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FOREWORD

I am pleased to put into the hands of readers Volume-9; Issue-2, March-April 2024 of **"International Journal of Environment, Agriculture and Biotechnology (IJEAB) (ISSN: 2456-1878)",** an international journal which publishes peer reviewed quality research papers on a wide variety of topics related to **Environment, Agriculture and Biotechnology**. Looking to the keen interest shown by the authors and readers, the editorial board has decided to release issue with DOI (Digital Object Identifier) from CrossRef also, now using DOI paper of the author is available to the many libraries. This will motivate authors for quick publication of their research papers. Even with these changes our objective remains the same, that is, to encourage young researchers and academicians to think innovatively and share their research findings with others for the betterment of mankind.

I thank all the authors of the research papers for contributing their scholarly articles. Despite many challenges, the entire editorial board has worked tirelessly and helped me to bring out this issue of the journal well in time. They all deserve my heartfelt thanks.

Finally, I hope the readers will make good use of this valuable research material and continue to contribute their research finding for publication in this journal. Constructive comments and suggestions from our readers are welcome for further improvement of the quality and usefulness of the journal.

With warm regards.

Editor-in-Chief
Date: May, 2024

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Response Evaluation of Chickpea Genotypes for Resisting *Helicoverpa armigera* (Hub) Throughout Growing Season & Correlating with Yield Parameter

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Abstract— *An investigation was worked out at the experimental Research Farm of the Department of Agricultural Entomology, VNMKV, Parbhani during Rabi, 2022 expecting resistive outcomes from responses of chickpea genotypes against gram pod borer (Helicoverpa armigera Hub.) for eggs and larval population including pod damage per cent from vegetative to maturity stage during growing meteorological weeks (MW). The correlation of the screening parameter with yield has also been calculated. The mean eggs, larval infestation and pod damage per cent of gram pod borer, H .armigera on genotypes under study is presented in the present investigation. It is found that on genotypes indicated significant differences regarding eggs, larval population and pod damage of H. armigera. The mean no. of eggs was reported on genotypes ICCL 86111 (0.24 eggs/plant). The least larval population was reported on genotypes ICCL 86111 (0.51 larvae/plant). The genotype ICCL 86111 had the least pod damage, 3.36 per cent and ICC 506 was (3.93 per cent) also fairly compatible genotype in this regard followed by BDNG 797 (4.26 per cent), ICC 92944 (5.46 per cent), ICCV 10 (5.88 per cent) and JG 62 (7.15 per cent) respectively. The chickpea yield showed significant correlation in negative manner with mean pod damage per cent (r = -0.774). Significantly Negative correlation was found with morning relative humidity with pest incidence in the genotypes viz., BDNG 797 and ICCL 86111.*

Keywords— ***Insect Ecology, Helicoverpa armigera, Eco-friendly, Host plant resistance, Pod damage, chickpea;***

I. INTRODUCTION

Chickpea (*Cicer arietinum* L.), being primary pulse crop of the Fabaceae family and often known as “Bengal gram” and locally as "chana." As a superior and less expensive source of protein than meat, chickpeas are locally referred to as "poor man's meat". Through biological nitrogen fixation of up to 140 kg of atmospheric nitrogen ha-1 year-1 ultimately preserves soil fertility. During the 2020–2021 growing season in Maharashtra, chickpeas were grown over an area of 16.94 lac/ha, yielding 13.97 lakh tonnes and 824 kg/ha of productivity. Production and productivity in the Marathwada region are 10.59 lakh/ha, 7.76 lakh tonnes and 707 kg/ha, respectively [1]. One of the most significant obstacles to the production of chickpeas worldwide is the chickpea pod borer (*H. armigera*). It infects more than 182 different crop types and is a serious pest. There is a large population of *H. armigera* throughout Asia, Africa, Australia, and Southern Europe [20]. *Helicoverpa armigera* affects chickpea from the early vegetative until podding stage, causing 60–80% of crop losses in Maharashtra state (India) [16]. The current research work was designed based on current hypotheses of *H. armigera* issue and the finding of pod borer (*H. armigera*) contesting genotypes of chickpea. Germplasm accessions have low to moderate levels of resistance to *H. armigera*. This has made it necessary to choose genotypes

with a higher capacity to withstand or regenerate from pod damage [13]. This promising method of pest management is even environmentally beneficial due to screening under natural environmental conditions and ultimately host plant resistance was evaluated. The researchers may use this data to adopt efficient and environmentally friendly chickpea genotypes and management strategies for this pest.

II. MATERIAL AND METHODS

Genotypes (ten) of chickpea with diverse growth habits were selected for their reaction to *H. armigera* under the screening studies. These genotypes were acquired from the Agriculture Research Station in Badnapur, VNMKV Parbhani (MH). viz., JG-11, JG 62, ICC 92944, BDNG-797, KAK 2 (Kabuli), ICCL-86111(R), ICC 506, ICCV 3137 (S), BDNGK-798 (Kabuli) and ICCV 10. The experiment was conducted at the experimental farm of the Department of Agricultural Entomology, VNMKV, Parbhani during Rabi 2021-22 under randomized block design (RBD) with three replications. The seeds were sown by dibbling during the 47 Meteorological week (25 Nov.). The standard dose of fertilizer was applied to all genotype tested for well growth. Other than that no other chemicals were sprayed. Three times hand weeding was done. Every plant of test genotype from each replication underwent weekly observations for the egg and larvae of *H. armigera* from 50 MW to 7 MW (Meteorological weeks) of *Rabi* 2021-22. The larvae of this insect pest rupture pods and penetrate into the pod and fed within, making seed unsuitable for human consumption [5]. To evaluate this effect of *H. armigera*, from pod initiation till plant harvest pod damage was monitored on every genotype. For estimation of pod damage per cent, from pod initiation until harvesting no. of healthy and injured pods per plant was recorded and the calculated per cent pod damage [6].

$$\text{Pod damage (\%)} = \frac{\text{No. of damaged pods}}{\text{Total no. of pods}} \times 100$$

Accordingly, from vegetative, flowering and pod formation stages of test genotypes of chickpea their mean no. of eggs and larvae including the percentage of pod damage throughout the season presented here.

Yield correlation:

Significant positive correlation examined between detrimental association of grain production/plant i.e., Yield and the no. of eggs and larva [8]. Thus correlation coefficient (r) was worked out between of *H. armigera* incidence with an average yield of chickpea. [10, 15, 19].

III. RESULTS AND DISCUSSION

Screening evaluation of ten chickpea genotypes in field for occurrence of eggs, larval population and pod damage per cent due to *H. armigera* under pesticide free condition was done during Rabi season of 2021-22.

3.1 Eggs of *H. armigera* on different chickpea genotypes during growing MW.

The information on eggs population of gram pod borer, *H. armigera* on different chickpea genotypes under study during 50 MW, 51 MW, 52 MW, 4 MW and 7 MW is presented in Table 1 and depicted in figure 1.

Table 1: Eggs population of H. armigera during 50, 51, 52, 4 and 7 Met. Week on chickpea genotypes.

Genotypes	No. of Eggs of *H.armigera*/plant				
	50 MW	51 MW	52 MW	4 MW	7 MW
JG-11	**0.59 (0.95)**	**0.30 (1.01)**	**0.15 (0.72)**	**0.77 (1.30)**	**0.83 (1.34)**
ICC 92944	**0.63 (1.10)**	**0.34 (1.10)**	**0.19 (0.80)**	**0.63 (1.26)**	**0.70 (1.29)**
KAK 2 (Kabuli)	**0.85 (1.17)**	**0.47 (1.20)**	**0.13 (0.68)**	**0.37 (1.16)**	**0.43 (1.20)**
ICC 506	**0.60 (0.92)**	**0.20 (1.10)**	**0.12 (0.65)**	**0.11 (1.05)**	**0.18 (1.09)**
BDNG-798 (Kabuli)	**1.00 (1.25)**	**0.57 (1.27)**	**0.26 (0.90)**	**0.79 (1.32)**	**0.86 (1.35)**
JG 62	**0.76 (1.04)**	**0.32 (1.04)**	**0.19 (0.78)**	**0.53 (1.22)**	**0.60 (1.25)**
BDNG-797	**0.83**	**0.40**	**0.16**	**0.19**	**0.26**

	(1.13)	**(1.13)**	**(0.75)**	**(1.08)**	**(1.12)**
ICCL-86111	**0.40 (0.86)**	**0.13 (0.92)**	**0.11 (0.57)**	**0.29 (1.13)**	**0.35 (1.16)**
ICC 3137	**1.10 (1.41)**	**0.70 (1.30)**	**0.27 (0.92)**	**1.23 (1.45)**	**1.30 (1.49)**
ICCV 10	**1.00 (1.25)**	**0.68 (1.29)**	**0.13 (0.67)**	**0.34 (1.15)**	**0.41 (1.18)**
SE(M)	**0.07**	**0.06**	**0.059**	**0.07**	**0.07**
CD @ 5%	**0.22**	**0.18**	**0.173**	**0.21**	**0.21**
CV %	**10.55**	**8.86**	**9.361**	**10.12**	**9.86**

Parenthesis figures of eggs population are √x+0.5

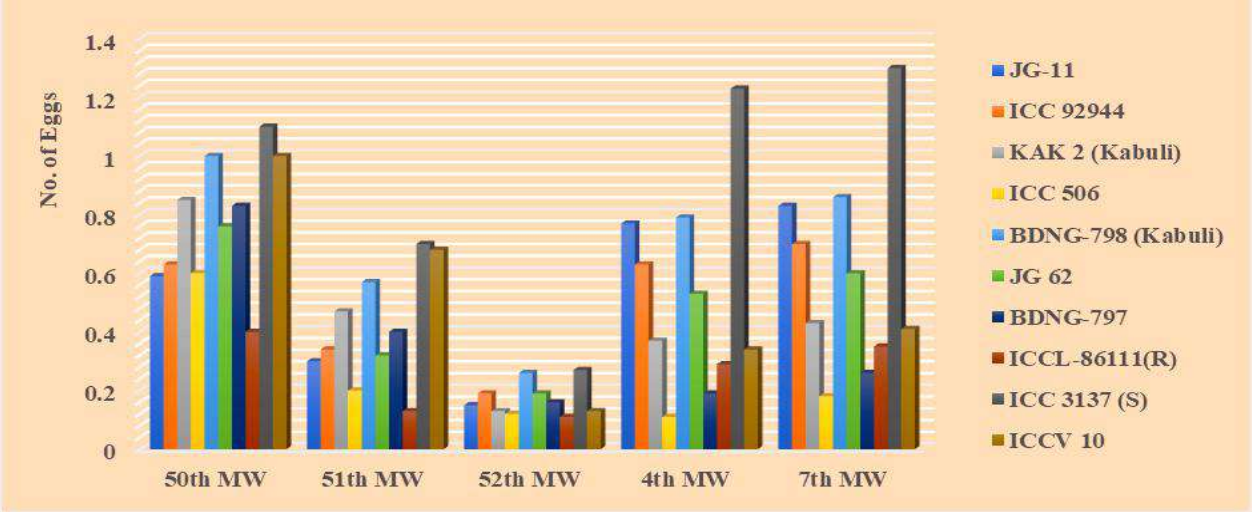

Fig.1: Eggs population of H. armigera during 50, 51, 52, 4 and 7 Met. week on chickpea genotypes.

The data revealed significant differences (Table 1; figure 1) among the genotypes regarding egg population of *H. armigera*. The eggs population per plant appeared in different parallel ranges of 0.40 to 1.10 eggs/plant, 0.13 to 0.70 eggs/plant, 0.11 to 0.27 eggs/plant, 0.11 to 1.23 eggs/plant and 0.18 to 1.30 eggs/plant respectively during five MW. This result coincides with reference research where 9 chickpea genotypes against *H. armigera* evaluated and the no. of eggs of *H. armigera* on different genotypes varied from 2.30 to 15.74 eggs and lowest oviposition was recorded on Genotype 5282. [4] ICCL 86111 performed well with the least no. of eggs reported during 50, 51 and 52 MW (0.40 eggs/plant, 0.13 eggs/plant and 0.11 eggs/plant respectively. After that during the 4 and 7 MW ICC 506 showed a least population of eggs viz., 0.11 eggs/plant and 0.18 eggs/plant respectively. ICC 3137 showed the highest level of eggs population appearance 11.10 eggs/plant, 0.70 eggs/plant, 0.27 eggs/plant, 1.23 eggs/plant and 1.30 eggs/plant during five weeks of observation respectively. During 50 MW JG 11 (0.59 eggs/plant) followed after ICC 506 (0.60 eggs/plant) and JG 62 (0.76 eggs/plant) performed at par with ICCL 86111. During 51 MW ICC 506 (0.20 eggs/plant) followed by JG 11 (0.30 eggs/plant), JG 62 (0.32 eggs/plant) and ICC 92944 (0.34 eggs/plant) were found statistically significant and at par with well-performing genotypes. 52 MW showed that ICC 506, KAK 2, ICCV 10 and JG 11 i.e., 0.12, 0.13, 0.13 and 0.15 eggs/ plant significantly at par with ICCL 86111. During the 4 MW BDNG 797 (0.19 eggs/plant) followed by ICCL 86111 (0.29 eggs/plant), ICCV 10 (0.34 eggs/plant) and KAK 2 (0.37 eggs/plant) performed well after ICCL 86111. [23] After evaluating 31 genotypes in the field for 100 days after germination showed the quantity of eggs/plants ranged highest in ICC 3137 (5.1 eggs/plants) among others. [22] This had also been discovered that the genotypes ICC-3137, K-850, and ICC-1403 were more defenseless and have privileged more eggs laying Ultimately during 7 MW BDNG 797 (0.26 eggs/plant) followed by ICCL 86111 (0.35 eggs/plant), ICCV 10 (0.41 eggs/plant), KAK 2 (0.43 eggs/plant) and JG 62 (0.60 eggs/plant) performed better for further recommendation.

3.2 Larval population of *H. armigera* on different chickpea genotypes throughout growing MW.

Larval population of borer catterpiller, on different chickpea genotypes were undertook for study throughout plant growing ten weeks (50 MW to 7 MW) showed here (Table 2 and depicted in figure 2). The figures revealed significant differences among the genotypes regarding larval population of *H. armigera*. The no. of larvae per plant of *H. armigera* during ten experimental MW appeared in different parallel ranges of 0.66 to 1.11 larvae / plant, 0.26 to 0.80 larvae / plant, 0.18 to 0.91 larvae / plant, 0.26 to 0.98 larvae / plant, 0.58 to 1.50 larvae / plant, 0.41 to 1.54 larvae per plant, 0.65 to 1.77 larvae / plant, 0.83 to 1.63 larvae / plant, 0.80 to 2.03 larvae / plant and 0.99 to 1.99 larvae / plant respectively on the chickpea genotypes under study. The result coincides with BG-372, HC-1, SAKI-9516, Vijay and Avrodhi were relatively less susceptible as these harbored lower larval population (1.07 to 1.32 larvae/plant) [7]. The lowest no. of larvae during 50, 51, 52, 2, 3, 6 MW recorded on genotypes ICCL 86111 (0.66 larvae/plant, 0.26 larvae/plant, 0.18 larvae/plant, 0.58 larvae/plant, 0.41 larvae/plant and 0.80 larvae/ plant respectively). ICC 506 during 4, 5 and 7 MW showed 0.65 larvae/plant, 0.83 larvae/plant and 0.80 larvae/ plant respectively. But during 1 MW least larval population was reported on genotypes ICC 92944 (0.26 larvae/plant) [12] Equivalent results were obtain according resistant genotype C 235 showed lowest larvae population (0.5/10 plants) and utmost no. of larvae (3.0/10 plants) and susceptible genotype H 82-2.

Table 2. Larval population of H. armigera on chickpea genotypes during growing meteorological weeks.

Genotype	No. of larval population of *H.armigera*/plant during MW									
	50 MW	**51 MW**	**52 MW**	**1 MW**	**2 MW**	**3 MW**	**4 MW**	**5 MW**	**6 MW**	**7 MW**
JG-11	0.76 (1.01)	0.77 (1.32)	0.42 (1.15)	0.90 (1.23)	1.20 (1.59)	1.24 (1.63)	1.43 (1.55)	1.57 (1.59)	1.63 (1.68)	1.63 (1.70)
ICC 92944	0.88 (1.12)	0.38 (1.11)	0.44 (1.16)	0.26 (1.00)	0.84 (1.41)	0.88 (1.43)	0.96 (1.39)	1.10 (1.43)	1.16 (1.45)	1.17 (1.45)
KAK 2 (Kabuli)	0.92 (1.15)	0.49 (1.20)	0.61 (1.28)	0.70 (1.30)	0.87 (1.40)	0.91 (1.47)	0.90 (1.36)	1.03 (1.40)	1.10 (1.43)	1.11 (1.43)
ICC 506	0.86 (0.98)	0.31 (1.06)	0.38 (1.12)	0.57 (1.11)	0.64 (1.17)	0.68 (1.21)	0.65 (1.25)	0.83 (1.32)	1.00 (1.38)	0.99 (1.38)
BDNG-798 (Kabuli)	1.05 (1.29)	0.76 (1.30)	0.67 (1.32)	0.83 (1.23)	1.28 (1.63)	1.32 (1.67)	1.53 (1.59)	1.67 (1.63)	1.73 (1.65)	1.74 (1.65)
JG 62	0.89 (1.09)	0.50 (1.28)	0.56 (1.25)	0.77 (1.34)	0.95 (1.45)	0.99 (1.49)	1.07 (1.42)	1.20 (1.46)	1.30 (1.50)	1.31 (1.50)
BDNG-797	0.8 (1.19)	0.43 (1.15)	0.47 (1.18)	0.47 (1.13)	0.64 (1.17)	0.59 (1.21)	0.86 (1.33)	0.86 (1.33)	1.04 (1.40)	1.02 (1.38)
ICCL-86111	0.66 (0.92)	0.26 (0.98)	0.18 (0.93)	0.52 (1.10)	0.58 (1.10)	0.41 (1.10)	0.75 (1.31)	0.97 (1.38)	0.80 (1.33)	1.04 (1.39)
ICC 3137	1.11 (1.28)	0.80 (1.45)	0.91 (1.46)	0.98 (1.42)	1.50 (1.72)	1.54 (1.79)	1.77 (1.66)	1.63 (1.71)	2.03 (1.74)	1.99 (1.73)
ICCV 10	1.05 (1.20)	0.68 (1.27)	0.23 (0.98)	0.70 (1.27)	0.91 (1.41)	0.95 (1.49)	0.87 (1.35)	1.01 (1.39)	1.07 (1.42)	1.08 (1.42)
SE(M)	0.06	0.04	0.08	0.15	0.13	0.17	0.06	0.076	0.05	0.07
CD @ 5%	0.18	0.12	0.26	0.45	0.39	0.38	0.18	0.227	0.15	0.22
CV %	10.44	9.92	8.864	10.62	10.15	10.19	9.80	8.947	8.69	8.57

Parenthesis figures of eggs population are $\sqrt{x+0.5}$

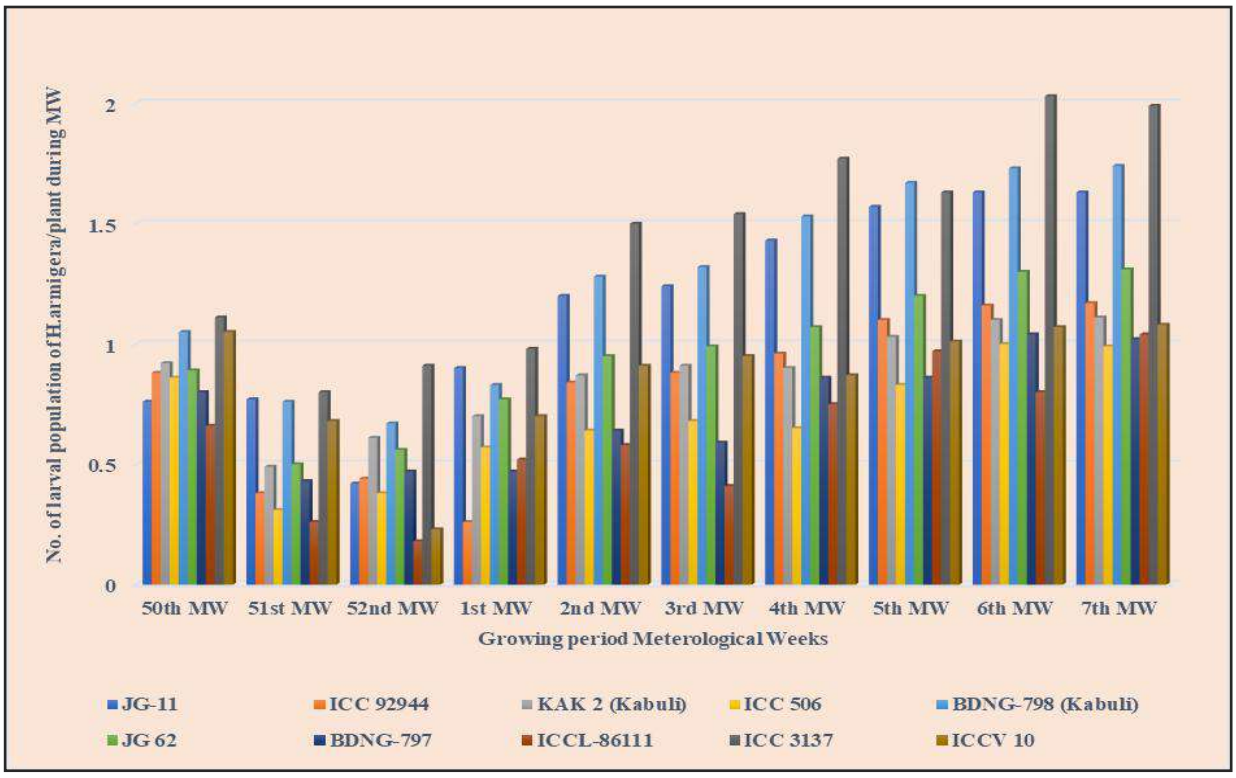

Fig. 2: Larval population of H. armigera on chickpea genotypes during growing meteorological weeks.

As discussed earlier least no. of larvae was observed on ICCL 86111 but after that ICC 506 (0.86, 0.31, 0.38, 0.57, 0.64, 0.68, 0.65, 0.83, 1.00 and 0.99 larvae / plant) and BDNG 797 (0.8, 0.43, 0.47, 0.47, 0.64, 0.59, 0.86, 0.86, 1.04, 1.02 larvae / plant) during every screening week proved themselves better fit after ICCL 86111. Kabuli chickpea types *viz.*, KAK 2 (0.92, 0.49, 0.61, 0.70, 0.87, 0.91, 0.90, 1.03, 1.10 and 1.11 larvae / plant) and BDNG 798 (1.05, 0.76, 0.67, 0.83, 1.28 1.32, 1.53, 1.67, 1.73 and 1.74 larvae / plant) also showed less than ICC 3137 but maximum positive response to larval feeding preference than other genotypes during screening weeks. Other genotypes *viz.*, JG-11, ICC 92944, JG 62 and ICCV 10 showed moderate level of resistance and medium level of infestation of larvae throughout MW. Accordingly highest population of *H. armigera* larvae throughout experimental growing season in respective ten MW was recorded on ICC-3137 (1.11 larvae/plant, 0.80 larvae/plant, 0.91 larvae/plant, 0.98 larvae/plant, 1.50 larvae/plant, 1.54 larvae/plant, 1.77 larvae/plant, 1.63 larvae/plant, 2.03 larvae/plant and 1.99 larvae/plant respectively. After assessing *H. armigera* resistance responses from 11 different chickpea cultivars Chaffe (14.32) and ICCV 10 had the lowest larvae whereas Phule G 5 (26.33), PG 8111 (24.90), GNG 465 (23.61) and BG 391 had the greatest larval incidence (23.31) [3].

3.4 Mean Eggs, Larvae, Pod damage, and effect on yield due to *H.armigera* during Met. Week on chickpea

The data pertaining mean eggs, larval infestation and per cent pod damage of gram pod borer, H .armigera on genotypes under study is presented in (Table 3 and depicted in fig. 3). It is found that on genotypes indicated significant differences regarding eggs, larvae and pod damage due to *H. armigera*. The mean results were observed in the range of 0.24 to 1.06 eggs per plant, 0.51 to 1.46 larvae/plant and 3.36 to 9.85 per cent respectively on the different genotypes. The lowest degree of mean infestation because of *H. armigera* was observed on genotype ICCL 86111 (0.24 eggs/plant, 0.51 larvae/plant and 3.36 per cent) which was at par with ICC 506 (0.27 eggs/plant, 0.64 larvae/plant and 3.93 per cent), BDNG 797 (0.35 eggs/plant, 0.67 larvae/plant and 4.26 per cent) and KAK 2 (0.43 eggs/plant, 0.80 larvae/plant and 7.25 per cent) respectively. Followed by ICCV 10 (0.49 eggs/plant and 0.83 larvae/plant), JG 11 (0.52 eggs/plant and 1.06 larvae/plant), ICC 92944 (0.52 eggs/plant and 0.82 larvae/plant). BDNG 798 (8.88 per cent) indicating moderate pod damage due to pod borer at par damage with after ICC 3137. Highest mean no. of eggs per plant, larvae per plant of *H. armigera* and per cent pod damage due to *H. armigera* was observed on genotype (1.06 eggs/plant, 1.46 larvae /plant and 9.85 per cent). Results concurs intervention with other reports [14] that the pod damage varies from 9.43 to 24.80 per cent. It is seen that variety Vijay had the least amount of pod damage (19.73 and 23.33%), followed by RSG 888 (20.46 and 27.67) but

variety Samrat had the most pod damage (30.40 and 34.33%) [9]. Also genotypes ICC 506, ICCV 10, ICCL 86102, and ICCV 95992 were shown to have low pod damage ratings of 3 from one to nine scale [2]. [18] Most promising strain, BRC-4, and the least vulnerable strain, BRC-1, both had pod damage levels of 9.38 and 21.49 per cent with grain yields of 0.333 and 0.137 kg/plot, respectively.

Table 2. Mean Eggs, Larvae, pod damage and yield due to H.armigera during Met. Week on chickpea

Genotype	Mean eggs-larvae and per cent pod damage			
	Eggs of *H.armigera*/plant	**Larva of *H.armigera*/plant**	**Pod Damage (%)**	**Yield (Kg/Ha)**
JG-11	0.52 (1.23)	1.06 (1.42)	7.72 (16.03)	1376
ICC 92944	0.52 (1.23)	0.82 (1.34)	5.46 (13.48)	1567
KAK 2 (Kabuli)	0.43 (1.19)	0.80 (1.33)	7.25 (15.55)	970
ICC 506	0.27 (1.12)	0.64 (1.28)	3.93 (11.43)	1654
BDNG-798 (Kabuli)	0.68 (1.29)	1.18 (1.47)	8.88 (17.24)	1421
JG 62	0.47 (1.21)	0.88 (1.36)	7.15 (15.28)	1341
BDNG-797	0.35 (1.16)	0.67 (1.29)	4.26 (11.88)	1477
ICCL-86111	0.24 (1.11)	0.51 (1.22)	3.36 (10.50)	1705
ICC 3137	1.06 (1.42)	1.46 (1.56)	9.85 (18.21)	876
ICCV 10	0.49 (1.21)	0.83 (1.35)	5.88 (13.92)	1620
SE(M)	0.03	0.03	0.56	19.40
CD @ 5%	0.08	0.10	1.61	58.10
CV %	8.46	8.89	9.82	17.80

Parenthesis figures of eggs population are $\sqrt{x+0.5}$

Figures of percentage in parenthesis are angular transformed values.

3.5 Average grain yield of test chickpea genotypes

The figures on average grain yield of test chickpea genotypes are specified in Table 3 and fig 4. It ranged from 870 to 1705 kg/ha. The uppermost grain yield was recorded by genotypes ICCL 86111 (1705 kg/ha) which was at par with the genotypes ICC 506 (1654 kg/ha) followed by ICCV 10 (1620 kg/ha), ICC 92944 (1567 kg/ha), BDNG 797 (1477 kg/ha), BDNG 798 (1421 kg/ha), JG 11 (1376 kg/ha) and JG 62 (1341 kg/ha) respectively. The lowest grain yield among the genotypes tested were recorded by KAK 2 (970 kg / ha) and ICC 3137 (876 kg/ha). It has been also found that the grain production per plot ranged from 23.33 to 192.00 gm /plant with larvae ranging from 1 to 50 [17]. [11] While comparing genotype ICC 506 to Annegeri, ICC 506 showed (2.08) and achieved yield of 797 kg/ha, whereas Annigeri with higher damage rating (8.33) and achieved 620 kg/ha. Chaffa, the cultivar that experienced the least pod damage (9.55%), was the most resilient.

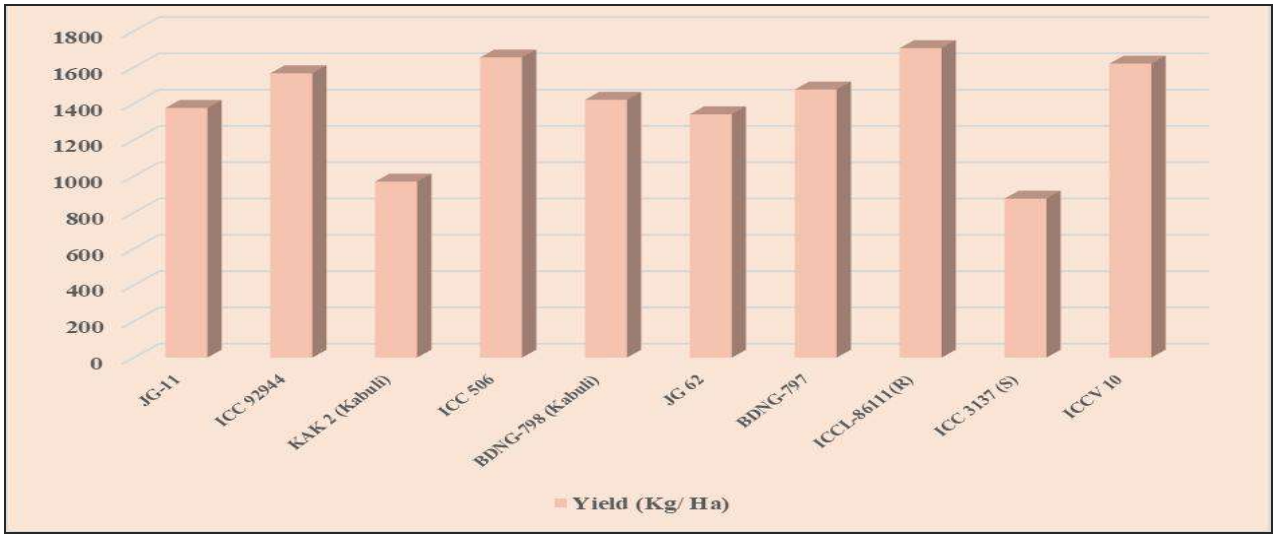

Fig. 2: Average grain yield (Kg/ha) of chickpea genotypes

3.6 Correlation of yield with *H. armigera* damage parameters.

The chickpea yield showed significantly negative correlation (Table 4) with mean per cent pod damage (r = -0.774) and Mean Eggs of H.armigera, Mean Larvae of H.armigera (r = -0.690, -0.688, respectively) indicating correlation of higher pod damage with lower the yield of chickpea.

Table 4. Correlation (r) of yield with screening attributes against H. armigera

Sr. no	**Screening attributes against *H. armiger***	**Yield**
1	Mean Eggs Population of *H.armigera*	-0.690
2	Mean Larval Population of *H.armigera*	-0.688
3	Mean Pod damage % due to *H.armigera*	-0.774**

**Significance Level at 0.01 % (0.765)

The present findings are supported by earlier research reports [10] yield obtained in ICCV 10, ICC 506 and ICCL 86111 were 1641, 1887 and 2120 kg/ha., respectively. [1] report depicts that ICCV 10 exhibited high yield potential and ICCV 09118 also showed a grain yield potential of >15.2 q/ha. The genotypes ICCV 07104 and ICCV 10 showed a yield potential of >15.0 q/ha compared to 5 q/ha in ICC 3137. Decrease in grain yield was lowermost in resistant check [21].

IV. CONCLUSION

The present findings are supported by earlier research reports [10] yield obtained in ICCV 10, ICC 506 and ICCL 86111 were 1641, 1887 and 2120 kg/ha., respectively. Anonymous (2010) reported that ICCV 10 exhibited high yield potential and ICCV 09118 also showed a grain yield potential of >15.2 q/ha. The genotypes ICCV 07104 and ICCV 10 showed a yield potential of >15.0 q/ha compared to 5 q/ha in ICC 3137. Decrease in grain yield was lowermost in resistant check [21].

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Intake of immunity boosters by Covid -19 patients of Bikaner city

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Abstract— *Corona virus disease 2019 (COVID-19) is an acute and contagious disease characterized by pneumonia and acute respiratory distress syndrome, which is caused by a novel corona virus (2019-nCoV). Three hundred post Covid-19 adult (19-39 years) patients were selected for present study comprising 150 males and 150 females. Subjects were selected after procuring the list of patients from Prince Bijay Singh Memorial and Shri Ram Hospital, Bikaner (Rajasthan) by purposive/ convenient random sampling on the basis of their willingness to cooperate during the study. The study had been carried out from January to June 2021. Majority of subjects consumed immunity boosters such as chyawanprash (74.0%), homemade decoction (84.3%), golden milk (78.7%) and citrus fruits (100%). Majority of subjects (74.0%) were aware about the meaning of immunity boosters. Majority (80.0%) of subjects were doing yoga daily. Almost equal number of subjects irrespective of gender drunk warm water whole day, it may be due to the reason that guidelines given by WHO has strongly recommended to drink warm water daily. In present investigation data unfurls that all patients (100%) consuming turmeric as immunity booster spices followed by 64.3 % cinnamon, 18.7% garlic and 5.3% coriander. Majority of subject's (74.0%) consumed chyawanprash, while 26.0 percent did not consume. During pandemic the consumption of turmeric milk found high as it is good antibiotic; also have positive effect on immunity.*

Keywords— ***Immunity boosters, immunity, , pandemic, antibiotic.***

I. INTRODUCTION

The Coronavirus disease 2019 (COVID-19) is an acute and contagious disease characterized by pneumonia and acute respiratory distress syndrome (ARDS). The pandemic caused by a novel coronavirus (2019-nCoV) recently reclassified and named severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) which belongs to the family of *Coronaviridae*. The first coronavirus that caused severe disease was severe acute respiratory syndrome (SARS), which originated in Foshan, China, and resulted in the 2002, SARS-CoV pandemic. The second was the coronavirus-caused Middle East respiratory syndrome (MERS), which originated from the Arabian Peninsula in 2012. SARS-CoV-2 is the third coronavirus that caused severe disease in humans to spread globally in the past 2 decades (Wiersinga *et al.*, 2020). Coronaviruses are large, enveloped, single-stranded RNA viruses found in humans and other mammals, such as dogs, cats, chicken, cattle, pigs, and birds. The first human coronavirus was identified in 1960 and to date seven human coronaviruses have been recognized. CoVs are divided into four genera: alpha, beta, gamma and delta-CoV. CoVs currently known to cause diseases in humans belong to the alpha or the beta-CoV. The average incubation period of SARS-CoV-2 is 5–6 days, but it can be up to 14 days. The routes from human to human of SARS-CoV-2 transmission can be direct, droplet inhalation, contact, through saliva, and via fecal–oral routes (Khan *et al*., 2020). Human-to-human transmission of SARS-CoV-2 occurs mainly between family members, including relatives and friends who intimately contacted with patients or incubation carriers (Guo *et al*., 2020).

The clinical outcomes of Covid-19 can vary from asymptomatic to a mild to severe state. Common symptoms of Covid-19 include headache, breathlessness, fever, cough, fatigue, dyspnea, diarrhea, and even conjunctivitis, occasionally leading to severe SARS-like viral pneumonia, acute respiratory distress syndrome (ARDS), multi-organ dysfunction, and even death (Balasubramanian and Ramalingam, 2020). Asymptomatic patients have infection (SARS-CoV-2) but never show any signs and symptoms while pre asymptomatic patients have no symptoms but when detected they were positive for SARS-CoV-2 and later develop symptoms. Older people and people with pre-existing medical conditions such as diabetes and heart disease are more vulnerable to the virus to become seriously ill (Syangatan *et al*., 2020).

Mrityunjaya *et al.* (2020) suggested that many nutritional supplements from various spices, herbs, fruits, roots, and vegetables can reduce the risk or severity of a wide range of viral infections by boosting the immune response, particularly among people with inadequate dietary sources and also by their anti-inflammatory, free radical scavenging, and viricidal functions. These nutrients can be repurposed in mitigating the pathological effects induced by the SARS-CoV-2 infection. Therefore, the use of natural compounds may provide alternative prophylactic and therapeutic support along with the therapy for Covid-19.

The immune system is indeed complex and is to a great extent impacted by the environment around us. Strengthening the immune system is the best protection against infectious diseases and a secret to good health. Counseling regarding immunity boosters is mandatory to make people aware and protect themselves from attack of virus by following certain habits like eating nutritious food, proper hygiene and sanitation, regular exercise, maintenance of emotional and mental health and usage of spices as well as herbal decoctions. However, a healthy lifestyle can help naturally build up your immune system to make your defense as strong as possible and prevent people to Covid-19.

II. REVIEW OF LITERATURE

Cinatl *et al.* (2003)**;** Chen *et al.* (2004) and Brush *et al. (*2006) reported that other anti-viral components like glycyrrhizin from liquorice have been known to show some activity against Corona virus and SARS, revealing the significance of glycyrrhizin to be used as trial drug in Covid-19 affected patients, in order to develop it as a potent cure drug. Similarly Carrasco *et al. (*2009); Kim and Lee (2009) and Bui *et al.* (2019) reported that the decoction of ginger, cloves and black peeper have been recommended to the healthy as well as Covid-19 infected person as it provides support in the humoral and cell mediated responses and also lowers the air way hyper responsiveness and nasal congestions. Apart from this, muti-vitamins intake within human body can wash away the sickness forces through recruiting the immune soldiers Combs *et al.* 2016. An *in-vitro* study by Lin *et al.* (2017) depicted the role of resveratrol from grape seeds against Corona viruses. Nevertheless, other plants like oregano, garlic, ginger, lemon, broccoli, mint, *tulsi*, fennel, thyme, cinnamon, star anise etc. should be tested in the different forms so as to form an effective drug to fight against Covid-19. It was further affirmed that Vitamin D was potent in reducing the hazards associated with viral pandemic.

According to Majumdar *et al*. (2020) **"**The global healthcare system is facing many challenges during the pandemic outbreak of Covid-19. India has a treasury of some indigenous medicines like Ayurveda, Unani and Siddha. Ayurveda is a classical medicinal system initiated in India from 2000 years ago. Ayurvedic can stimulate the immune-modulators within human body and enhance the immunity system. Ayurveda and yoga may support the patients of Covid-19 with improvement of quality of standard care". Recently in India, it was suggested by the Ministry of AYUSH, to drink *Kadha* as a booster of immunity and lowering the tenderness caused during Covid-19 catastrophe. A *Kadha* is an extract prepared from less juicy or dry ingredients like spices and herbs. The Ministry of AYUSH with its conventional acquaintance has an extensive custom of maintenance of nation's health and its participation has augmented manifolds in this Covid-19 pandemic crisis (AYUSH Advisory, 2020). All ayurvedic healthcare professional generally recommend classical ayurveda medicine, however AYUSH-64 a novel formulation prepared by CCRAS provides resistance against malaria and other fevers.

According to Ding *et al.* (2018) and Adem *et al.* (2020) Polyphenols are known to increase the immune to cells to foreign infestations and in response permits cellular accumulation of different types of polyphenols through varied receptors. This subsequently triggers signaling pathways and initiate immune responses. The natural polyphenols were identified as potent Covid-19 protease inhibitors. Similarly Lewis *et al.*, (2019) and Petric (2020) reported that diet comprising of multi-vitamins wield immune modulatory possessions on numerous immune cells such as monocytes, neutrophils, lymphocytes, NK cells and dendritic cells that bump up the immunity against pathogens**.** In the same way Sarfraz *et al.*, (2020) stated that dietary minerals are essential immunity mediators should be incorporated into the diet i.e. different fruits, spices and vegetables are abundant with immuno-

stimulators that in turn fortify the innate as well as adaptive immune responses against viral elements**.**

Pal *(*2020) also revealed that zinc one of the anti-inflammatory and anti-oxidant micro-nutrient found in food with well-established role in immunity is currently being used in some clinical trials against Covid-19**.** Likewise Arshad *et al.* (2020) stated that Covid-19 is affected people with low immunity response. Plant-based foods increased the intestinal beneficial bacteria which are helpful and make up of 85 per cent of the immune system. By the use of plenty of water, minerals like magnesium and zinc, micronutrients, herbs, food rich in vitamins C, D & E and better life style can promote the health and can overcome this infection. In the same way Jayawardena *et al.* (2020) reported that scientific expedition of escalating immune system through appropriate sleep, judicious exercise, stress-free environment, proper nutritive foods, water intake and consumption of fresh and healthy fruits and vegetables would anticipate the citizenry to cope with corona virus battle via naturally vaccinating their systems. While Yasmin *et al.* (2020) revealed that high dose of Vitamin D reduced the hazard of several chronic ailments such as cardiovascular diseases, diabetes mellitus, hypertension, cancer and respiratory tract infection. Similarly Joachimiak (2021) reported the current estimates of 40 per cent to 70 per cent individuals worldwide will become infected over the course of this pandemic in the absence of strong mitigation efforts. Zinc should be included as part of preventative supplementation for Covid-19 and in general for support of immune health. Zinc supplementation should also be considered in the context of zinc deficiency acquired during a viral infection and host immune response. Healthy individuals with a robust immune system have clearly a better starting point for the difficult Covid-19 viral infection with expected positive effects on clinical outcomes such as shortening the duration of even just the sub-severe cases.

III. MATERIALS AND METHOD:

Locale of the study

The list of patients after seeking prior permission and having discussion with the respective hospital authorities was procured from Prince Bijay Singh Memorial and Shri Ram Hospital, Bikaner (Rajasthan). After that researcher visited respondents residential site and most of the patients belongs to the east & West Bikaner.

Selection of sample

Researcher visited hospital for subject selection after obtaining authorized consent from the respective hospital authorities. The post Covid-19 adult patients were selected for the present investigation. Three hundred both male and female were selected by purposive/ convenient random sampling on the basis of their willingness to cooperate during the study.

Development of interview schedule for data collection

A structured interview schedule with sufficient number of items prepared short, crisp, scientifically structured, validated, easy-to-use and applicable for each group was developed to collect the data from the subjects. The schedule approved by the experts before implementing on the subjects. The pretested interview schedule consists following section:

General information of the subjects

This section included gathering of general information about the subjects regarding their name, age, gender, category, religion, educational qualification, type of activity, family type etc.

Age: The chronological age of the respondents at the time of data collection was recorded.

Gender: It refers to the range of characteristics pertaining to and differentiating between male and female.

Religion: The categories framed as Hindu, Muslim, Sikh, Christian and Jain.

Category: Caste referred to the class or distinct hereditary order of society. Information regarding caste of the respondents recorded fewer than four categories: General, Other Backward class, Schedule Caste and Schedule Tribes.

Educational Qualification: The term education was operational zed as the level of school education perused by the respondents. Level of education assessed in terms of Literate, Primary, Middle, High school, Graduate (Bachelor degree) and above (Post graduate and Doctorate).

Type of activity: The quality or state of being active was assessed in terms of sedentary, moderate and heavy.

Family Type: Nuclear family consists husband wife and their children and any other members of unit which are living with them. Joint family often includes multiple generations in the family. Three to four generations stay together under a single roof Rayangoudar (2009).

RESULTS AND DISCUSSION :

Distribution of subjects according to their demographic profile

General information

General information of three hundred Covid-19 patients (150 males and 150 females) aged 19-39 years selected randomly from Bikaner district of Rajasthan

collected thorough a pre-tested interview schedule which is narrated below.

Category

The table 1 illustrates that majority of the subjects (59.0%) belonged to general category followed by 24.7 per cent from OBC category, 9.7 per cent from schedule caste and 6.7 per cent of them belonged to Schedule tribes. Gender-wise data shows that the percentage of females (62.0%) in general category recorded higher as compared to males (56.0%) and equal percentage noted in case of schedule tribes (6.7%).

Religion

Information regarding religion of the subjects explicated that majority of them Hindu (94.3%) followed by Jain, Muslim, Sikh and Christian (2.3%), (2.0%), (1.0%) and (0.3%) respectively (Table 1).

Table 1: Distribution of subjects according to their demographic profile

Parameters	Males n=150(50)	Females n=150 (50)	Total N=300(100)
Category			
General	84 (56.0)	93 (62.0)	177 (59.0)
Other Backward Class	42 (28.0)	32 (21.3)	74 (24.7)
Schedule Caste	14 (9.3)	15(10.0)	29 (9.7)
Schedule Tribes	10 (6.7)	10(6.7)	20 (6.7)
Religion			
Hindu	140 (93.3)	143 (95.3)	283 (94.3)
Muslim	2 (1.3)	4 (2.7)	6 (2.0)
Sikh	3 (2.0)	0 (0)	3 (1.0)
Christian	1 (0.7)	0 (0)	1 (0.3)
Jain	4 (2.7)	3 (2.0)	7 (2.3)
Educational qualification			
Primary	0 (0)	0 (0)	0 (0)
Middle	2 (1.3)	3 (2.0)	5 (1.7)
High School	23 (15.3)	17 (11.3)	40 (13.3)
Graduate	84 (54.8)	100 (66.7)	182 (60.7)
Above	43(28.7)	30 (20.0)	73(24.3)
Type of activity			
Sedentary	150 (100)	150 (100)	300(100)
Moderate	0 (0)	0 (0)	0(0)
Heavy	0 (0)	0 (0)	0(0)
Family type			
Nuclear	110 (73.3)	118 (78.7)	228 (76.0)
Joint	40 (26.7)	32 (21.3)	72(24.0)

Note- Figures in parenthesis indicates percentage of subjects

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Educational Qualification

Subjects inquired about their academic qualifications. Table 1 unfurls that majority of the subjects (60.7%) finished graduation followed by post-graduation and above (24.3%), studied up to high school (13.3%) and only 1.7 percent attended middle school. None of the subject found educated up to primary level. Higher percentage of females (66.7%) found educated as compared to males (54.8%) up to graduation level. Similar trend observed for middle school while opposite result found at high school and above graduation.

Type of activity

Data unfolds that all the subjects found to be engaged in sedentary activities irrespective of gender. None of the subject engaged in moderate and heavy activities (Table 1).

Family Type

Table 1 discloses the data of type of family in terms of joint and nuclear. It can be seen that most of the subjects (76.0%) belonged to nuclear family followed by joint family (40.0%).

Information related to Immunity booster

Immunity is the main mechanism of host defense against infectious agents and Covid- 19 badly affects the individuals with low immunity. Vitamins, minerals, antioxidants, probiotics, and functional foods are some immune boosters which contribute to strong immunity. Information regarding Immunity is depicted in table 2 and portrayed beneath.

Meaning of Immunity booster

Immunity boosters are the supplements which can increase our immunity. Table (2) reflects that majority of subjects (74.0%) was aware about the meaning of immunity boosters. None of the subjects found unaware about the immunity booster, reason may be due to wide awareness campaign done by government and other agencies on print and electronic media. It also plays a significant role in pandemic for faster recovery and reduces the severity. Irrespective of gender almost equal number of subjects had proper knowledge of immunity booster.

Meditation yogasan and pranayama

Stress is one of the leading killers of immunity. Meditation yog asana and pranayama lead to a positive state of mind that promotes better health and immunity (Bushell *et al.*, 2020). The data in table (2) unfurls that majority (80.0%) of subjects were doing yoga daily. Only 20.7 percent males and 18.3 percent females did not doing mediation and pranayama.

Intake of warm water

Drinking warm water daily may keep our digestive system intact, help in improving blood circulation and reduce stress level. Ninety one percent of the subjects in the present study drunk warm water throughout the day. Almost equal number of subjects irrespective of gender drunk warm water whole day, it may be due to the reason that guidelines given by WHO has strongly recommended to drink warm water daily [Table 2].

Spices

Herbs and species are well known to boost immunity. It was observed that nations with lower consumption of species per capita showed greater number of Covid-19 cases per million populations. In present investigation data unfurls that all patients (100%) consuming turmeric as immunity booster spices followed by 64.3 % cinnamon, 18.7% garlic and 5.3% coriander. While 43.0 percent consumed other spices such as cumin, ginger, *munkka, kalimirch* and *tulsi.* None of the female subjects consumed coriander as a immunity booster [Table 2]

Ayush Ministry *kadha*

Ayush ministry has suggested measures which may help the public to remain healthy in pandemic times. The ministry recommended decoction made out using from *tulsi*, *dalchini*, *kalimirch*, *shunthi*, *Munkka*, jaggery and fresh lemon juice to be consumed once or twice a day which have a positive effect on the respiratory system. Data reflects that majority of subjects (88.3%) consumed ayush ministry *kadha.* Irrespective of gender almost equal number of subjects consumed kadha. It has been suggested that decoction kills the virus in respiratory itself and protects individuals to suffer with severity [Table 2].

Intake of *Chyawanprash*

Chyawanprash is a household remedy in north india. It is formulated by processing medicinal herbs and their extracts, including the prime ingredient *amla* (Indian gooseberry), which is the world's richest source of vitamin C subsequent mixture with honey and addition of aromatic herb powders (clove, cardamom, and cinnamon). It has positive effect on immune system. Majority of subject's (74.0%) consumed c*hyawanprash*, while 26.0 percent did not consume. Higher percentage of males (78.7%) than females (69.3%) used *chyawanprash* in Covid-19 period [Table 2].

Ayush Ministry guidelines

Ministry of Ayush recommended guideline to stop spread of Covid-19 virus. The ministry has given some measures of boosting immunity which included ayurvedic spices to be added in their diet to enhance immunity, using oil therapy for mouth and nose and steam inhalation. Most of the subjects (87.7%) following Ayush ministry guideline only 12.7 percent did not aware about these measures which are taken by government. More male (97.3%) subjects followed Ayush ministry guidelines than females (77.3%) [Table 2].

Homemade *kadha*

Homemade *kadha* is easily available source to improve immunity. Consuming herbal drinks and taking proper rest have helped Covid -19 patients survive from the deadly viral disease. Results unfurl that in present study most of the subjects (84.3%) consumed *kadha* prepared at home. Subjects convinced with the fact that immunity-boosting *kadha* is a life saviour and has helped them get through Covid -19 and also decrease the risk of severity [Table 2].

Golden milk or turmeric milk

India is a leading producer and exporter of turmeric in the world. Ninety percent of the total produce is consumed internally and only a small portion exported. During pandemic the consumption of turmeric milk found high as it is good antibiotic; also have positive effect on immunity. The results of present investigation demonstrated that 78.7 percent of subjects consumed turmeric milk daily followed by only 21.3 percent not consuming [Table 2].

Vitamin C

Vitamin C acts as antioxidant by combine with and scavenge many types of oxidizing free radicals and also used in healing wounds. To combat this pandemic, administration of high dose of ascorbic acid, in addition to standard conventional supportive treatments, has been shown to be a safe and effective therapy for severe cases of respiratory viral infection (Hong *et al*., 2020). Interestingly, result of present investigation showed that all (100%) the subjects consume vitamin c by including citrus fruits in their diet while some taking supplement of vitamin c. Use of vitamin c for Covid-19 patients is useful tool to fight against Covid-19 [Table 2].

Results about intake of immunity boosters revealed that majority of subjects found aware about meaning of immunity booster and most of them follow measures to boost immunity. The results of present study is alignment with the study conducted by Gautam *et al*., 2020 concluded that herbs such as *Tulsi, Marich, Sunthi, Dalchini* are the most commonly used and easily available spices in home and can be effective in immuno-regulation for controlling viral infections like Covid-19. Similarly another study conducted by Singh *et al*., 2021 that majority of subjects (71.8%) are taking *kadha* for combating infection and boosting immunity. Most of the subjects (86.1%) thought that there is no side effect of kadha while 13.9% think vice versa. A total of 93.6 percent of people used spices for treating coronavirus or other viral infection as well as boosting immunity. However in present findings, consumption of spices, warm water, chyawanprash, golden milk, citrus fruits, homemade and ayush ministry decoction found as immunity boosters. Hence, it can be concluded that use of immunity boosters may reduce the risk of severity of the disease and helps in faster recovery.

Table 2: Distribution of subjects according to information related to immunity boosters

S.No.	Parameters	Male n=150(50.0)	Female n=150(50.0)	Total N=300(100)
1.	**Immunity boosters**			
	Helps in increase immunity	16(10.7)	18(12.0)	34 (11.3)
	Protect against disease	25(16.7)	19(12.7)	44 (14.7)
	Both	109(72.6)	113(75.3)	222(74.0)
	Don,t know	0(0)	0(0)	0(0)
2.	**Yogasana, pranayama and meditation**			
	Yes	119 (79.3)	121(80.7)	240(80.0)
	No	31(20.7)	28(18.3)	60(20.0)
3.	**Warm water**			
	Yes	138(92.0)	135(90.0)	273(91.0)
	No	12(8.0)	15(10.0)	27(9.0)
4.	**Spices most used for immunity booster**			

	Turmeric	150(100)	150(100)	300(100)
	Garlic	30(20.0)	26(17.3)	56(18.7)
	Coriander	16(10.7)	0(0)	16(5.3)
	Cinnamon	102(68.0)	91(60.7)	193(64.3)
	Any other	83(55.3)	46(30.7)	129(43.0)
5.	**Ayush ministry *kadha***			
	Yes	136(90.6)	129(86.0)	265(88.3)
	No	14(9.3)	21(14.0)	35(11.7)
6.	**Intake of *chyawanprash***			
	Yes	118(78.7)	104(69.3)	222(74.0)
	No	32(21.3)	46(30.7)	78(26.0)
7.	**Ayush ministry guideline**			
	Yes	146(97.3)	116(77.3)	262(87.3)
	No	4(2.7)	34(22.7)	38(12.7)
8.	**Homemade *kadha***			
	Yes	132(88.0)	121(80.7)	253(84.3)
	No	18(12.0)	29(19.3)	47(15.7)
10.	**Golden milk**			
	Yes	121(80.7)	115(76.7)	236(78.7)
	No	29(19.3)	35(23.3)	64(21.3)
11.	**Vitamin C**			
	Yes	150(100)	150(100)	300(100)
	No	0(0)	0(0)	0(0)

Note: Figures in parenthesis indicates percentage of subjects

IV. CONCLUSIONS

An interview schedule developed for obtaining the required information about all the subjects. While assessing general information about the subjects it was observed that majority of the subjects (59.0%) belonged to general category followed by 24.7 per cent from OBC category, 9.7 per cent from schedule caste and 6.7 per cent of them belonged to schedule tribes. Gender-wise data showed that the percentage of females (62.0%) in general category recorded higher as compared to males (56.0%) and equal percentage noted in case of schedule tribes (6.7%). Majority of them were Hindu (94.3%) followed by Jain, Muslim, Sikh and Christian i.e. percent 2.3%, 2.0%, 1.0% and 0.3% respectively. Majority of the subjects (60.7%) were graduated followed by post-graduate and above (24.3%), studied up to high school (13.3%) and only 1.7 percent attended middle school. Higher percentage of females (66.7%) was observed educated as compared to males (54.8%) up to graduation level. All the subjects found to be engaged in sedentary activities irrespective of gender. Most of the subjects (76.0%) belonged to nuclear family followed by joint family (40.0%). Immunity boosters have a significant impact in Covid-19, also in this present study various immunity boosters taken by maximum number of subjects. The results of present investigation demonstrated that 78.7 percent of subjects consumed turmeric milk daily followed by only 21.3 percent not consuming. The ministry has given some measures of boosting immunity which included ayurvedic spices to be added in their diet to enhance immunity, using oil therapy for mouth and nose and steam inhalation. Most of the subjects (87.7%) following Ayush ministry guideline only 12.7 percent did not aware about these measures which are taken by government. Use of vitamin c for Covid-19 patients is useful tool to fight against Covid-19.

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Selection strategies for yield enhancement in barley (*Hordeum vulgare* L.)

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Abstract— *A field experiment with 42 genotypes of barley was conducted in order to select key components and promising genotypes for yield enhancement. The experiment was laid in randomized block design with four replications during 2022-23 at Barley Research Area, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University, Hisar. The genotypic correlation estimates showed significant positive association of grain yield with days to maturity, spike length and 1000-grain weight. The trait viz., days to maturity exhibited the highest positive direct effect on grain yield followed by 1000 grain weight and spike length. Hence, these traits could be considered as suitable selection criterion for the development of high yielding barley varieties. Cluster analysis assigned all the genotypes into four distinct clusters. Cluster I, contained 14 genotypes recorded with shortest plant height having highest number of effective tillers per meter. Cluster II consisted of 8 genotypes, characterized by long spikes and high 1000-grain weight. Cluster III had 11 genotypes with highest grain yield. Cluster IV comprised of nine early maturing genotypes. The average inter-cluster distance was found to be highest between the cluster I and III followed by between cluster I and IV while the lowest inter-cluster distance was observed between clusters III and IV. The improvement in six rowed barley could be achieved through the use genotypes assigned in clusters III, whereas the genotypes which contained in cluster II might be considered as potential parents for two rowed barley to obtain high heterotic response and consequently better segregants for grain yield.*

Keywords— ***Barley, Cluster, Correlation, Direct Effects, Genetic divergence***

I. INTRODUCTION

Barley is a coarse cereal, which is being utilized for human food, livestock feed and as well as for malting and brewing purposes. The barley cultivation requires low inputs in terms of fertilizers and irrigation and has better adaptability to harsh environments. Barley offers many health benefits and has a huge potential to be a future staple cereal substituting wheat and rice in diet for diabetic and people suffering from high cholesterol. It has low glycemic index as well as low amount of anti-nutritional factor *i.e.* phytate along with high beta glucan, that offers many health benefits. In India, the area under barley is about 0.62 m ha with the production and productivity of 1.69 m t and 27.33 kg/ha, respectively. Barley was cultivated on 15,300 hectares with a grain production of 53,300 tons in Haryana which ranks second in average productivity (34.86 q/ha) after Punjab (36.54 q/ha) during 2022-23 [1].

Genetic reconstruction of a plant type is essential for development of high yield potential genotypes by selection and incorporation of yield components. The evaluation of breeding material for genetic variability is vital for present as well as for future crop improvement approaches. The extent and nature of interrelationship among different characters and their contribution towards yield helps in formulating the efficient scheme of multiple trait selection. Genetic diversity is one of the important mechanism of stability of biological systems and is always remained the key parameter of plant breeding [2]. Hence,

their information facilitates the plant breeders in isolation of trait specific superior donors for commencing targeted hybridization programme, as heterotic expression is believed to be associated with genetic divergence among the genotypes used as parents. In addition, appropriate utilization of genotypes requires screening of genetic potential under specific environmental condition for which the breeding programme is designed. The Bioversity International as well as International Union for the Protection of New Varieties of Plants recommended the morphological characterization as criterion to identify accessions for the assessment of genetic diversity [3]. Some findings on genetic diversity of barley have also focused on importance of morphological traits [4, 5]. Several approaches are available to examine genetic diversity in the breeding material. Often cluster analysis has widely been used in order to identify the traits specific donors to be used in a fresh breeding programme for meaningful achievement.

Keeping in view the above, the present investigation was therefore undertaken to identify the key yield components and to select the trait specific promising genotypes for inclusion in yield enhancement strategies in barley under timely sown irrigated conditions.

II. MATERIALS AND METHODS

A total of 42 barley genotypes comprised of two and six row types were grown in a randomized block design with three replications during 2022-23 at Barley Research Area, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University, Hisar. The experimental location is situated between 29°10'N latitude, 75°46'E longitude and at an altitude of 215.2 m above mean sea level in subtropical region of North Western Plain Zone of India. Each genotype was consisted of 6 rows of 5.0 m length grown under timely sown irrigated condition, with spacing of 23 cm between rows. The observations were recorded for eight morphological traits *viz*., days to heading, days to maturity, plant height (cm), effective tillers per meter, spike length (cm), number of grains per spike, 1000-grain weight (g) and grain yield (q/ha). The crop was raised following recommended package of practices. For recording data, five randomly selected competitive plants in each replication were chosen for all the traits under study except of days to heading and maturity, and grain yield which were recorded on plot basis. The mean performance of each genotype was subjected to statistical analysis using statistical software R Studio [6].

III. RESULTS AND DISCUSSION

Correlation coefficient analysis measures the natural relation between various plant characters and determines the component traits on which selection can be used for yield enhancement. The genotypic correlation coefficients between studied traits are depicted in Table 1. The results revealed significant positive correlation of grain yield with days to maturity, spike length and 1000-grain weight. These finding are in accordance with result of Kumar *et al*. [7] for days to maturity and 1000-grain weight. The positive correlation of grain yield with these traits signifies that improvement in one or more of these traits could result in higher grain yield in barley.

Table 1: Estimates of genotypic correlation coefficients for different traits in barley

Traits	DH	DM	PH	ET/m	SL	G/S	TGW	GY
DH	1.00	0.732**	0.300	0.101	0.215	-0.034	0.20	0.143
DM		1.00	0.245	0.139	0.341*	-0.187	0.437**	0.385*
PH			1.00	-0.387*	0.312*	0.369*	-0.174	0.135
ET/m				1.00	-0.452**	-0.619**	0.226	-0.132
SL					1.00	0.245	0.223	0.307*
G/S						1.00	-0.606**	-0.047
TGW							1.00	0.322*

DH: Days to heading, DM: Days to maturity, PH: Plant height, ET/m: Effective tillers per meter, SL: Spike length, G/S: Number of grains per spike, TGW: 1000-Grain weight, GY: Grain yield, *, ** Significant at 0.05 and 0.01 level, respectively

Plant height showed positive and significant correlation with spike length and number of grains per spike. However, it registered significant negative correlation with effective tillers per meter. Further, the results of correlation coefficient implied the significant positive association for days to maturity with days to heading, spike length and 1000-grain weight. Kumar *et al*. [7] also reported positive but non-significant correlation of days to maturity with 1000-grain weight. Similarly, significant negative correlation was found for spike length

and number of grains per spike with effective tillers per meter; and number of grains per spike with 1000-grain weight, thereby indicating that these traits could not be improved simultaneously. Some authors also reported significant positive correlation for days to heading with maturity [8], plant height with spike length [9], days to maturity with days to maturity and spike length; plant height with spike length and number of grains per spike [10]. The negative and significant relation of effective tillers per meter with plant height and number of grains per spike; 1000-grain weight with number of grains per spike also corroborated with the findings of Devi *et al*. [10] under different environments. The negative but non-significant association between spike length and effective tillers per meter was also confirmed by Kumar *et al*. [7] in barley.

Path coefficient presents an effective way of finding direct and indirect sources of correlation. Direct and indirect effects of these components determined on grain yield at genotypic level are presented in Table 2. The results of path coefficient analysis revealed that days to maturity (0.477) exerted the highest positive direct effect on grain yield followed by 1000-grain weight (0.218), spike length (0.060), plant height (0.041) and number of grains per spike (0.040), which support the findings of Devi *et al*. [10] except for number of tillers per meter however, Kumar *et al.* [9] in their findings mentioned positive direct effect of number of tillers per meter on grain yield. Therefore, these traits could be considered as main components for selection in a breeding program for higher grain yield. Path analysis further revealed that though effective tillers per meter had negative direct effect (-0.154) but it highly contributed to grain yield *via* number of grains per spike and spike length. In addition, days to heading contribute *via* number of grains per spike to grain yield as this traits also showed high negative direct effect (-0.257).

Table 2: Direct and indirect effects of different traits on grain yield in barley

Traits	DH	DM	PH	ET/m	SL	G/S	TGW	rg with GY
DH	**-0.257**	-0.188	-0.077	-0.026	-0.055	0.009	-0.052	0.143
DM	0.349	**0.477**	0.117	0.066	0.163	-0.089	0.209	0.385*
PH	0.012	0.010	**0.041**	-0.016	0.013	0.015	-0.007	0.135
ET/m	-0.016	-0.021	0.059	**-0.154**	0.070	0.095	-0.035	-0.132
SL	0.013	0.020	0.019	-0.027	**0.060**	0.015	0.013	0.307*
G/S	-0.001	-0.008	0.015	-0.025	0.010	**0.040**	-0.024	-0.047
TGW	0.044	0.095	-0.038	0.049	0.049	-0.132	**0.218**	0.322*

DH: Days to heading, DM: Days to maturity, PH: Plant height, ET/m: Effective tillers per meter, SL: Spike length, G/S: Number of grains per spike, TGW: 1000-Grain weight, GY: Grain yield, rg: Genotypic correlation, *, ** Significant at 0.05 and 0.01 level, respectively; **Residual effect=0.259**

The distribution of fourty two genotypes of barley into different diverse clusters and their genetic distances are depicted in Table 3. All the genotypes were classified into four distinct clusters (*Fig*. 1). The clustering pattern of genotypes recognized cluster I as largest one with 14 genotypes, followed by cluster III with eleven genotypes. Cluster II and IV possessed eight and nine genotypes, respectively. Kumar *et al.* [11] studied and classified 87 barley genotypes into different clusters based on 10 qualitative traits and also selected the promising genotypes for future barley breeding strategies. The estimates of intra-cluster distances showed highest genetic distance in cluster I (120.20) followed by clusters II with genetic distance of 61.57. The results also revealed that cluster I is the most distantly placed from cluster III which is the maximum (154.12) among all cluster combinations, followed by clusters I and IV (152.32). However, cluster III is most closely placed to cluster IV which is minimum distance (55.25) among inter cluster distances. It is well known that higher the distance between clusters, more the genetic diversity would be between the genotypes. Therefore, highly diverse genotypes would produce better segregants in the following generations enabling further selection and trait improvement. Sarkar *et al*. [12], Ebrahim *et al.* [13], Hailu *et al.* [14] and Devi *et al.* [15] also reported the existence of adequate genetic diversity in their findings in barley crop.

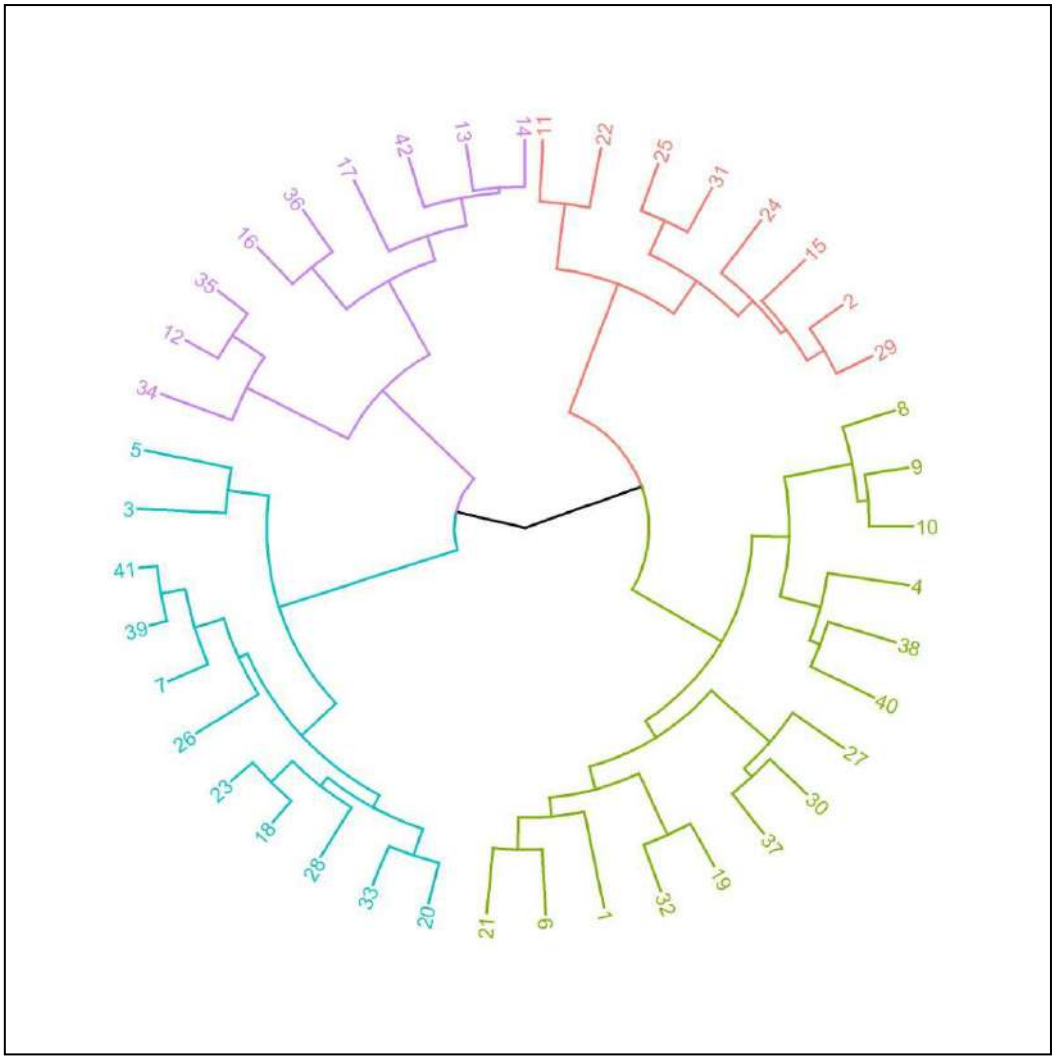

Fig. 1. Dendrogram showing the clustering pattern of 42 genotypes of barley

Table 3: Distribution of 42 genotypes into different clusters and their genetic distances

Genotypes	**No. of Genotypes**	**Cluster Number**	**Genetic distances between clusters**			
			1	**2**	**3**	**4**
BH 21-02 (1), BH 21-01 (4), DWRB 101 (6), BH 21-28 (8), BH 21-37 (9), BH 21-21 (10), BH 21-23 (19), DWRB 182 (21), BH 20-15 (27), BH 20-16 (30), BH 885 (32), DWRB 92 (37), BH 21-33 (38), BH 20-38 (40)	14	1	**120.20**	123.05	154.12	152.32
BH 21-09 (2), DWRB 160 (12), BH 21-05 (15), BH 20-13 (22), BH 20-09 (24), BH 20-10 (25), BH 20-02 (29), BH 20-11 (31)	8	2	123.05	**61.57**	97.48	95.49
BH 21-06 (3), BH 21-22 (5), BH 21-38 (7), BH 21-03 (18), BH 21-12 (20), BH 21-35 (23), BH 22-27 (26), BH 21-26 (28), BH 946 (33), BH 20-40 (39), BH 21-24 (41)	11	3	154.12	97.48	**51.30**	55.25
BH 21-07 (11), BH 21-31 (13), BH 21-10 (14), BH 393 (16), BH 21-36 (17), BH 22-28 (34), BH 22-29 (35), DWRB 137 (36), BH 21-11 (42)	9	4	152.32	95.49	55.25	**50.60**

Values in parenthesis indicates the serial number of genotypes

Almost all of the clusters exhibited substantial variation in mean performance for the characters under study (Table 4). Cluster I possessed genotypes with short plant height having highest effective tillers per meter. Cluster II was characterized by genotypes with long spikes and with high 1000-grain weight. Cluster III was found promising for the traits *viz*., number of grains per spike and grain yield. Cluster VI illustrated with early maturing genotypes. Most diverse and promising genotypes for specific traits selected from different clusters and are shown in Table 5. For improvement of a particular component trait, the promising donors thus identified which plunged in different clusters could be used in crossing programme to obtain high heterotic response and thus better segregants in subsequent generations for yield enhancement in barley. Several studies have also been conducted for assessment of genetic diversity in barley based on different morphological characters for selecting genetically diverse genotypes for hybridization [16, 17, 18].

Table 4: Average performance of clusters for different traits in barley

Traits	Clusters			
	1	**2**	**3**	**4**
Days to heading	85	88	88	**82**
Days to maturity	128	131	130	**124**
Plant height (cm)	**99**	101	111	101
Effective tillers per meter	**156**	117	96	90
Spike length (cm)	7.4	**8.6**	8.5	7.9
Number of grains per spike	26	25	**65**	64
1000-Grain weight (g)	47.9	**57.6**	44.9	39.8
Grain yield (q/ha)	43.6	49.2	**50.3**	39.8

Table 5: Trait specific promising barley genotypes in different clusters

Trait	Promising donors
Days to heading (≤ 80 Days)	BH 21-10, BH 393, BH 21-36, BH 885, DWRB 137, BH 21-11
Days to maturity (≤125 Days)	BH 21-31, BH 21-10, BH 393, BH 21-36, BH 885, DWRB 137, BH 21-11
Plant height (≤90 cm)	DWRB 182, BH 20-11, DWRB 137
Effective tillers per meter (>150)	BH 21-02, DWRB 101, BH 21-37, BH 21-21, BH 21-23, DWRB 182, BH 20-16, BH 20-11, BH 885, DWRB 92, BH 21-33
Spike length (>8.5 cm)	BH 21-22, DWRB 160, BH 21-09, BH 21-06, BH 21-10, BH 21-03, BH 21-35, BH 20-09, BH 20-10, BH 21-26, BH 20-11
Number of grains per spike Six rowed (≥ 70) Two rowed (≥27)	**Six rowed:** BH 21-06, BH 21-22, BH 22-29, BH 21-24, BH 21-11 **Two rowed:** BH 21-09, BH 21-37, DWRB 160
1000 grain weight (>55.0 g)	DWRB 160, BH 21-05, BH 20-13, BH 20-09, BH 20-10, BH 20-16, BH 20-11, DWRB 92
Grain yield (>50 q/ha)	**Six rowed:** BH 21-06, BH 21-38, BH 21-03, BH 21-12, BH 21-35, BH 22-27, BH 21-24 **Two rowed:** BH 21-09, BH 20-09, BH 20-10, BH 20-15, BH 20-16, BH 20-11, BH 21-33

IV. CONCLUSION

From this study, it can be concluded that the improvement in six rowed barley could be achieved through the use genotypes assigned in clusters III, whereas the genotypes which contained in cluster II might be considered as potential parents for two rowed barley keeping days to maturity, spike length and 1000-grain weight traits as suitable selection criterion for the development of high yielding barley varieties under timely sown irrigated conditions.

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Effect of Priming Treatment to Enhance Seed Quality of (*Sorghum Bicolor* (L.) Moench)

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Abstract— *With a view to find out the effect of priming on germination and seedling vigour on sorghum (Sorghum bicolor L.) seeds, experiments were conducted during Kharif 2018 in Seed Technology Laboratory, Department of Seed Science & Technology, CCS Haryana Agricultural University, Hisar (Haryana), India.The seed of two forage sorghum variety (HC 136 and HJ 541) were evaluated under five priming treatments viz. T_1: Untreated (control), T_2: Hydration -dehydration (6 h), T_3:Hydration – dehydration (6 h) + 0.25 % thiram treatment.T_4: Hydration with GA_3 (50 ppm for 6 h),T_5:Hydration with sodium molybdate (500 ppm for 6 h) in complete randomized block design and replicated thrice.Maximum germination percentage and seedling length were observed when seeds primed with T_4:Hydration with GA_3 (50 ppm for 6 h), followed by Hydration – dehydration + 0.25 % Thiram treatment (T_3) similar trend was also observed in all parameters. Among the treatments T_1 Untreated (control) recorded minimum germination %, Seedling Length, seedling dry weight, Seed Vigour index and dehydrogenase activity. Among all the priming treatments T_4 Hydration with GA_3 (50 ppm for 6 h) was found to be the best priming treatment. Moreover, priming treatments have more pronounced effect on HJ541 maintained highest quality parameters followed than HC136 of sorghum seeds.*

Keywords— ***Sorghum, Seed priming, GA_3, Thiram, Vigour index.***

I. INTRODUCTION

Sorghum [(*Sorghum bicolor* (L.)Moench] is the fifth major cereal crop in the world and occupies fifth position in acreage after wheat, rice, maize and barley. It is grown as a staple food crop throughout the Asian and African regions, besides as a forage and fodder crop for livestock in the developed countries like USA, Europe and Japan. Major producers of sorghum in the world are Nigeria, USA, India, Mexico, Argentina, Sudan, Ethiopia, Brazil, China and Australia. It has been classified under family poaceae, tribe Andropogonae and genus sorghum, Sorghum is considered to be one of the drought tolerant crop. The sorghum is cultivated as dual purpose crop ranking fourth among all cereals. Sorghum possesses a variety of anatomical, morphological, and physiological features that enable it to survive in water-limited environments [1]. The fodder sorghum is grown in 8.3 million ha mainly in Western UP, Haryana, Punjab, and Rajasthan and fulfils over two third of the fodder demand during *Kharif* season. The area under fodder cultivation is estimated to be about four per cent of the gross cropped area, which remained static for the last four decades. The traditional grazing lands are gradually diminishing because of urbanization, expansion of cultivable area, grazing pressure and industrialization etc. These factors resulted in severe shortage of feed and fodder to the extent of 26 per cent in dry-crop residues, 35.6 per cent in green fodder and 41 per cent of concentrates. To reduce the demand and supply gap, the production and

productivity of fodder crops needs to be enhanced. As per an estimation only 25-30 per cent of required quantity of quality seed is available in cultivated fodders and <10 per cent in range grasses and legumes in India. Presently, the seed demand of cultivated forages, range grasses and legumes is increasing tremendously. Now, with the development of a number of improved and high yielding varieties in forage crops, it has become important that quality seed should be readily available and supplied to the tanners at reasonable price (http://www.igfri.res.in/publications). Seed is considered as one of the important basic agricultural inputs for obtaining higher yield. Good quality seed acts as a catalyst for realizing the potential of all other inputs in agriculture. Without good seed, the investment on fertilizer, water, pesticides and other inputs will not play the desired dividends. Its importance has been realized with the passage of time and greater realization that efficiency is the key factor to be competitive in all the agricultural ventures. Therefore, the availability of quality seed to the farmer at an affordable price and in time is considered crucial for enhancing and sustaining the agricultural productivity. Therefore, production of quality seed and maintenance of high seed germination is of utmost importance in a seed programme. Seeds are practically worthless if upon planting they fail to germinate and give adequate plant stand in the field in addition to healthy and vigorous plants. The great quality seed is pre-essential to improve the production and yield. It has been affirmed to understand that utilization of value seeds broadened efficiency of yield increased by 15-20 percent[2]. Seed possesses maximum viability and vigour at physiological maturity [3], thereafter, seeds gradually aged and decline in viability and vigour. Seed deterioration leads to reduction in seed quality, performance and stand establishment .Higher moisture content along with high temperature of storage environment, the sooner is the loss of viability [4].Seed ageing causes regular deterioration in all vital cellular components causing thereby advanced loss of viability .Lipid auto-oxidation has also been proposed to be one of the causes of seed ageing [5], which involve the production of free radicals .Such problems convey severe threat to agriculture; hence require management to sustain viability and vigour[6-8].The most sensitive stages, for many crop species submitted to the stress conditions, are seed germination and early seedling growth [9].

Heydecker [10] reported that seed priming is one of the most important developments to help rapid and uniform germination and emergence of seeds and to increase seed tolerance to adverse environmental conditions. Seed priming has presented promising and even surprising results, for many crop seeds. Primed seeds usually show improved germination parameters [11]. Seed priming with nitrate solutions gave better seed quality and field establishment in maize [12]. Potassium permanganate has oxidizing properties and can act as ethylene neutralizer or an antiseptic. It helped in germination of some legume seeds stored for 20 - 44 years [13]. It is found that on-farm' seed priming with KH2PO4 improved fertilizer- use efficiency and increased yield and profit for different crops grown on P deficient soils [14]. Priming in its traditional sense, is soaking of seeds in water before sowing, has been the experience of farmers in India in an attempt to improve crop stand establishment but the practice was without the knowledge of the safe limit of soaking duration. On-farm seed priming involves soaking the seed in water, surface drying and sowing the same day. The rationale is that sowing soaked seed decrease the time needed for germination and allow the seedling to escape deteriorating soil physical conditions, However it's the investigated research, effect of priming treatments viz., Untreated (control), Hydration -dehydration (6 h), Hydration - dehydration (6 h) + 0.25 % thiram treatment, Hydration with GA_3 (50 ppm for 6 h) Hydration with sodium molybdate (500 ppm for 6 h) and evaluate seed quality parameters *viz*., germination per cent, seedling length, seedling dry weight and dehydrogenase activity of sorghum.

II. MATERIALS AND METHODS

The present investigation on seed invigouration aspects of sorghum was conducted during 2017-18 in the laboratory Department of Seed Science and Technology, CCSHAU, Hisar. The details of the materials used and methods adopted for the conduct of various experiments on seed invigouration are described here under.

***Source of seeds*:** Seed material consist of two varieties *viz.* HC136, HJ541 of sorghum crop were taken. Three seed lots of each variety include-fresh, one year and two year old seed stored under ambient conditions. The seed were collected from the Forage Section, Department of Genetics & Plant breeding, CCS Haryana Agricultural University, Hisar.

Treatment details

For this experiment, natural aged seeds of both the varieties were treated with following priming treatments;T_1: Untreated (control), T_2: Hydration -dehydration (6 h) , T_3: Hydration –dehydration (6 h) + 0.25 % thiram treatment, T_4: Hydration with GA_3 (50 ppm for 6 h), T_5: Hydration with sodium molybdate (500 ppm for 6 h). After each treatment seed were dried back to original moisture content. Then different test was directed on the treated seeds to find out the viability percentage of the seed lot.

Observations recorded

The different observations recorded were

I. Final count germination (%)

II. Total seedling length (cm)

III. Total seedling dry weight (mg)

IV. Seedling vigour index –I (Germination percentage × Seedling length)

V. Seedling vigour index –II (Germination percentage × Seedling dry weight)

VI. Dehydrogenase activity test

Standard Germination (%)

100 seeds of each variety with three replications were placed in between adequate moistened rolled towel papers (BP) and kept at 25°C in seed germinator. The first count was taken on 4th day and last count on 7th day and only normal seedlings were considered for percent germination giving to the rules of International Seed Testing Association [15].

Seedling length (cm)

Ten normal seedlings were randomly selected from each replication of both the varieties at the time of final count of standard germination and average seedling length was calculated and expressed in centimetres.

Dry weight per seedling (mg)

Seedling dry weight was evaluated after the final count in the standard germination test (7th day.) The 10 seedlings of each variety replicated thrice were taken. Seedlings dried in a hot air oven for 24 h at 80±1ºC. The dried seedlings of each replication were weighted and average seedling dry weight of each variety was calculated .

Seedling vigour index

Seedling vigour indices were calculated according to the method suggested by [16]

I. ***Vigourindex-I*** =Standard Germination (%) × Average seedling length (cm)

II. **Vigour index-II**= Standard Germination (%)× Average seedling dry weight (mg)

Dehydrogenate activity (O D g^{-1} ml^{-1})

In DHA test, the basic principle for topographical tetrazolium test for seed viability is the reduced of 2, 3, 5-Triphenyl tetrazolium chloride to red formazan by dehydrogenase enzyme in seed embryo. It is a quantitative method which may be used to determine varying dehydrogenase activity between seeds of similar viability and therefore, it is measure of seed vigour. Sample of one gram seed of each variety in three replications were ground and passed through a 20 mesh screen. 5 ml of 0.5% tetrazolium solution was used to soak 200 mg flour for 3-4 h at 38ºC. Then centrifugation was done at 10000 rpm for 3 minutes and the supernatant was poured off. 10 ml acetone was used to extract the formazan for 16 h followed by centrifugation and spectrophotometer was used to determine the absorbance of the solution at 480 nm. These observations were indicated as optical density (O.D.) and thisprocedure as per procedure suggested by [17].

III. RESULT AND DISCUSSION

Priming improved the germination per cent and vigour of sorghum seed significantly over no-priming. The response of low vigour (aged) seeds to seed priming was much higher when compared to high vigour (unaged) seeds. Data showed in table 1 to 3 reveal that all the treatments improved the standard germination, seedling length and seedling dry weight in all the seed lots and varieties. The freshly harvested seed lot (L_1) was observed highest germination percentage and seedling length as compared to One year old (L_2) and Two year old(L_3). The HJ541 variety performed better than HC136 with each priming treatments. The treatment GA_3 (T_4) showed highest improvement in germination percentage and seedling length among varieties and seed lots followed by Hydration -dehydration + 0.25% thiram (T_3) and the lowest improvement was observed in untreated (T_1). The dry matter was increased in all the lots and varieties after treatments. The maximum dry matter was observed under the GA_3 (T_4) treatments as compared to control (T_2), hydration-dehydration (T_3), hydration-dehydration + 0.25% thiram (T_5) and hydration with sodium molybdate (500 ppm) in all lots and varieties. Variety HJ541 found more responsive than HC136 in all the priming treatments and treatment GA_3 (T_4) found more effective than others.Similar finding were also reported in mustard seed by [18], in sunflower seeds by [19] and in sesame seed by [20].Data presented in table 4-6reveal that all the treatments improved the vigour index-1 in all the lots of both the variety. However, lot L1 showed maximum improvement and lot L3, showed minimum improvement within lots when treated with different priming treatments. In both the varieties HJ541 (V2) perform better and HC136 (V1) showed minimum performance. Treatment with GA_3 (50 ppm) showed maximum improvement in both varieties and all three lots.Data presented in Table 5 reveal that the results of all treatments found promising in improving vigour index- II for both the variety and all the seed lots. However, freshly harvested seed lot (L_1) show maximum improvement in vigour Index-II followed by one year old seed lot (L_2), two year old seed lot (L_3). Among treatments GA_3 treatment (T_4) show maximum improvement in vigour index -II followed by Hydration – dehydration + 0.25% Thiram treatment (T_3) and Hydration - dehydration (T_2). In

both the varieties HJ 541 (V_2) shows maximum improvement whereas the minimum improvement was recorded in HC 136 (V_1) when treated with different treatments. Data presented in Table 6 revealed that the results all the treatments enhanced the dehydrogenase enzymes activity in both variety and all the seed lots of sorghum. The maximum increase in dehydrogenase enzymes activity was reported in variety (HJ541) followed by (HC136). Among different seed lot fresh year seed lot (L_1) show maximum increase in enzyme activity followed by one year old seed lot (L_2) and two year old seed lot (L_3). The GA_3 (T_4) treatment shows highest improvement in dehydrogenase enzyme activity in two varieties and each seed lot followed by Hydration – dehydration + 0.25 % Thiram treatment (T_3) and Hydration with sodium molybdate treatment (T_5). These observations were parallel to those already depicted by various workers in different crop such as [21] in cotton; [22] in *Brassica juncea*; [23] in maize; and [24] in Pearl millet. Toselli and Casenave[25] reported that hydro-priming and osmo-priming increase the germination and vigour index.Similar findings were reported by [26] in sunflower seed and in sorghum seeds by [27].

Table 1.Effect of pre-sowing treatments on standard germination (%) of natural aged seed of sorghum

Table 1 (a). Interaction between varieties and treatments

Varieties	**Treatments**					**Mean**
	T_1	**T_2**	**T_3**	**T_4**	**T_5**	
HC136 (V_1)	60.33 (51.41)	61.33 (52.08)	64.22 (53.97)	69.11 (57.36)	61.56 (52.26)	**63.31 (53.41)**
HJ541 (V_2)	71.33 (58.044)	74.00 (59.92)	75.11 (60.63)	80.78 (64.65)	74.33 (60.20)	**75.11 (60.69)**
Mean	**65.83 (54.73)**	**67.67 (56.00)**	**69.67 (57.30)**	**74.94 (61.00)**	**67.94 (56.23)**	

CD (P = 0.05) V= 0.349, T= 0.551, V × T= 0.780

Values in parenthesis are angular transforme.

Table 1 (b). Interaction between treatments and seed lots

Seed lots	**Treatments**					**Mean**
	T_1	**T_2**	**T_3**	**T_4**	**T_5**	
Fresh seed (L_1)	82.21 (65.51)	85.00 (67.24)	85.00 (69.21)	91.59 (73.12)	84.17 (67.55)	**85.58 (68.52)**
One-year-old (L_2)	73.83 (59.22)	75.00 (59.99)	78.50 (61.39)	84.00 (64.91)	77.83 (61.04)	**77.83 (61.31)**
Two-year-old (L_3)	41.45 (39.45)	43.00 (40.76)	45.50 (41.31)	49.33 (44.98)	41.83 (40.10)	**44.22 (41.32)**
Mean	**65.83 (54.73)**	**67.67 (56.00)**	**69.67 (57.30)**	**74.94 (61.00)**	**67.94 (56.23)**	

CD (P = 0.05) L=0.427, T=0.551, L × T=0.955, V × L × T=1.350

Table 2. Effect of pre-sowing treatments on seedling length of natural aged seed of sorghum

Table 2 (a). Interaction between varieties and treatments

Varieties	**Treatments**					**Mean**
	T_1	**T_2**	**T_3**	**T_4**	**T_5**	
HC136 (V_1)	30.01	30.72	32.38	33.77	31.98	31.77
HJ541 (V_2)	30.57	32.00	32.88	34.94	33.27	32.73
Mean	30.29	31.36	32.63	34.36	32.62	

CD (P = 0.05) V=0.077, T=0.121, V × T=0.171

Table 2 (b).Interaction between treatments and seed lots

Seed lots	**T_1**	**T_2**	**T_3**	**T_4**	**T_5**	**Mean**
Fresh seed (L_1)	31.14	33.95	35.66	37.05	35.61	**34.68**
One year old (L_2)	31.54	31.70	32.72	35.55	33.15	**32.93**
Two year old (L_3)	28.20	28.43	29.50	30.49	29.10	**29.14**
Mean	**30.29**	**31.36**	**32.63**	**34.36**	**32.62**	

CD (P = 0.05), L=0.094, T=0.121, L×T=0.210, V× L×T=0.297

Table 3.Effect of pre-sowing treatments on seedling dry weight of natural aged seed of sorghum

Table 3 (a). Interaction between varieties and treatments

Varieties	**Treatments**					**Mean**
	T_1	**T_2**	**T_3**	**T_4**	**T_5**	
HC136 (V_1)	12.04	12.07	12.14	12.34	12.08	**12.13**
HJ541 (V_2)	12.358	12.43	12.47	12.67	12.45	**12.48**
Mean	**12.20**	**12.25**	**12.35**	**12.51**	**12.26**	

CD (P = 0.05) V=0.012, T=0.019 V x T=0.027

Table 3 (b). Interaction between treatments and seed lots

Seed lots	**Treatments**					**Mean**
	T_1	**T_2**	**T_3**	**T_4**	**T_5**	
Fresh seed (L_1)	13.62	13.43	13.50	13.80	13.44	**13.51**
One year old (L_2)	12.50	12.55	12.61	12.81	12.56	**12.60**
Two year old (L_3)	10.73	10.77	10.81	10.92	10.78	**10.80**
Mean	**12.20**	**12.25**	**12.31**	**12.512**	**12.26**	

CD (P = 0.05) L=00.15, T=0.019, L x T=0.034, V x L x T=0.048

Table 4.Effect of pre-sowing treatments on vigour Index-I & II of natural aged seed of sorghum

Table 4 (a). Interaction between varieties and treatments

Varieties	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
HC136 (V_1)	1,870.18	1,941.05	2,108.02	2,395.69	2,059.48	**2,074.88**
HJ541 (V_2)	2,221.37	2,393.24	2,496.82	2,837.49	2,480.76	**2,485.93**
Mean	**2,045.77**	**2,167.15**	**2,302.42**	**2,616.59**	**2,270.12**	

CD (P = 0.05) V=17.089, T=27.020, V x T=38.212

Table 4 (b). Interaction between treatments and seed lots

Seed lots	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
Fresh seed (L_1)	2,765.67	2,886.98	2,997.87	3,235.19	2,914.84	**2,960.11**
One year old (L_2)	2,304.36	2,378.03	2,569.66	2,928.25	2,593.52	**2,554.76**
Two year old (L_3)	1,067.30	1,236.42	1,339.73	1,686.33	1,302.00	**1,326.36**
Mean	**2,045.77**	**2,167.15**	**2,302.42**	**2,616.59**	**2,270.12**	

CD (P = 0.05) L=20.930, T=27.020, L x T=46.800, V x L x T=66.185

Table 5.Effect of pre-sowing treatments on vigour Index-II of natural aged seed of sorghum

Table 5 (a). Interaction between varieties and treatments

Varieties	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
HC136 (V_1)	752.014	772.061	793.158	844.06	781.022	**788.463**
HJ541 (V_2)	894.706	929.652	962.058	1,032.35	940.214	**951.797**
Mean	**823.36**	**850.857**	**877.608**	**938.207**	**860.618**	

CD (P = 0.05) V=4.076, T=6.445, V x T=9.114

Table 5 (b). Interaction between treatments and seed lots

Seed lots	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
Fresh seed (L_1)	1,106.830	1,142.000	1,181.340	1,255.630	1,155.570	**1,168.27**
One year old (L_2)	923.515	953.897	980.715	1,042.990	955.540	**971.332**
Two year old (L_3)	439.735	456.673	470.767	515.998	470.747	**470.784**
Mean	**823.360**	**850.857**	**877.608**	**938.207**	**860.618**	

CD (P = 0.05) L=4.992, T=6.445, L x T=11.163, V x L x T=13.243

Table 6. Effect of pre sowing treatment on dehydrogenase enzyme activity of natural aged seed of sorghum

Table 6 (a). Interaction between variety and treatments

Varieties	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
HC136 (V_1)	0.449	0.452	0.456	0.465	0.451	**0.455**
HJ541 (V_2)	0.505	0.511	0.518	0.527	0.511	**0.514**
Mean	**0.477**	**0.482**	**0.487**	**0.496**	**0.481**	

CD (P = 0.05) V=0.001, T=0.002, V x T=0.003

Table 6 (b). Interaction between seed lot and treatments

Seed lots	Treatments					Mean
	T_1	T_2	T_3	T_4	T_5	
Fresh seed (L_1)	0.586	0.595	0.606	0.619	0.594	**0.600**
One year old (L_2)	0.460	0.464	0.466	0.474	0.463	**0.466**
Two year old (L_3)	0.385	0.387	0.388	0.395	0.387	**0.388**
Mean	**0.477**	**0.482**	**0.487**	**0.496**	**0.481**	

CD (P = 0.05) L=0.001, T=0.002, L x T=0.003, V x L x T=0.0

IV. CONCLUSION

Significant amount of variation was observed in both of the varieties and all the seed lots for all characters. All priming treatments enhance the seed quality considerably in case of all physiological constraints in natural aged seed lots. Among several priming treatments, hydration with GA_3 (50 ppm for 6 h) was discovered predominant for improving seed quality in both the variety of all the lots of sorghum seed. HJ541 was recorded superior variety established on majority of the germination and vigour constraints results whereas HC 136 was recorded inferior.Priming of the seeds with different treatments was found effective to enhance the seed value in fresh as well as marginal seed lot *i.e*. one year seed lot. GA_3 (50ppm for 6 h) was discovered well priming treatment for improving the quality of seeds followed by hydration- dehydration (6 h) + 0.25% thiram treatments. All the priming treatments indicated maximum effect on HJ 541 followed by HC136.

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Evaluation of Land Use and Road System for Urban Planning in Luohu District, Shenzhen, Based on GIS

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Abstract— *This study aims to evaluate the urban planning system of land use and road system in Luohu, Shenzhen. Based on remote sensing images, 30 m DEM data, population data, and various urban planning standards, the land use types in the Luohu built-up area are determined through comprehensive analysis and evaluation: a road system that covers the slope, area, area ratio, and intersection spacing of road longitudinal sections, as well as a bus system that covers the density of the bus network, non-linear coefficient, network length, average station spacing, repetition coefficient, and station coverage. The study results show that the overall urban planning of Shenzhen Luohu is incomplete, and the land use, road system, and bus routes in the built-up area are not reasonable enough. Finally, suggestions are proposed to address the shortcomings, such as revitalizing existing land resources and promoting the efficient and intensive use of industrial land.*

Keywords— ***Geographic Information System (GIS), Land use; Road system, Bus routes, City planning***

I.INTRODUCTION

Urban planning is the blueprint for urban development and construction, as well as the basic basis for urban construction and management. It has a huge, complex, and long-term impact on urban development and the ecological environment of surrounding areas. Therefore, evaluating urban planning can promote the implementation of urban sustainable development strategies (Mo, 2006). Generally speaking, study on urban planning systems can be evaluated from three aspects: land use, road network systems, and public transportation routes. Among them, land use planning is an indispensable link in urban development, referring to the process of rational allocation of land resources within the city, and is a concept that is carried out to meet the needs of sustainable urban development (Cheng, 2024). Based on the current socio-economic and natural conditions as well as the requirements of national economic development coordinate the total supply and demand of land and arrange the total amount of land use. It has the characteristics of protecting and regulating land use, so it

is also known as the "gate" of urban development (Hu, 2010).

In addition, the planning of the road network system is the main foundation of urban transportation construction. Assuming that land use is likened to the body of the city, the road network system is like the blood of the body, responsible for the transportation of blood and nutrients to the city. Whether its planning is scientific and reasonable directly affects various conditions of urban development, including whether it can adapt to social and economic development and whether various modes of transportation can coordinate and closely cooperate with each other. Whether the road network system produces the best socio-economic benefits and whether the transportation of people and things is convenient, fast, comfortable, economical, and safe (Chen, 2009), Among them, urban public transportation is an indispensable element of urban travel, an artery to ensure the normal operation of urban production and life, and an important infrastructure to improve the comprehensive function of the city. It plays an important role in the development of various industries in the city, the prosperity of the economy and cultural undertakings, and the connection between urban and rural areas (Ma, 2003; Meng, 2020).

Based on this, this study adopts GIS technology as the foundation and evaluates land use, road network systems, and public transportation roads through method analysis and standard indicator evaluation, attempting to interpret whether the various standards in the study area comply with modern urban planning indicators and propose relevant suggestions.

II. STUDY AREA AND DATA SOURCES

2.1 Study Area

Luohu District is located in the south-central part of Shenzhen City and the central part of the Shenzhen Special Economic Zone. Its geographical coordinates are 114 ° 06′ -114 ° 22′ E and 22 ° 53′ -22 ° 62′ N. It is bounded by the Wutong Mountain Bogong Ao watershed and Yantian District in the east, connected to Futian District by the middle line of Hongling Road in the west, and bordered by Longgang District and Longhua District in the north (Figure 1). The total area of the jurisdiction is 78.79 square kilometers, including 36.02 square kilometers of built-up areas.

According to the 2022 National Economic and Social Development Statistics Bulletin, the urban green coverage area of Luohu District is 5176.74 hectares, with a green coverage rate of 64.6%. The area of garden green space is 4987.3 hectares, of which the area of park green space is 1712.1 hectares. There are 111 parks, including 15 municipal parks and 96 community parks. As of 2022, the permanent population of Luohu District is 1.018 million. The built-up areas in Luohu District are concentrated in the central and southeastern parts, with mostly mountainous areas on the east and west sides, scattered in the built-up areas, and a large area of water in the northeast. To serve the transportation of people in the area, there are 11 main roads in both horizontal and vertical directions, with a total length of 208.42 kilometers. There are currently 183 bus routes, 15.27 hectares of bus stops, and more than 600 bus stops.

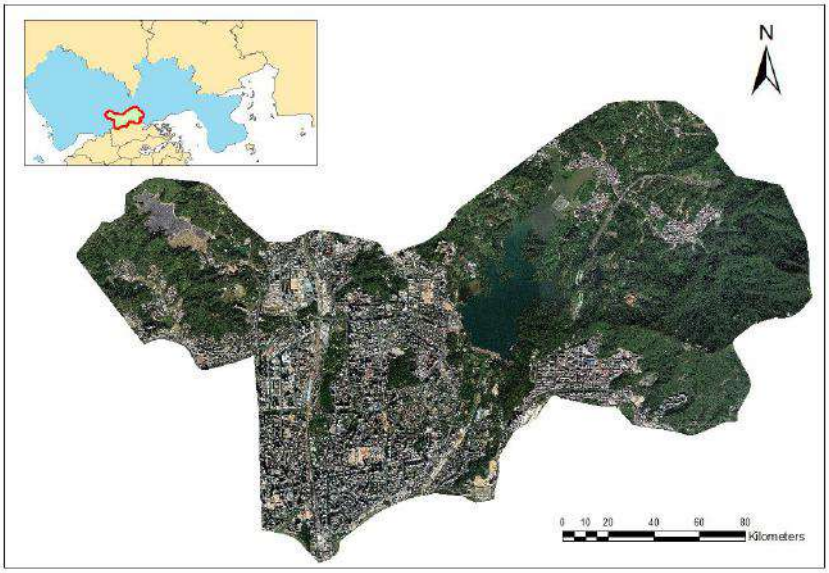

Fig.1 Map of the Study Area

2.2 Data Sources

This study mainly used three types of data: vector boundary range data in Luohu District, image data, and 30m DEM elevation data (Table 1).

Table 1 Data Sources

Data type	Data sources	Data usage
Vector boundary range data	Resource and Environmental Science and Data Center（https://www.resdc.cn/）	Used to delineate the scope of the research area
Image data	LocaSpace Vewer（http://www.locaspace.cn/）	Used for vectorizing urban functional areas
Demographic data	Shenzhen Luohu District People's Government http://www.szlh.gov.cn/wxlh/zjlh/lhsj/）	Used to calculate per capita urban construction land and per capita individual construction land
30mDEM	Geospatial data cloud (https://www.gscloud.cn/）	Used to calculate the slope of road profiles
Code for Urban Road Traffic Planning and Design (GB 50220-95)	Ministry of Housing and Urban Rural Development of the People's Republic of China（https://www.mohurd.gov.cn/）	Used to evaluate the density, non-linear coefficient, repetition coefficient, length, average station distance, and station coverage of urban road longitudinal section slope, road area, and bus route network
Classification of Urban Land and Standards for Planning and Construction Land (GB 50137-2011)		Used to evaluate per capita urban construction land, per capita individual construction land, construction land structure, and classification of urban construction land built-up areas
Design Specification for Urban Road		Used to evaluate the spacing between road

Intersections (CJJ 152-2010)		intersections
Design Code for Urban Road Traffic Facilities (GB 50688-2011)		
Design Specification for Urban Road Engineering (CJJ37-2012)	Ministry of Housing and Urban Rural Development of the People's Republic of China(http://swj.sz.gov.cn/attachment/1/1151/1151906/2945061.pdf）	Used for graded urban roads
Shenzhen Urban Planning Standards and Guidelines	Shenzhen Planning and Natural Resources Bureau(https://www.sz.gov.cn/attachment/1/1133/1133902/10013132.pdf）	Write a land balance sheet

III. METHODOLOGY

3.1 Study Schema

In order to evaluate the urban planning of Luohu District, this study starts with the land use, road system, and public transportation roads in the built-up area. Firstly, the registered image maps are vectorized to obtain the urban transportation roads, urban functional area division, and public transportation routes and stations. Based on the processed transportation road network, the longitudinal slope, area, and area ratio of the road, as well as the spacing between intersections, are obtained. Based on bus routes and stations, the density of bus routes is obtained using fishing net tools. The non-linear coefficient, network length, and average station distance of bus routes are obtained using distance measurement tools. The bus repetition coefficient is obtained through buffer zone analysis. The 800 m bus service area is obtained through network analysis, and the coverage rate of bus stations is obtained (Figure 2).

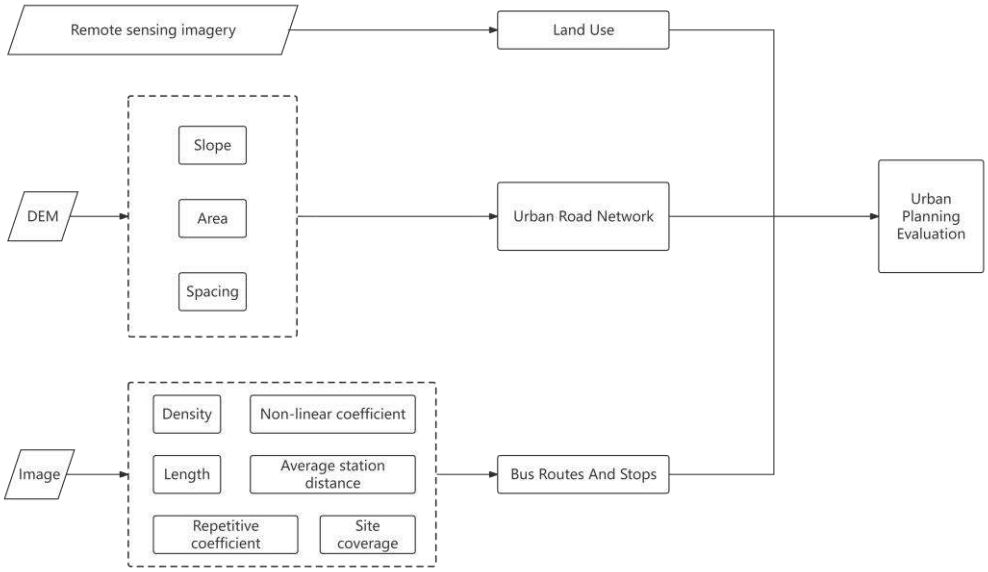

Fig.2 The Flowchart of the Study Schema

3.2 Land Use in Built-up Areas

3.2.1 Evaluation Criteria for Per Capita Urban Construction Land

According to the Classification and Planning of Urban Land and the Standard for Construction Land (GB 50137-2011), per capita urban construction land refers to the area of urban construction land within a city divided by the number of permanent residents within that area, with a unit of m2/person. The evaluation standard is that the per capita urban construction land index for planning should be determined comprehensively based on the current per capita urban construction land index, the climate zone where the city is located, and the planned population size, using Table 2 as the standard. Meanwhile, it should comply with the limit requirements of the allowed per capita urban construction land index and the allowed adjustment range in the table.

Table 2 Planning per capita urban construction land indicators（m^2/ person）

Climate zone	Current per capita urban construction land indicators	Permissible per capita urban construction land indicators for planning	Allow adjustment range		
			Planned population size ≤200 thousand people	Planned population size 201 ~ 500 thousand people	Planned population size ＞500 thousand people
I、II、VI、VII	≤65.0	65.0 ~ 85.0	＞0.0	＞0.0	＞0.0
	65.1 ~ 75.0	65.0 ~ 95.0	+0.1 ~ +20.0	+0.1 ~ +20.0	+0.1 ~ +20.0
	75.1 ~ 85.0	75.0 ~ 105.0	+0.1 ~ +20.0	+0.1 ~ +20.0	+0.1 ~ +15.0
	85.1 ~ 95.0	80.0 ~ 110.0	+0.1 ~ +20.0	-5.0 ~ +20.0	-5.0 ~ +15.0
	95.1 ~ 105.0	90.0 ~ 110.0	-5.0 ~ +15.0	-10.0 ~ +15.0	-10.0 ~ +10.0
	105.1 ~ 115.0	95.0 ~ 115.0	-10.0 ~ -0.1	-15.0 ~ -0.1	-20.0 ~ -0.1
	＞115.0	≤115.0	＜0.0	＜0.0	＜0.0
III IV V	≤65.0	65.0 ~ 85.0	＞0.0	＞0.0	＞0.0
	65.1 ~ 75.0	65.0 ~ 95.0	+0.1 ~ +20.0	+0.1 ~ 20.0	+0.1 ~ +20.0
	75.1 ~ 85.0	75.0 ~ 100.0	-5.0 ~ +20.0	-5.0 ~ +20.0	-5.0 ~ +15.0
	85.1 ~ 95.0	80.0 ~ 105.0	-10.0 ~ +15.0	-10.0 ~ +15.0	-10.0 ~ +10.0
	95.1 ~ 105.0	85.0 ~ 105.0	-15.0 ~ +10.0	-15.0 ~ +10.0	-15.0 ~ +5.0
	105.1 ~ 115.0	90.0 ~ 110.0	-20.0 ~ -0.1	-20.0 ~ -0.1	-25.0 ~ -5.0
	＞115.0	≤110.0	＜0.0	＜0.0	＜0.0

Based on the analysis of current land use statistics and the principles of conservation and intensive land use, this standard sets the upper and lower limits of per capita urban construction land indicators for urban planning located in climate zones I, II, VI, and VII at 65.0~115.0 m^2/person and the upper and lower limits of per capita urban construction land indicators for urban planning located in climate zones III, IV, and V at 65.0~110.0 m^2/person.

The per capita urban construction land quota for newly built cities should be determined within 85.1~105.0 m^2/person. The per capita urban construction land quota for the capital should be determined within the range of 105.1 to 115.0 m^2/person. When remote areas, cities in ethnic minority areas, as well as some mountainous cities, industrial and mining cities with small populations, scenic tourism cities, etc., if do not meet the requirements, a special demonstration should be conducted to determine the planned per capita urban construction land index, and the upper limit should not exceed 150.0 m^2/person.

3.2.2 Evaluation Criteria for Per Capita Individual Construction Land

Per capita single construction land refers to the area of residential land, public management and service land, road and transportation facility land, as well as green space and square land within the city, divided by the number of permanent residents within the urban construction land range, in m^2/person. According to the "Classification of Urban Land and Standards for Planning and Construction Land" (GB 50137-2011), the per capita residential land indicators for planning should comply with the provisions of Table 3.

Table 3 Indicators of Per Capita Residential Land Area（m^2 / person）

Climatological zoning for buildings	I、II、VI、VII Climate zone	III、IV 、V Climate zone
Per capita residential land area	28.0 ~ 38.0	23.0 ~ 36.0

The planned per capita area of public management and public service land should not be less than 5.5 m^2/person. The planned per capita land area for roads and transportation facilities should not be less than 12.0 m^2/person. Then, the planned per capita green space and square land area should not be less than 10.0 m^2/person, among which the per capita park green space area should not be less than 8.0 m^2/person.

3.2.3 Evaluation Criteria for Construction Land Structure

The structure of construction land refers to the proportion obtained by dividing the area of residential land, public management and service land, industrial land, road and transportation facilities land, as well as green space and square land within a city by the area of urban construction land (%). According to the Urban Land Classification and Planning Construction Land Standard (GB 50137-2011), the proportion of residential land, public management and public service land, industrial land, road and transportation facility land, and green space and square land in urban construction land planning should comply with the provisions of Table 4.

The planned urban construction land structure of industrial and mining cities, scenic tourism cities, and other cities with special circumstances can be determined based on the actual situation.

Table 4 Planning Urban Construction Land Structure

Land use name	Proportion of urban construction land（%）
Residential land	25.0 ~ 40.0
Public management and public service land	5.0 ~ 8.0
Industrial land	15.0 ~ 30.0
Road, street and transportation	10.0 ~ 25.0
Green space and square land	10.0 ~ 15.0

3.2.4 Land Balance Sheet

The urban land balance sheet is a planning tool for urban development, used to display and plan the area and proportion of various types of land within the city. It provides a detailed list of eight types of land, including public management and public service land, logistics and warehousing land, industrial land, green space and square land, commercial service facility land, residential land, road and transportation facility land, and urban construction land. It also provides the land area, proportion of urban construction land, per capita urban construction land status, and planned area for each type of land. The land use balance sheet plays an important role in urban planning, resource allocation, sustainable development, policy formulation, and public participation. It is an important tool for urban planning and management, which helps to achieve rational development and an optimized layout of the city.

3.3 Road System Evaluation Methods

3.3.1 Road Longitudinal Section Slope

The longitudinal slope of urban roads refers to the ratio of the height difference between two points on the same slope section of a route and its horizontal distance, expressed as a percentage. The maximum longitudinal slope is the maximum value of the longitudinal slope determined by factors such as road grade, natural conditions, driving requirements, and street-facing buildings. The minimum longitudinal slope is the minimum longitudinal slope specified for sections with poor drainage for longitudinal drainage needs.

In practical situations, the slope of a road is generally expressed in two ways: percentage display or the degree method. The percentage method is the most commonly used method for measuring slope, which is the percentage

$$S = \frac{(h-l)}{100}$$

of the elevation difference between two points and their total length. Its calculation formula is as follows:

In the formula, S is the longitudinal slope of the road, h is the elevation difference of the road, and l is the total length of the road

3.3.2 Road Area

The urban road area refers to the length of the road multiplied by the width of the road. According to the "Shenzhen Urban Planning Standards and Guidelines" issued by the Shenzhen Municipal Planning and Natural Resources Bureau, the control of road width is shown in Table 5:

Table 5 Corresponding widths of roads of different levels

Road class	Total Road Width
Expressway	35 - 80
Main road	30 - 50
Secondary trunk road	26 - 35
by-pass	16 - 25

The urban road area ratio, also known as the "urban road area density," is expressed as the percentage of the area of roads (roads refer to roads with a width of 3.5 meters or more paved, excluding sidewalks) within the urban built-up area. The urban road area ratio is an important economic and technical indicator that reflects the ownership of urban roads in urban built-up areas.

3.3.3 Road Intersection Spacing

Road intersection spacing refers to the distance between the centers points of two adjacent road intersections. From a transportation perspective, in general, there is an intersection every 200 meters, which may feel too dense and conflicted, making it inconvenient for vehicle driving and traffic management. However, there is only one intersection between 800-1000m, which is not convenient for the entry and exit of residential areas and neighborhoods. Therefore, in order to facilitate the walking and driving of pedestrians and vehicles, the distance between road intersections should be 300–800 m. Different levels of roads have different functions and design speeds, resulting in varying distances between corresponding road intersections. The spacing between intersections of urban expressways, main roads, secondary roads, and branch roads should be 1500–250 m, 700–1200 m, 350–500 m, and 150–250 m, respectively (Shi, 2007).

3.4 Public Transport Network

The public transportation network is composed of fixed stations arranged for urban public transportation based on urban streets. This study will use the evaluation index system summarized by Li et al. (2003) to evaluate the public transportation network in Luohu District, including road network density, road network length, non-linear coefficient, repetition coefficient, average station distance, and station coverage.

3.4.1 Road Network Density

Road network density refers to the ratio of the total length of the road centerline of the bus route to the area of the bus service city. It is used to reflect the relative size and average distribution of the bus network and can also reflect the degree to which residents are close to the bus route. The formula is as follows:

$$\delta = \frac{L_0}{F}$$

In the formula, δ refers to the density of the public transportation network, L0 refers to the total length of the road centerline of the bus route, and F refers to the area of the bus service city, unit in km/ km^2.

According to the Code for Urban Road Traffic Planning and Design (GB 50220-95), the density of the public transportation network planned in the city center should reach δ = 3–4 km/ km^2. In urban fringe areas, it is necessary to achieve δ = 2–2.5 km/ km^2. According to theoretical analysis, the average density of the urban public transportation network is δ = 2.5 km/ km^2 (Kuang, 2005).

The density of a city's public transportation network must be appropriate. It is generally believed that the higher the density of the public transportation network, the more convenient the transportation connections will be. However, if the density is too high, it can cause problems such as inefficient urban land use, severe barriers between urban areas, increased investment in urban road construction, and the formation of too many intersections, affecting the speed of vehicle travel and the capacity of main roads. A low density can cause traffic vehicles to detour, increase travel time, and fail to fully utilize the distribution function of the public transportation system, leading to traffic congestion (Li, 2008).

3.4.2 Road Network Length

$$l_{\min} \le l \le l_{\max}$$

In the formula, lmin is the upper limit of the route length (km), and lmax is the lower limit of the route length (km). According to the operating requirements of the "Urban Road Traffic Planning and Design Specification (GB 50220-95)" issued by the National Bureau of Technical Supervision, the lmin is about 5km and the lmax is about 15km (Liu, 2008).

3.4.3 Non Linearity Coefficient

$$[\frac{l_k}{d_k}] \le [\frac{l}{d}]_{\max}$$

In the formula, l_k is the length of line k (km), and d_k is the spatial straight-line distance between the starting and ending stations of the line (km). According to the "Code for Urban Road Traffic Planning and Design (GB50220-95)," the non-linear coefficient of public transportation lines should not exceed 1.4, and the average non-linear coefficient of the entire network is 1.15–1.2, which is suitable for evaluating the convenience of the connection between different road network types and the distribution points of passenger and freight flow routes (Li, 2008).

3.4.4 Repetition Coefficient

The repetitive coefficient refers to the ratio of the total length of the bus operation route to the total length of the bus network. The formula used to reflect the density of bus routes on major urban roads and the degree of repetition of bus routes is as follows:

$$\mu = \frac{L}{L_0}$$

In the formula, μ refers to the repetition coefficient of the public transportation network, L refers to the total length of bus operation routes, L0 refers to the total length of the bus network, and the ratio has no units. According to the Code for Urban Road Traffic Planning and Design (GB50220-95), the repetition coefficient μ, the upper limit

is 3.1, and the lower limit is 1.2. It is generally believed that μ = 1.2–1.5 is a reasonable value (Kuang, 2005).

3.4.5 Average Stop Spacing

The calculation of average station distance is based on the average distance between the bus stop and its nearest bus stop, calculated using the "distance matrix" tool of QGIS software. According to the "Code for Urban Road Traffic Planning and Design (GB 50220-95), the average station distance in the urban area should not exceed 400m, and the average station distance in the suburbs should not exceed 800m.

3.4.6 Bus Stop Coverage

Station coverage is the percentage of the service area of bus stops in the urban land area. It refers to the ratio of the total number of stops N on the bus route to the service area during the statistical period. It is used to represent the average distribution of bus stops within the bus service area. The coverage rate of bus stops reflects the degree to which residents are close to bus stops. It is one of the important indicators used to measure whether it is convenient for citizens to use buses. It can be expressed as follows (Wang, 2008):

Bus stop coverage = number of bus stops x service area of bus stops / land area of urban zoning

According to the "Code for Urban Road Traffic Planning and Design (GB 50220-95), the service radius of urban bus stops is divided into three ranges: 300m, 500m, and 800m. The service area of public transportation stations is calculated based on the 500-meter service radius and shall not be less than 90%. The 300m and 500m radius service areas are more meaningful for the study of conventional ground bus systems, and the 800m radiation radius is more suitable for the analysis of backbone public transportation with large operating volumes, such as subways. Therefore, this study takes the 500-meter-radius service area as the main evaluation indicator (Li, 2015). However, according to the above calculation formula, the sum of areas is not the algebraic sum of all areas covered by the region, but the area after geometric merging of the regions, that is, the result of subtracting the overlapping area from the algebraic sum of areas.

IV. ANALYSIS AND RESULTS

4.1 Analysis of Land Use in Built-up Areas

The analysis of functional zones for urban construction land is based on Figure 3. Among them, the functional areas of urban construction land are classified according to the "Classification and Planning of Urban Land and Construction Land Standards (GB50137-2011)". In addition to public facility land, they are divided into seven types of land: residential land, commercial service facility land, public management service facility land, industrial land, logistics and warehousing land, road and transportation land, green space, and square land. In addition, water areas are also divided. Among them, the area and total area data of each functional area of urban construction land are shown in Table 6:

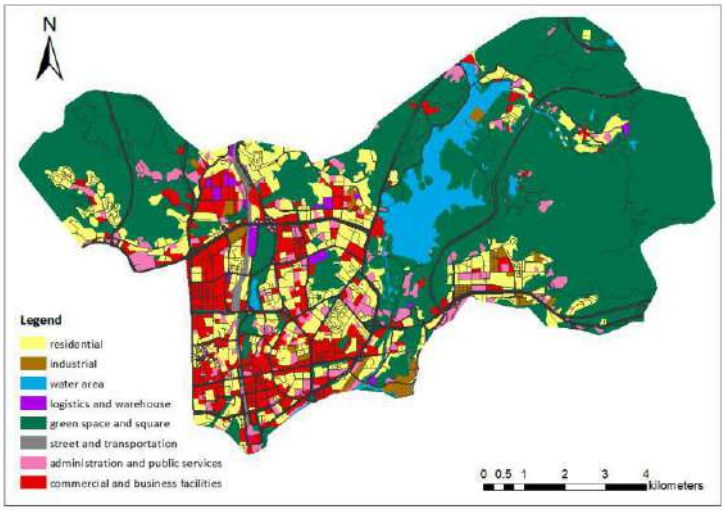

Fig.3 Classification of Functional Areas for Urban Construction Land

Table 6: Area and Total Area of Each Functional Area of Urban Construction Land

Functional Area	Unit（km2）
Public management and public service land	4.28
Logistics and warehousing land	0.55
Industrial land	1.20
Green space and square land	43.91
Land for commercial service facilities	7.29
Residential land	13.15
Roads and transportation land	0.46
total area	71.32

4.1.1 Analyze Per Capita Urban Construction Land

Based on the per capita urban construction land standard, Luohu District in Shenzhen belongs to the Class IV climate zone, with a per capita urban construction land area range of 65.0 to 110.0 square meters (Table 7).

Table 7 Per Capita Urban Construction Land Standards

Climatological zoning for buildings	I、II、VI、VII Climate zone	III、IV 、V Climate zone
Per capita urban construction land area	65.0 ~ 115.0	65.0 ~ 110.0

According to the data vectorized by ArcGIS software, the total area of urban construction land in Luohu District, Shenzhen, is 70.06 km2. The permanent population is 1.018 million, and the per capita urban construction land area in Luohu District is calculated by dividing the total urban construction land area by the local permanent population. The per capita urban construction land area in Luohu District is 70.06 square meters per person. Meet the standard range of 65.0 to 110.0 square meters (Table 8).

Table 8 Per Capita Urban Construction Land Area in Luohu District

Total area （㎢）	Permanent population （ ten thousand people）	Per capita urban construction land area （㎡/ people）
71.32	101.8	70.06

4.1.2 Analyze Per Capita Individual Construction Land

Based on the per capita construction land standard, Luohu District in Shenzhen belongs to the Class IV climate zone, with an average residential land area of 23.0–36.0 square meters per person. The per capita area of public management and public service land in the plan should not be less than 5.5 ㎡/person. The planned per capita land area for roads and transportation facilities should not be less than 12.0 ㎡/person. The planned per capita green space and square land area should not be less than 10.0 ㎡/person, among which the per capita park green space area should be greater than 8.0 ㎡/person (Table 9).

Table 9 Per Capita Single Construction Land Standards

Climatological zoning for buildings	I、II、VI、VII Climate zone	III 、IV 、V Climate zone
Per capita residential land area	28.0 ~ 38.0	23.0 ~ 36.0

The public management and public service land in Luohu District is 4.3 ㎡/person, which is lower than the national standard of 5.5 ㎡/person and does not meet the requirements of the national standard. The land for green spaces and squares is 43 ㎡/person, which is higher than the national minimum standard of 10.0 ㎡/person and meets the national standard requirements. The road and transportation land area is 0.5 ㎡/person, which is

significantly lower than the national standard of 12.0 ㎡/person and does not meet the requirements. The residential land area is 13 ㎡/person, which is lower than the national standard of 23.0 ㎡/person and does not meet the requirements (Table 10).

Table 10: Per Capita Area of Various Construction Land in Luohu District

Functional Area	Unit (km^2)	Per capita area (㎡/ person)
Public management and public service land	4.36	4.28
Logistics and warehousing land	0.56	0.55
Industrial land	1.23	1.20
Green space and square land	43.91	43.91
Land for commercial service facilities	7.42	7.29
Residential land	13.38	13.15
Roads and transportation land	0.46	0.46

4.1.3 Analyze the Structure of Construction Land

Based on the construction land structure standards and the analysis of the proportion of various land uses in Luohu District, it is found that the proportion of residential land is 19%, which does not meet the range of 25.0–40.0% in the national standard. The proportion of public management and public service land is 6%, which meets the national standard of 5.0–8.0%. The proportion of industrial land is 2%, which does not meet the national standard range of 15–30%. The land for roads and transportation facilities is 1%, which does not meet the national standard range of 10.0–25.0%. The proportion of green space and square land is 62%, exceeding the appropriate range of national standards of 10.0–15.0 (Table 11).

Table 11 Proportion of Land Use in Luohu District

Functional Area	The proportion of construction land structure standards（%）	The proportion of various land uses（%）
Public management and public service land	5.0 ~ 8.0	6%
Logistics and warehousing land		1%
Industrial land	15.0 ~ 30.0	2%
Green space and square land	10.0 ~ 15.0	62%
Land for commercial service facilities		10%
Residential land	25.0 ~ 40.0	19%
Roads and transportation land	10.0 ~ 25.0	1%

4.1.4 Land Balance Sheet

Based on the planning and requirements for various types of land in the Shenzhen Urban Planning Standards and Guidelines and combined with actual situations, the land area, proportion of urban construction land, and per capita urban construction land status and planning for eight types of land are obtained. Among them, the area of land for public management and public services, industrial land, residential land, roads, and transportation facilities is planned to expand, while the area of land for logistics and warehousing, commercial service facilities, and urban construction remains the same. The area of land for green spaces and squares is planned to decrease (Table 12).

Table 12 Land Balance Table of Luohu District, Shenzhen

Land use code	Land use name	Land area（km2）		Proportion of urban construction land（%）		Per capita urban construction land （ km2/ person）	
		Present situation	Plan	Present situation	Plan	Present situation	Plan
A	Public management and public service land	4.36	5.59	6	8	4.28	5.5
W	Logistics and warehousing land	0.56	0.56	1	1	0.55	0.55
M	Industrial land	1.23	10.70	2	15	1.20	10.51
G	Green space and square land	43.91	11.43	62	16	43.13	11.22
B	Land for commercial service facilities	7.42	7.42	10	10	7.29	7.29
R	Residential land	13.38	23.41	19	33	13.15	23
S	Roads and transportation land	0.46	12.21	1	17	0.46	12
H	Urban construction land	71.32	71.32	100	100	70.06	70.06

4.2 Urban Road Analysis

According to the road classification standards of the "Design Specification for Urban Road Engineering (CJJ37-2012)" standard, the road classification map of Luohu District in Shenzhen (Figure 4) and the road classification map at all levels of Luohu District in Shenzhen (Figure 5) are classified. Expressways can be roughly divided into "four vertical and two horizontal," with the main road mainly concentrated in the urban center and extending to the eastern and western mountainous areas, and the branch road connecting the traffic of the urban center and the eastern and western mountainous areas. The secondary road connects urban buildings and residential buildings.

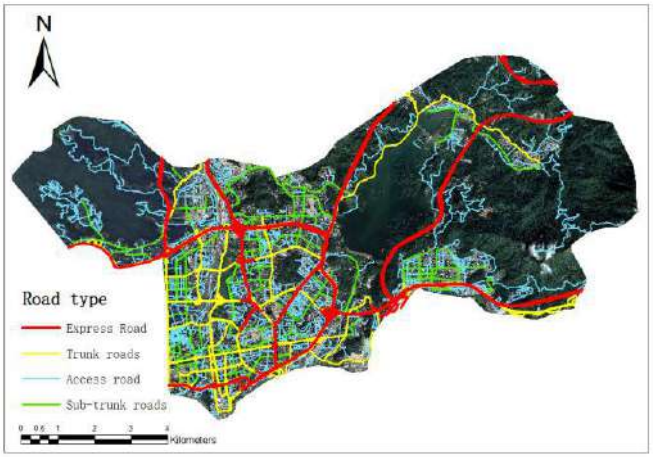

Fig.4 Road Classification Map of Luohu District, Shenzhen

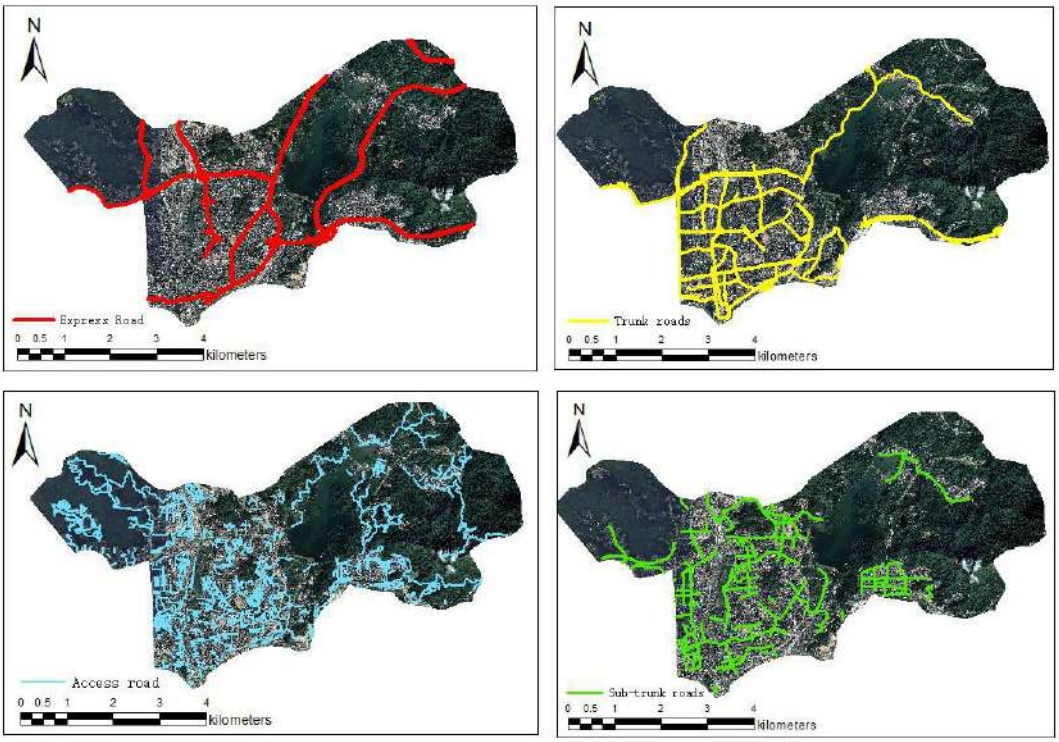

Fig.5 Classification map of various roads in Luohu District, Shenzhen

4.2.1 Analysis of Vertical Section Slope of Urban Main Roads

According to the distribution of main roads obtained from Figures 4 and 5, as well as the limit values formulated based on vehicle power characteristics and considering economic factors, they should not be easily adopted in the design, leaving room for error. Generally speaking, it is better to have a gentle longitudinal slope, but for the drainage of the road surface and side ditches, the minimum longitudinal slope should not be less than 0.3% to 0.5% (Table 13) (Liu, 2011).

Table 13 Main Technical Indicators of Chinese Highways

Highway grade	Expressway		一		二		三		四	
terrain	Plain hills	Mountains and heavy hills	Plain hills	Mountains and heavy hills	Plain hills	Mountains and heavy hills	Plain hills	Mountains and heavy hills	Plain hills	Mountains and heavy hills
Calculate driving speed (km/h)	120	80	100	60	80	40	60	30	40	20
Maximum longitudinal slope (%)	3	5	4	6	5	7	6	8	6	9

Usually, the design of longitudinal section alignment follows the following principles (Ma, 2009):

1) The vertical section design should refer to urban road planning to control the elevation and adapt to the layout of

building facades along the street and the drainage of surface water along the road range;

2) To ensure safe and comfortable driving, the longitudinal slope should be gentle and smooth, and the undulations should not be frequent.

3) The design of longitudinal sections of mountain roads and newly opened roads should take into account the balance of earthwork and the economic effects of automobiles. Reasonably arrange the road surface design elevation;

4) The lane where motor vehicles and non-motor vehicles are mixed should be designed with a longitudinal slope based on the climbing ability of non-motor vehicles;

5) The longitudinal section design should comprehensively address the requirements of terrain, underground pipelines, geology, hydrology, climate, and drainage.

By analyzing the longitudinal slope of the main road in Luohu District, it is found that all of them meet the technical indicators of Chinese highways and the roads are smooth (Table 13).

Table 13 Longitudinal Section Slope of Main Roads in Luohu District

Road name	Road length（m）	Profile grade（%）	Whether it complies with national standards（≤5%)
Honghu West Road	1900	0.63%	YES
Ja bin Road 1	3400	1.41%	YES
Honggui Road1	1700	1.06%	YES
Beihuan Avenue 1	2400	1.38%	YES
Riverside Drive 1	1500	1.73%	YES
Heping Road	4400	0.86%	YES
Honggang Road 1	3900	0.90%	YES
Honggang Road 2	4000	1.03%	YES
Honggang Road 1	1800	1.06%	YES
Honggui Road 2	1700	1.06%	YES
Hongling North Road 2	342100	0.05%	YES
Ja bin Road 2	4800	1.44%	YES
Jianshe Road 1	4000	1.85%	YES
Jiaohu Road 1	1800	1.50%	YES
Jiaohu Road 2	1800	1.39%	YES
Nigang East Road 1	3600	0.53%	YES
Nigang East Road 2	7100	0.76%	YES
Qingping Expressway 1	1800	0.89%	YES
Qingping Expressway 2	1600	0.88%	YES
People's Park Road 1	2800	1.25%	YES
People's Park Road 2	2800	1.36%	YES
Drying Cloth Road 1	800	4.13%	YES
Drying Cloth Road 2	800	4.13%	YES
Shennan East Road 1	9500	0.46%	YES

Shennan East Road 2	9000	0.67%	YES
Sungang Road 1	5600	0.66%	YES
Sungang Road 2	5500	0.60%	YES
Xinxiu Road	1800	1.94%	YES
Xinyuan Road	1800	1.78%	YES

4.2.2 Analysis of urban road area

The urban road area ratio, also known as the "urban road area density," is expressed as the percentage of the area of roads within the urban built-up area (roads refer to roads with a paved width of 3.5 meters or more, excluding sidewalks) to the built-up area. The formula is: road area ratio=total area of road land/total area of construction land. The urban road area ratio is an important economic and technical indicator that reflects the ownership of urban roads within the urban built-up area. According to calculations, the built-up area of Luohu District is 71320591 square meters. Therefore, the road area ratio of Luohu District is shown in Table 14:

Table 14 Road Area Ratio in Luohu District

Road category	Length（m）	Road area（㎡）	Urban road area ratio（%）
Expressway	123941.76	4337961.72	6.08
Main road	149252.53	4477575.76	6.28
Secondary trunk road	110780.04	2880280.95	4.04
by-pass	243602.13	3897634.08	5.46

The area ratio of urban roads refers to the proportion of the land area of roads to the urban construction land area. According to the "Code for Urban Road Traffic Planning and Design" (GB50220-95), the land area of urban roads should account for 8–15% of the urban construction land area. For large cities with a planned population of over 2 million, it is recommended to be 15%–20%. The population of Luohu District in 2022 will be 1.018 million people, and the total area ratio of urban roads will be 21.87%, which is consistent with the "Code for Urban Road Traffic Planning and Design." According to the regulations (GB50220-95), the road area in Luohu District is 6.87% higher than the urban construction land area.

Therefore, the proportion of the road area in Luohu District to the urban construction land area is too high. The urban road area ratio is an important economic and technical indicator that reflects the ownership of urban roads in the urban built-up area. A high road area ratio in Luohu District means an increase in road investment. With relatively more roads, traffic congestion will naturally decrease. However, a high road area ratio is not conducive to the construction of the urban environment. A high road area ratio can cause an increase in intersections, potentially leading to traffic congestion. If the road area is too high, the urban green area will decrease. It also means that the already tense residential activity land is further squeezed, which is not conducive to the efficient development of land resources.

4.2.3 Analysis of Road Intersection Spacing

According to the industry standard "Design Regulations for Urban Road Intersections (CJJ 152-2010)" issued by the Ministry of Housing and Urban Rural Development of China, the spacing between level crossings should be determined based on the size of the city, road network planning, road types, and their regional location in the city; the spacing between main road intersections should be roughly equal; the minimum spacing between various types of intersections should meet the minimum length required for turning vehicles to

change lanes, the maximum queue length for vehicles during the red light period, and the total length of the entrance and exit lanes, and should not be less than 150m.

According to the national standard "Design Specification for Urban Road Traffic Facilities (GB 50688-2011)" issued by the Ministry of Housing and Urban Rural Development of China, pedestrian crossing facilities should be installed at road intersections. The distance between pedestrian crossing facilities on expressways and main roads should be 300–500 m, and the distance between pedestrian crossing facilities on secondary roads should be 150–300 m.

The spacing data of major road intersections in Luohu District, Shenzhen (Table 15) is based on the table data, which shows that the spacing of all intersections is greater than 150m, in accordance with the national standards.

The main road, Shennan Middle Road, has six intersections and three sections, among which only the distance from the intersection of Bao'an South Road to the intersection of Heping Road meets the national requirement of 300–500 meters. The distance from the intersection of Jianshe Road to the intersection of Dongmen Middle Road and from the intersection of Hongling Middle Road to the intersection of Bao'an South Road has reached over 600 meters, exceeding the national requirement of 300–500 meters.

There are a total of five intersections and three sections on the two secondary roads. Among them, only the distance between the intersection of Bao'an South Road and Guiyuan Road meets the national requirement of 150–300 m. The distance between the intersection of Caiwuwei Third Street and Bao'an South Road, as well as the intersection of Leyuan Road and Wenjin Middle Road, has reached over 300m, exceeding the national requirement of 150–300 m.

Table 15 Distance between Main Road Intersections in Luohu District, Shenzhen

Road category	Road name	Starting point	Termination	Road intersection spacing（km)	Average intersection spacing of roads（Km)
Expressway	Yanhe South Road	Wenjin South Road	Shennan East Road	1.163	0.622
	Yanhe Road Viaduct	Dongmen South Road	Wenjin South Road	0.311	
	Yanhe Road Viaduct	East Ring Expressway Viaduct	Dongmen South Road	0.392	
Main road	Shennan East Road	Jianshe Road	Dongmen Middle Road	0.666	0.543
	Shennan East Road	Bao'an South Road	Heping Road	0.326	
	Shennan East Road	Hongling Middle Road	Bao'an South Road	0.637	
Secondary trunk road	Jiefang Road	Cai Wuwei Third Street	Bao'an South Road	0.488	0.33
	Jiefang Road	Bao'an South Road	Guiyuan Road	0.203	
	Hubei Road	Leyuan Road	Wenjin Middle Road	0.307	
by-pass	Qiaoxing Road	Luofang Road	Jing'er Road	0.254	0.21
	Feipeng Road	Hubei Road	Shennan East Road	0.132	

	Antarctic Road	Chunfeng Road	Shennan East Road	0.236	

4.3 Public Transport Network

4.3.1 Analyze Road Network Density

According to the "Code for Urban Road Traffic Planning and Design" (GB 50220-95) in China, the standard density of the urban road network is 3–4 km/km2 in the central area and 2–2.5 km/km^2 in the urban edge. Luohu District includes 10 streets, all of which belong to the main urban area. The built-up area of Luohu District is 71.32 km^2, and the length of the research bus route is 170.46km. The calculated density of the bus route network in the main urban area of Luohu District is 2.39 km/ km^2. It does not meet the design standards for the density of bus routes in the central urban area, and there are still some areas with bus gaps, which affect the normal travel of local residents. The low density of the urban public transportation network can lead to a decrease in the scope of public transportation services and an increase in passenger walking distance, which is not conducive to passenger travel.

The distribution characteristics of the density of public transportation routes in Luohu District are: The density of public transportation routes in Luohu District is unevenly distributed in space. The density of public transportation routes in the central and southern regions is relatively dense, with a maximum value of 3.5 km/ km^2. The density of public transportation routes in this area meets the requirements of the density standard for urban center areas, making it convenient for residents to travel in this area. The public transportation network in the northeast and northwest regions is relatively sparse, with a maximum value of only 0.6 km/ km^2. The density of the public transportation network in this area has not met the requirements of the density standard of the urban center area, and the travel of residents is not convenient enough.

4.3.2 Analyze the Length of the Road Network

The appropriate length of a bus route depends on the average distance traveled by passengers on that route. If the route is too long, it will reduce the turnover rate of the bus, increase the waiting time for passengers, and increase the probability of vehicle delay, while also increasing the operating cost of the bus route, reducing the transportation efficiency and economic benefits of the bus route. If the route is too short, it will reduce the utilization rate of buses, increase the number of transfers for residents to take public transportation, and, to some extent, increase the pressure on urban transportation.

According to the "Code for Urban Road Traffic Planning and Design (GB50220-95)," the length of bus routes should not exceed the maximum limit: the one-way length of general urban bus routes is 8–12 kilometers, not exceeding 13 kilometers, and the length of the public transportation network in Luohu District is 170457.21 meters.

4.3.3 Analyze the Non-linearity Coefficient of the Road Network

56% of the bus routes in Luohu District meet the design specifications, and the non-linear coefficient values of the routes are higher than the design specifications. This is mainly due to the influence of the urban spatial structure and natural conditions in Luohu District.

From the non-linear coefficient characteristics of each route, the non-linear coefficient of suburban lines is smaller than that of urban lines. Among the 56 bus routes that meet the design specifications and standards, only 18 urban lines account for 32.1%. The suburban lines are relatively straight and have small detours, while the urban lines are influenced by buildings and have more detours, resulting in a relatively high non-linear coefficient. Among them, M485, M140, B909, E13, B621, 333, 337, E13, M103, M482, 62, peak lines 33, 29, M482, etc. are particularly obvious and even affect the travel time of passengers. The route direction should be adjusted appropriately to reduce detours.

4.3.4 Analyze the Road Network Repetition Coefficient

The repetition coefficient of the public transportation network reflects the degree of waste in the capacity of public transportation routes. In the "Code for Urban Road Planning and Design (GB50220-95)," only the definition is given: the repetition coefficient of public transportation lines refers to the ratio of the total length of public

transportation lines to the length of the regional road network, without specifying specific regulatory requirements. The coefficient of route repetition is also the ratio of route density to network density, where route density refers to the ratio of the total length of bus operating routes to the land area of cities with bus services, and network density refers to the ratio of the road mileage of routes arranged in the area to the land area of cities with bus services. For a given section, its value is the number of lines passing through that section. According to the Transportation Engineering Manual, it is recommended to have a network repetition coefficient of 1.25–2.5.

The repetition coefficient of Luohu District is about 1.52 (as shown in Table 16), which complies with the provisions of the Traffic Engineering Manual.

Table 16 Repetitive coefficient of bus routes

Road name	Length（km）	Repetition coefficient
Bus routes in Luohu District	170457.21	1.52
Luohu District Road Network	259404.54	

4.3.5 Analyze Average Station Distance

According to the requirements of the Urban Road Traffic Planning and Design Standard (GB-50220-95), the average station distance in the urban area should not exceed 400m, and the average station distance in the suburbs should not exceed 800m.

The average distance between bus stops in Luohu District, Shenzhen, is 168.67m, which is much lower than the standard and meets the standards (Table 17). Among the 361 intervals, only 15 intervals in the above table have a distance greater than 400m, indicating that the infrastructure level in Luohu District is relatively good.

Table 17 Average Bus Station Distance

Starting station	Arrival station	Distance
Tongxin Garden	Changling East Bus Station	862.82
Sanjiu Garden	Sanjiu Road Intersection	555.18
Qinghu Villa	Qingshuihe Logistics Park	476.41
Luofang Village Archway	Anfang Primary School	470.81
Anfang Primary School	Luofang Village Archway	470.81
Luofang Sancha River	Luofang Water Purification Plant	462.41
Wutongshan Bus Station	Tiger Bamboo Scare	454.66
San Tin Village	Lanke Center	451.34
Taojinshan Greenway South Station	Taojinshan Community	425.48
Taojinshan Community	Taojinshan Greenway South Station	425.48
Luofang Water Purification Plant	Luofang South Road	424.03
Cuirong Garden	Xianhu Maple Scenery	417.41
Sungang Railway Station	Tianbei New Village	416.97
Honghu West Road North	Honghu Park 2	414.41

4.3.6 Analyze Station Coverage

According to the requirements of the Urban Road Traffic Planning and Design Standard (GB-50220-95), the service area of public transportation stations, calculated with a radius of 300m, shall not be less than 50% of the urban land area; Calculated with a radius of 500m, it should not be less than 90% (Figure 6):

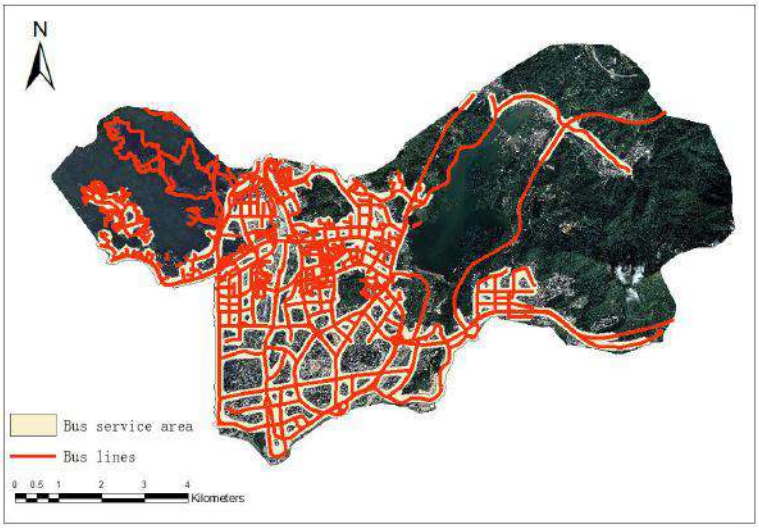

Fig.6 800m Bus Service Area in Luohu District

The data on the coverage rate of bus stops in Luohu District, Shenzhen, shows that the urban bus coverage rate within 500m is only 34% (Table 18), far below the standard value.

Table 18 Bus Station Coverage

Number of bus stops	Bus stop 500m service area (square kilometers)	Built-up Area	Bus stop coverage rate (%)
360	24.83	71.32	34.82

V. CONCLUSION

This article uses GIS to conduct spatial analysis on land use data, road data, and public transportation data in the built-up area of Luohu District, Shenzhen. Combined with national standards, the following conclusions are drawn:

1. Analysis of Land Use in Luohu District

From the perspective of per capita construction land indicators, Luohu District in Shenzhen belongs to a Class IV climate zone, with a per capita urban construction land area ranging from 65.0 to 110.0 square meters and a per capita urban construction land area of 70.1 square meters per person in Luohu District. It meets the scope of this standard. From the perspective of per capita individual construction land, the land for green spaces and squares is 43 square meters per person, which meets the national standard requirements; the road and transportation land is 0.5 square meters per person, which does not meet the requirements; and the residential land is 13 square meters per person, which does not meet the requirements. From the perspective of land structure proportion, residential land accounts for 19%, which does not meet the range of 25.0–40.0% in national standards, while public management and public service land account for 6%, which meets national standards. The proportion of industrial land is 2%, which does not meet national standards. 1% of the land used for roads and transportation facilities does not meet national standards. The proportion of green space and square land is 62%, which does not

meet national standards. Overall, the per capita construction land in Luohu District meets the standards, and the ratio of per capita individual construction land to land structure does not meet the standards (Table 19).

Table 19 Analysis of Land Use condition

Index	Types	Present situation	Standard	Evaluate whether it meets the requirements
Per capita construction land	Per capita construction land area	70.1 ㎡/person	[65.0,110.0]㎡/ person	yes
Per capita single construction land	Green space and square land	43 ㎡/ person	≥10.0m2/ person	yes
	Roads and transportation land	0.5 ㎡/ person	≥12.0 m2/ person	no
	Residential land	13 ㎡/ person	≥23.0m2/ person	no
Land use structure ratio	Green space and square land	62%	[10.0%,15.0%]	no
	Roads and transportation land	1%	[10.0%,25.0%]	no
	Residential land	19%	[25.0%,40.0%]	no
	Public management and public service land	6%	[5.0%,8.0%]	yes
	Industrial land	2%	[15%,30%]	no

2. Urban Road Analysis

The maximum longitudinal slope of the main road in Luohu District meets the technical indicators of Chinese highways, and the road is gentle. Due to the population of 1.018 million people in Luohu District in 2022, the total urban road area ratio is 21.87%, which is 6.87% higher than the standard. Therefore, the proportion of road area in Luohu District to the urban construction land area is too high and does not meet the standard. The spacing between the intersections of expressways and branch roads in Luohu District complies with the Design Specification for Urban Road Traffic Facilities (GB 50688-2011), while the main and secondary roads do not comply. Therefore, excessive road area will affect the land occupation of other types of land, and excessive spacing between intersections of main and secondary roads will affect the convenience of transportation. Overall, the roads in Luohu District still need to be adjusted (Table 20).

Table 20 Analysis of Land Use condition

Index	Types	Present situation	Standard	Evaluate whether it meets the requirements
Maximum slope of vertical section	Main road	[0.05%,4.13%]	≦5%	yes
Road area	Four levels of roads	21.87%	[8%,15%]	no
Road intersection spacing	Expressway	622m	≥150m	yes
	Main road	543m	[300m,500m]	no
	secondary trunk road	330m	[150m,300m]	no
	by-pass	210m	≥150m	yes

3. Analysis of Urban Bus Routes and Stops

The density of bus routes in the main urban area of Luohu District is 2.39 km/ km2, which does not meet the design standards for bus route density in the central urban area. Some areas still have bus blank areas, which affects the normal travel of local residents. The average length of the road network is 7.51km, which meets the standard of [8, 13] km. The non-linear coefficient is 1.62, which does not meet the standards of the design specifications. The repetition coefficient of Luohu District is 1.52, which meets the standard. The average distance between bus stops is 168.67m, which meets the standards. The coverage rate of urban public transportation within 500m is only 34%, which meets the standard. Therefore, the overall situation of bus routes in Luohu District is unreasonable, reducing the utilization rate of buses and increasing the number of transfers for residents to travel by bus, which to some extent increases the pressure on urban transportation (Table 21).

Table 21 Analysis of urban bus routes and stops condition

Index	Present situation	Standard	Evaluate whether it meets the requirements
Road network density	2.39 km/km2	[3,4] km/km2	no
Road network length	7.51km	[8,13] km	yes
Non-linear coefficient	1.62	≦1.4	no
Repetition coefficient	1.52	[1.25,2.5]	yes
Average stop spacing	168.67m	≦400m	no
Site coverage	34%	≥90%	no

In summary, based on the evaluation of land use, road system, and public transportation routes in Luohu District, it is recommended to actively implement the "Management Regulations on Supporting the Development of the Real Economy and Promoting the Conservation and Intensive Use of Industrial Land in Shenzhen" in future development, further promote the structural reform of the land supply side, better utilize existing land resources, and promote the conservation and intensive use of industrial land; to build more roads and expand the road area; and properly convert the green area into roads and industrial land.

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Antibacterial Activity of Silver Nanoparticles Synthesized from *Aloe Vera* Extract

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Abstract**—** *Material science has advanced significantly due to nanotechnology, which enables researchers to work with materials at the nanoscale to produce novel goods. It has been suggested that using plant extract nanoparticles biosynthesis is an economical, and environmentally benign process. Despite various chemical and physical approaches, green nanomaterial synthesis is the most contemporary. We used the Aloe vera plant to synthesize silver nanoparticles (AgNPs) and tested their antibacterial characteristics. UV-Vis spectroscopy, Scanning Electron Microscope (SEM), and X-ray diffraction were used to analyze the produced AgNPs. The XRD peaks, which were indexed, represented the face-centered cubic configuration of AgNp's. The exhibited peak indicates nanocrystalline-structured silver particles. The average particle size of bio-synthesized NPs which was between 30 and 35 nm is confirmed by SEM. The antimicrobial activity of our synthesized Nps was tested against bacteria (Escherichia. E. coli, Pseudomonas luteola, Bacillus Subtillis) as a function of nanoparticle concentration and the test was done by Disc diffusion method and antimicrobial activity was found to be quite robust. When the outcomes were compared to the effect of antibiotics like Ciprofloxacin 5, Vancomycin 30, and Ampicillin 10, these antibiotics were shown to be less effective than nanoparticles*

Keywords**— Aloe Vera, Biosynthesis, Nanotechnology, Nanoparticles (NPs) Silver Nanoparticle (AgNPs).**

I. INTRODUCTION

The 'nano' is a Greek prefix that indicates 'dwarf' or minuscule and indicates 1 billionth of a meter (10−9 m) [1]. the study of structures and substances on a nanometer scale is known as Nanoscience. nanotechnology is a technology used in practical applications like electronic gadgets. Nanoscience dates back to the 5th century BC Greek and Democritus eras [2]. From the huge molecule to the small solid item to be a strong link between surface and volume, the nanoworld is the gateway between the atom and the solid. The nanoworld concept is based on the coming together of a diverse range of scientific and technological fields that were distinct earlier [3].

Nanotechnology is the study of the very small; it is the small-scale manipulation of material. Atoms and molecules have various functions at this scale and have various remarkable and intriguing uses. It provides opportunities for the development of materials, including medical applications [4].

In current material science, nanotechnology has been the most actively studied topic. Nanoparticles show entirely new and innovative properties based on certain qualities such as dimension, shape, and distribution [5, 6].

NPs differ in shape, size, and structure. It's between 1 and 100 nanometres and its shapes include round, hollow, spiral, flat, and uneven in shape with a tabular, conical hollow

center. Surface variation might be uniform and uneven. Certain NPs are crystal solids that maybe loosely or densely packed and they are crystalline or amorphous. [7].

CLASSIFICATION OF NANOPARTICLES:

Carbon-based, inorganic, and organic NPs are the 3 subclasses of NPs.

ORGANIC NANOPARTICLES:

Organic NPs include dendrimers, micelles, liposomes, and ferritin, among others [8]. In addition to their typical traits including size, composition, surface morphology such as, and so on, their unique characteristics make them ideal organic nanoparticles, medication delivery systems may also employ organic NPs[9]. These NPs are environment-friendly. [10]. The medication carrying capacity, stability, and distribution mechanisms, such as liposomes, have a vacuous core and are delicate to temperature and targeted heat.

INORGANIC NANOPARTICLES:

Inorganic NPs didn't contain carbon. Inorganic NPs are not harmful. Biocompatible and hydrophilic inorganic nanoparticles, contrastingly with organic nanoparticles [11], inorganic NPs are extremely stable. Inorganic NPs are categorized into Metal and metal oxide NPs.

METAL-BASED NANOPARTICLES:

Metals are converted into nanometric-sized particles by constructive destructive methods [12]. Almost any metals can be converted into Nanoparticles [13]. Aluminum (Al), cadmium (Cd), cobalt (Co), copper (Cu), gold (Au), iron (Fe), lead (Pb), silver (Ag), and zinc (Zn) theses metal are commonly used in the synthesis NPs [14]. Aside from their sizes, which range from 10-100nanometer. [7].

METAL OXIDE-BASED NANOPARTICLES:

The goal of metal oxide NPs is to correspond with relvant metals. [15]. Iron nanoparticles, for example, are oxidized to iron oxide nanoparticles. Iron oxide nanoparticles have a higher degree of reactivity than iron NPS. An improvement in efficiency and reactivity results in the generation of metal oxide NPs [16]. Zinc oxide, silicon dioxide, iron oxide, aluminum oxide, cerium oxide, titanium oxide, and magnetite are examples of metal oxide nanoparticles [7].

CARBON-BASED NANOPARTICLES:

Carbon-based nanomaterial is made entirely of carbon. Carbon-based NPs include fullerenes, graphene carbon nanotubes (CNT), carbon black, and carbon nanofibers.

METHODS FOR SYNTHESIS OF NANOPARTICLES:

Different techniques including chemical, physical, and biological techniques are used to create nanoparticles.

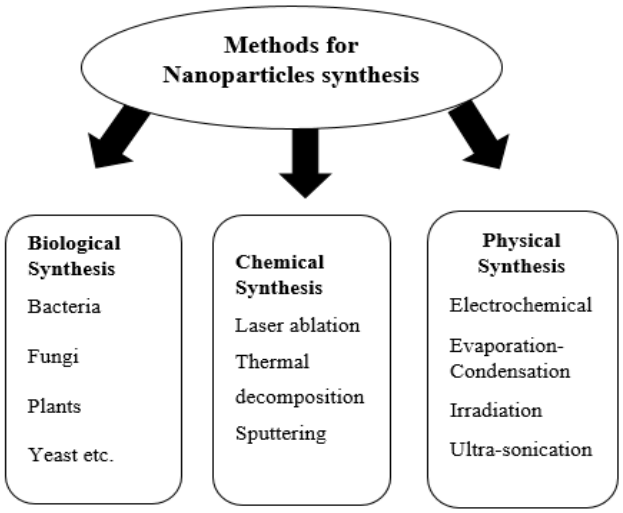

Fig.1. Methods for Synthesis of Nanoparticles

PHYSICAL METHOD:

physical methods used to make nanoparticles are UV irradiation, sonochemistry, laser ablation, and radiolysis. during the physical synthesis process, metal atoms evaporate. Immediately after this, condensation on various supports, where the metallic atoms rearrange as tiny collection of metals nanoparticles. using physical methods we may create NPs that are extremely pure and have a certain form [17]. Evaporation/condensation has previously been used to make nanoparticles of several materials such as Ag, Au, and fullerene using this physical technique [7]. Conversely, these techniques usually need highly advanced instruments, chemicals, and radiant heating, along with significant power consumption, leading to elevated operational expenses.[17].

CHEMICAL METHOD:

Mechanical milling, laser ablation, thermal decomposition, sputtering, etc. [17] are the chemical techniques for the synthesis of nanoparticles but demands a short period in order to produced enormous amount of NPs. The majority of chemicals employed in nanoparticle synthesis and stabilization are harmful and produce non environment-friendly wastes [5].

BIOLOGICAL METHOD:

There is a current demand to design environment-friendly synthesis techniques that do not have harmful compounds and have advantages over traditional processes that use harmful chemicals. To synthesize nanoparticles, biosynthesis employs bacteria, plant extracts, fungi, and other microbes, as well as precursors [18]. The need for eco-

friendly NP synthetic procedures. Synthesis has led to an increasing interest in biological methods without using hazardous chemicals as byproducts and green nanotechnology has emerged. Because they don't contain hazardous compounds and offer natural capping agents, plants make a superior starting point for the creation of nanoparticles.[19].

Several plant extract has been used to accomplish the Green synthesis of NPs we use *Aloe vera* in this study.

ALOE VERA:

The succulent plant *Aloe vera* belongs to the Aloe genus [20]. There are roughly 500 species and many places in the world, and consider them invasive. The botanical names *aloe* (also from Greek) and *Vera* ("true") are both Latin. *Aloe* is commonly known by its common names which include *Chinese aloe*, *Cape aloe*, or *Barbados aloe.* Nicolass Laurens Burman described it in 1768 as *Aloe vera* in *Flora*. [21]. Aloe species are succulent perennial xerophytes with thick fleshy leaves that allow water to be stored in the well-known gel [22]. Aloe species thrive in semi-arid, warm conditions and best grow on dry, sandy, calcareous soil. Aloe, on the other hand, is not a cactus. Africa the Arab peninsula, Madagascar, and all the Indian Ocean islands are home to the Aloeis. Some of their species can be also found in the Asia California and in the United States [23].

Because of its plethora of biomolecules, *Aloe vera* is a fleshy plant that has therapeutic benefits [24][25]. The researchers were successful in synthesizing nanoparticles using microorganisms and extracts of plant-specific items such as *Aloe vera gel* to make silver and gold nanoparticles [26]. The nanoparticles were embedded into cotton fabric and screened for their antimicrobrial properties against *Escherichia coli* and *Staphylococcus aureus* [19]. Because of its antimicrobial, antioxidant, anti-inflammatory, and other functional qualities, *Aloe vera* is one of the oldest therapies for a variety of human maladies, as a result, it is found in favor in disciplines including, preserving food, sustainable packaging cosmetics, and pharmaceuticals [27].

SILVER NANOPARTICLES:

Several new products are using silver nanoparticles to generate antimicrobial action due to a large surface area [31]. Because of its dichroic property when integrated into glass, silver nanoparticles were of long-term interest for centuries. Because of their biocidal qualities, silver-based substances have been utilized as harmless, inorganic, and antimicrobial agents for ages in a variety of applications including wood preservatives and water purification [32]. AgNps are widely employed characteristic in catalysis, chemical sensing, biosensing, photonics, electronics, and pharmaceuticals due to their unique properties [33]. Antimicrobial activity is another application of AgNps. With a low toxicity profile, silver is a powerful antimicrobial. Silver and silver nanoparticles are most commonly used in scientific applications, such as topical ointments to prevent burns and open wound infections [34]. Owing to their intriguing physicochemical properties, biology, and medicine both greatly benefit from the use of AgNp [35]. Because of its remarkable aspects, researchers are now concentrating on metal nanoparticles, and nanostructure nanomaterial production [36]. The AgNps has piqued the curiosity of the scientific community due to their vast range of applications. Additionally, These AgNps are being used to effectively identify and treat cancer [37].

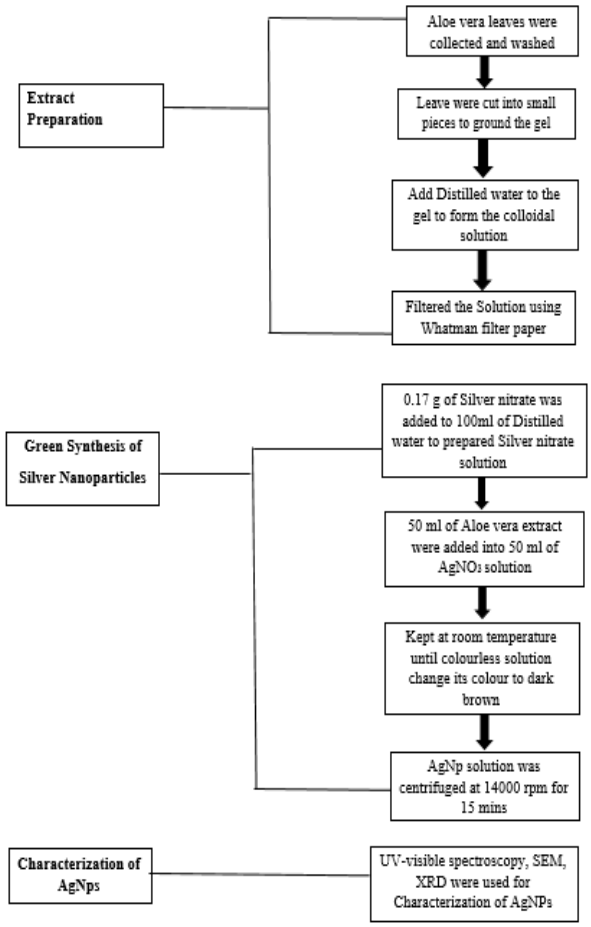

Fig 2. Schematic Diagram of Synthesis of Silver Nitrate

SAMPLE COLLECTION:

We harvested fresh *Aloe vera* leaves from the garden of Women University Mardan and *Escherichia coli, Pseudomonas luteola, and Bacillus Subtillis* were provided by the Central Lab of Women University Mardan.

SILVER NANOPARTICLES SYNTHESIS:

***ALOE VERA* EXTRACT PREPARATION:**

Fresh *Aloe vera* leaves were collected and were washed two to three times with tap water to eliminate any dirt and dust, after surface sterilization the green peel was removed, and the leaves were chopped into minute pieces, a gel emerged. the gel was then blended to produce an aqueous solution which was sieved through Filter paper and the filtered leaf extract was then placed at room temperature.

PREPARATION OF SILVER NITRATE SOLUTION:

To make the solutin of 1mM Silver Nitrate, add 0.17 grams of silver nitrate in 100ml of distilled water. After stirring for a few minutes, the silver nitrate solution was prepared and covered with Aluminum foil.

SYNTHESIS OF SILVER NANOPARTICLES FROM *ALOE VERA* EXTRACT:

In a reaction process, both the AgNO3 solution and extract were taken in 1:1 concentration. To alter the pH of the medium 50ml of 1mM $AgNO_3$ solution was added into a flask having 50ml of plant extract and dissolved with 1% ammonia solution. The prepared solution was kept at room temperature the synthesis of spherical-shaped AgNP was indicated by the appearance of orange-yellow coloration. The solution was centrifuged and the Suprenant was eliminated from the prepared solution and the pellet was rinsed in ethanol and placed directly on a glass coverslip for air drying.

II. CHARACTERIZATION

UV-VISIBLE SPECTROSCOPY:

The AgNp's were studied using a UV-visible spectrophotometer (Uviline Connect Series 940) to compare the synthesis and kinetic properties of our designed nanoparticles. The spectrometer was equipped with "Spectra lab" software to collect and examine the results. The formation and stability of AgNp were seen in wavelengths between 300-550 nm. UV-Vis absorption spectra recorded the numerical data were plotted as a graph.

X-RAY DIFFRACTION METHOD (XRD):

The developed AgNp solution was centrifuged for 30 min at 14000 rpm. The Suprenant was removed with the help of a disposable syringe and the pellet was washed with ethanol and placed for air drying to get powder AgNps for XRD. X-ray diffraction (JDX 3532 Jeol Japan) Cu K radiation is used for the characterization of synthesized nanoparticles. The samples were imaged with the generator's current and voltage parameters set at 35 kilovolts and 25 milliamperes, and they scanned in the 15 to 700 C range. 0.04/sec was the scan rate.

SCANNING ELECTRON MICROSCOPE (SEM):

The JSM-5910 Jeol Japan SEM machine was utilized to characterize the mean particle size and shape of our synthesized nanoparticles. After sonicating the AgNP solution with distilled water, tiny droplets of these samples were applied on a slide and waited for it to dry. A tiny plated platinum layer was added to the samples to make them conductible. A vacuum of 10-5 torr was applied to the JSM-5910 Jeol Japan SEM machine. The microscope's accelerating voltage was maintained between 10 and 20 kV. The SEM micrograph gives the shape and size of our synthesized AgNPs.

ANTIBACTERIAL ACTIVITY:

Different bacterial strains including *Bacillus Subtillis, Escherichia coli, and Pseudomonas Luteola* were utilized antibacterial activity of synthestic ANPs was determined using Well Diffusion technique.

SUB-CULTURING:

To obtain the pure colonies, the bacterial strains were grown on nutrient agar and incubated at 37˚ Celsius for 24 hrs. Using the streaking technique, the bacteria were cultured on nutrient agar and incubated the cultures at 37˚ Celsius for 24 hrs. Inoculate the bacterial strain's single colony and keep it at 37˚ Celsius in the incubator for 24 hours.

WELL DIFFUSION METHOD:

MHA media was ready to use for the Well Diffusion technique to evaluated the produced silver nanoparticles' antimicrobial activity against bacterial species. The glass plates were filled with MHA medium and the inoculated bacterial strain using a disinfected cotton bud the holes were made with the assistance of micropipette blue tips and wells were filled with silver nanoparticle solution at different concentrations of 40, 60, and 80 µl/ml and incubated for 24hrs at 37°C. The antimicrobial action was determined by examining the region of embarrassment (mm) that was developed all around the discs.

III. RESULTS

The alteration in hue of silver nanoparticles is due to surface plasmon vibration. A visible color shift in the reaction medium was used to initially identify the silver nanoparticles. Over time, the intensity of the color grew to turn from orange-yellow to dark brown.

UV-VISIBLE SPECTRAL ANALYSIS:

The appearance of brown color is due to surface plasmon vibration. A surface plasmon absorption band with a maximal of 350 nano-meters was seen in brown. This

indicates the AgNp absorbance at 350 nm-53 0nm as shown in Figure 5 and their structure was confirmed by SEM images.

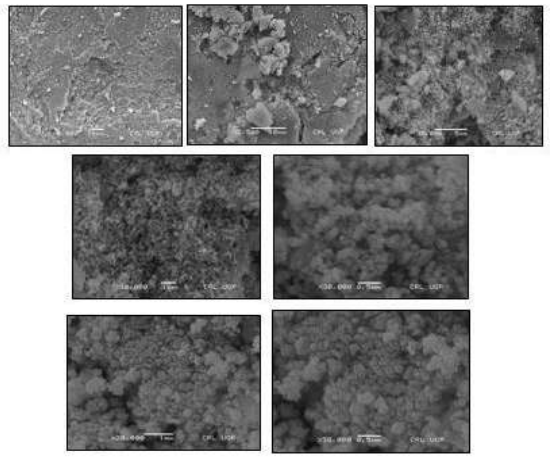

Fig 3. SEM images of nanoparticles at various magnifications

X-RAY DIFFRACTION ANALYSIS:

X-RD is utilized to examine the particle's overall oxidative condition throughout period, i.e., phase identification and depiction of the nanoparticles' crystal structure. XRD investigations were used to perform structural analysis. Figure 10 shows the XRD form of *Aloe vera* plant extract in the manifestation of AgNP. The face-centered cubic pattern of AgNP was represented by the XRD peaks, which were indexed. The nanocrystalline-shaped silver particles are indicated by the depicted peak.

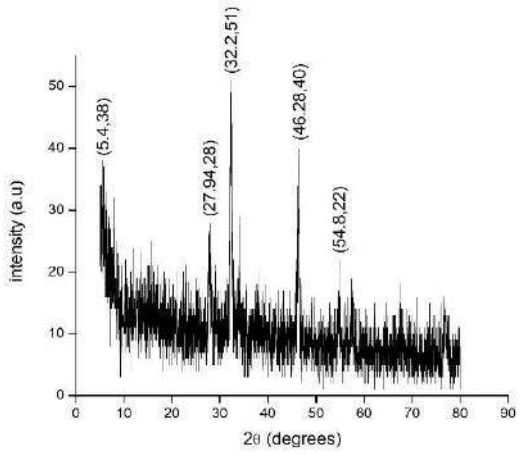

Fig.4. XRD Graph Of Silver Nanoparticles

4.4 ANTIBACTERIAL ACTIVITY OF THE AgNPs:

The antimicrobial characteristics of the AgNPs were concluded by monitoring the inhibition zones surrounding wells at various volumes of 40, 60, and 80 µl/ml, following 24 hrs of the incubation period at 37°C. The result revealed that sliver nanoparticles had an inhibition zone against *Escherichia. Coli, Pseudomonas luteola, Bacillus Subtilis*.The antibacterial activity enhanced as the concentration of AgNps was increased. Figure 11 shows Ag nanoparticles were used to create a clear zone of inhibition. *Bacillus Subtilis* relative to the most significant zone of inhibition as compared to other bacterial strains.

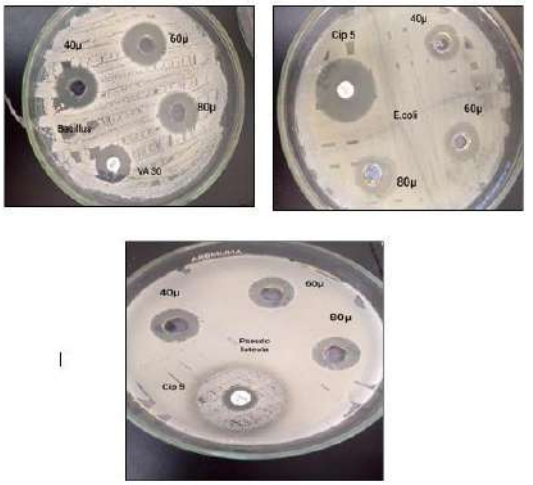

Fig 5. Antibacterial Activity of Nanoparticles with Different

Concentrations Ranging From 40 µL, 60 µL, and 80 µL

Table 1. The antimicrobial activity of silver nanoparticle synthesized using extract of Aloe vera

Bioactive agent		Inhibition Zone		
		Escherichia coli	Bacillus subtilis	Pseudomona luteola
Ag nanoparticles	40µl	13	18	12
	60µl	15	22	14
	80µl	16	21	16
Ciprofloxacin 5		22	nil	10
Vancomycin 30		Nil	17	Nil

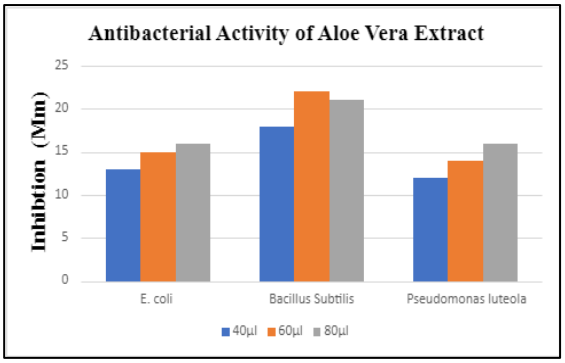

Fig 6. Antibacterial Activity of Nanoparticles with different

concentrations ranging from 40 µl, 60 µl, and 80 µl

IV. DISCUSSION

When AgNO3 solution was added to the *Aloe vera* extract the color was slightly changed into an orange-yellow color and over time, the intensity of the color grew to turn from yellowish to brown. Since within the increase in time of reaction from 10 to 15 mins from 10 hrs the colorless $AgNO_3$ solution turned pale yellow to deep red-brown during silver reduction (*Cotton Fabric*, n.d.). The study showed that further increases in reaction time such as 24 hrs and 48 hrs resulted in little change in color intensity, indicating that reaction time is complete. In this research study generated NPs were studied under UV-Vis spectrophotometer the peak absorbance was between 350 nm-530 nm and the maximum peak was observed at 350 mn. But in [38] study the Plasmon peak was observed at 450 nm and illustrates the making of AgNps. Another study showed that spectral peaks were between 430 nm to 530 nm [19].

According to SEM studies the size of synthesized AgNps was between 30 nm to 35 nm and is spherical and clumped together. According to [39] nanoparticles synthesized from The agglomerated nanoparticles were measured to be between 287.5 and 293.2 nm in size, although the usual particle dimension is predicted to be 70 nm. Moreover, Vélez et al., 2018 synthesized AgNps that were mostly spheroid within a usual size of 25 nm according to SEM studies.

The analysis of the nanoparticles we manufactured through Alovera extract was conducted through X-ray diffraction XRD. The XRD analysis shows the face-centered cubic crystalline structure. In another study the XRD examination of AgNPs produced Bragg reflection peaks at 111,200,200, and 311 were observed in the examination after biological reduction and stabilization with Alovera, these confirm that there is a cubic lattice structure centered on the nanoparticle face [19].

In this study nanoparticles synthesized from *Aloe vera* extract have strong antimicrobial properties towards *Escherichia. Coli*, (Gram-negative) *Pseudomonas luteola*, (Gram-negative) *Bacillus Subtilis* (Gram-positive), and the antibacterial activity was strong against *Bacillus Subtilis* as compared to the other two bacterial strains. From previous studies, it was observed that nanoparticles synthesized from Elettaria Cardamom the nanoparticles showed the inhibition zone but the maximum inhibition zone was shown against *Bacillus Subtilis* [40]

V. CONCLUSION

The following are the conclusions from the current research

- AgNps were effectively produced by the green synthesis method
- Organisms ranging from simple to very complex can be used to synthesize nanoparticles of the correct size and shape.
- UV-visible spectrophotometer, SEM, and XRD were used for detailed characterization of AgNps.
- We explored the antimicrobial efficacy of the AgNps we manufactured against bacteria (*Escherichia. E. coli, Pseudomonas luteola, Bacillus Subtillis*) at varying nanoparticle concentrations. The evaluation was conducted using Disc Diffusion method, revealing notably strong antimicrobial activity.
- When the outcomes were compared to the effect of antibiotics like Ciprofloxacin 5, Vancomycin 30, and Ampicillin 10, these antibiotics were shown to be less effective than nanoparticles.

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Relationship Between Compliance Level of Good Agriculture Practices with Increased Production of Sugar Cane (*Saccharum officinarum* L)

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Abstract— *The increase in sugar cane yields is influenced by many factors, one of which is farmer compliance in implementing good agriculture practices (GAP). The implementation of various cultivation methods and technologies cannot be carried out partially and must be integrated into a technology package. This technology package then becomes effective and efficient in producing high sugar yields. We conducted this research to determine to what extent the implementation of GAP by farmers affects production, including % Yield, % Brix, and % Pol sugarcane. We conducted research at the sugar cane production centre in Malang Regency. The stages of this research were carried out by a) a survey of 20 farmers and locations by looking at the compliance of the GAP with a total area of approximately 10 Ha. This sampling is Purpose Random Sampling with a modified Likert scale technique. b) Carrying out Yield Analysis and Correlation with Rank-Spearman Correlation is carried out to see the relationship between the application of GAP to the yield of sugarcane. The results show the compliance of GAP, which are based on the five main indicators of successful sugarcane cultivation, including land cultivation, fertilization, pest and disease control, defoliation, the process of sending the row material to the factory and all of them has a positive correlation to the yield of sugarcane with farmer compliance to implement GAP reaching 70 %. Then farmers' compliance with GAP in cultivation shows a positive correlation in increasing the results of %Yield, % Brix, and % Pol.*

Keywords— ***Yield, good agricultural practices (GAP), Farmer compliance, Sugar Factory***

I. BACKGROUND

Sugar is a commodity that has strategic value in the Indonesian economy because it is a staple material used directly by the community and industry (Supriyati, 2011, Syahnaz et al., 2022). National sugar consumption in 2021 will reach 7.3 million tons consisting of 3.2 million tons of consumption sugar and 4.1 million tons of industrial sugar. The sugar industry in Indonesia currently uses sugar cane as a raw material of 2.36 million tons in 2021. This number has increased by 2.58% from last year's 2.13 million tons (Puspitasari et al., 2021). Two main factors determine sugar production in Indonesia: farm or cultivation carried out by farmers and off-farm or sugarcane processing in sugar factories (Sudiarso & Prigandarini, 2022). These two factors must work together to achieve high sugar production. From the on-farm side, implementing Good Agriculture Practices (GAP) greatly determines the weight or productivity of sugarcane and its yield value. While off-farm, namely processing sugar cane into sugar, is determined by factory conditions and calculation of sugar yield (Hariadi, 2015; Yunitasari, 2015; Maghfiroh et al., 2017).

In the on-farm aspect, to ensure the success of sugarcane cultivation, the government regulates Good Agriculture

Practices for sugarcane commodities through regulations on Guidelines for Good Milled Sugar Cane Cultivation by a) variety arrangement, b) determination of planting period, c) land determination, d) soil preparation e) seed preparation f) planting g) maintenance h) harvesting (cutting, loading and transport) i) worker/labour health. In this study, we then determined GAP activities which refer to selecting seeds/seedlings, land preparation, determining varieties, planting, maintenance, pest control and harvesting (cutting, loading and transport) to be the main factors for successful cultivation. Compliance with GAP by farmers becomes absolute. Several research results evaluating the implementation of GAP in the production of several types of plants showed positive results. Shofi et al. (2019) state that applying organic SOP GAP positively correlates with organic red rice production. Mahyuda et al. (2018) state that applying GAP for Gayo arabica coffee cultivation has an impact on improving quality and production yields.

The application of GAP is a matter of serious study that needs to be carried out, considering that farmers need to pay more attention to the quality and quantity of their sugarcane production through exemplary GAP implementation. On the other hand, farmers' motivation in implementing GAP is influenced by the purchase price, which is based on determining the yield obtained (Santosa, 2021). Determination of the yield is still under the sugar factory's authority as the buyer of the farmers' crops, which determines the yield value partially carried out unilaterally. The case study found the yield was the same, namely around 7% for all sugarcane in one milling period. Thus, the yield determination results do not reflect differences in the type and quality of sugarcane. This condition decreased the quality of sugarcane because farmers were reluctant to improve the quality of the sugarcane they produced. This is because the yield value will impact farmers' income. The lower the yield value, the lower the income earned, and it is not proportional to the production inputs provided (Amir, 2010). Lukman et al. (2008) explained that the income factor is the main objective of farmers in practising sugarcane farming. So the guarantee of income is the motivation to maintain continuity and increase the quantity and quality of sugarcane production.

The polemic about yield determination for sugarcane farmers seems to be a classic problem that traps farmers and sugar mills in an ambivalent situation, Istiawati, (2006). According to Cahyani (2017) that mutual distrust arises. On the one hand, there is distrust of farmers in the Sugar Factory due to the determination of yield yields and the incentives obtained. On the other hand, the Sugar Factory knows that farmers are no longer implementing GAP properly, so the yield is determined unilaterally. Ultimately, farmers choose to convert sugarcane land into land for other crops or convert the land into non-agricultural use because all factors are within their control (Hariadi, 2015). Then, the Sugar Factory finally obtained raw materials that were rudimentary. Because farmers who apply GAP do not get good yields, they only cultivate without regard to GAP. If conditions like this are allowed to continue, it will threaten food security at the national level. The production of white crystal sugar will decrease due to the absence of raw materials, and the import of gross sugar for refined sugar materials will be higher.

The purpose of this research is to determine the appropriateness of farmers in implementing GAP and to see how far the implementation affects the sugarcane yield. Ultimately, the Sugarcane mill's trust in farmers reappeared, and vice versa, leading to a more transparent sugar yield calculation policy that satisfies the people's sugar cane farmers and benefits sugarcane mills.

II. METHOD

Selection of location and time of research

Research locations were selected purposively in sugarcane production centres in Malang Regency consisting of Gondanglegi District and Pagelaran District. The research was carried out in October-November 2021.

Methods of Data Collection and Sample Selection

1. Level of Adoption of Good Agriculture Practices

Data collection was carried out through direct interviews with 20 sugarcane farmers using a closed-method questionnaire, where the possible answer choices were predetermined, and respondents were not given alternative answers. The respondents were selected using a purposive random sampling technique with the criteria of farmers who are partners of the Gondanglegi Village Unit Cooperative and the Krebet Sugar Factory. Respondents were given a questionnaire in which each described the implementation of the GAP, including land cultivation and fertilization, such as the type of fertilizer, the amount of fertilizer, sugarcane treatment, and others that affected the yield value of sugarcane. 5% of the prominent farmers sampled with an even distribution.

2. Sugarcane Yield Results

Sugarcane samples for yield calculations were taken from sugarcane planted by 20 interviewees. A sampling of sugarcane was carried out at 5 points using the diagonal sampling method; at each point, two ovals were taken. For analysis, the calculation of the yield value was carried out at the Pasuruan Indonesian Sugar Plantation Research Center. Sugarcane samples were taken from the

same type to reduce the bias in yield measurement. The number of samples was taken according to the standard operating procedure for the sample and immediately sent to Pasuruan Indonesian Sugar Plantation Research Center to measure the yield, Brix % and Pol %. The yield value of the measurement results will be matched with the sample farmer management system and compared with the measurement of the yield value by the sugar factory.

Varieties Arrangement Sampling

The arrangement of varieties also influences the basis for returning sampling. The variety arrangement is carried out by farmers by determining the superior varieties to be planted according to the typology of the land, determining the maturity composition, suitability of the superior varieties with the cutting plan and planting period, as well as the availability of healthy, pure and timely planting materials when needed. Planting of sugarcane is carried out based on the composition of ripeness (early ripe, medium early, medium and slow middle) which is adjusted to the raw material needs of each sugar factory. The varieties used are superior by technical standards and are certified. The varieties used and the samples for this research were BL and Late Ripe (Table 5).

Table 5. Sampling-based on varieties, various criteria and types of seeds

Variety	Criteria for maturity	Type of seedling	Number
PR	Early Ripe	New Plant cane	1
BL	Late Ripe	New Plant cane	6
BL	Late Ripe	Ratoon cane	13

Data analysis

Qualitative data from the interviews were converted into quantitative using a scoring test technique using a modified Likert scale for each variable indicator of the GAP component. This study uses an ordinal scale. According to Sugiyono (2018), an ordinal scale is a measurement scale that states categories and the rank of the construct being measured. The scoring index obtained is used to determine the percentage of GAP adherence using the following formula (Apriani, 2019):

% GAP adoption rate = (Actual score)/(Maximum score) x 100%

The percentage value of applying GAP to each component obtained is analyzed about the obtained yield value of the test results. To find out the relationship between the level of application of GAP and the yield obtained using a non-parametric statistical approach with the Rank-Spearman correlation method and we used the Kruskal Wallis Test ($P<0.05$) to see a comparison between the yields produced on the new planting species and ratoons.

III. RESULTS AND DISCUSSION

Results

GAP compliance by farmers

In this study, it is examined to what extent GAP affects yields. To know the determining factors for the success of sugarcane farmers in improving their welfare. The efficiency of the use of production inputs will also be considered in this study. Determining yields by sugar factories will not burden farmers and will become a vicious circle in their partnership between farmers and sugar factories. The results of the study are shown in Figure 1. The graph shows the compliance of Good Agriculture Practices by Farmers where it is known that the new planting cane or plant cane and ratoons are trending similarly in fulfilling their GAP. Even ratoon cane tends to be more intensive in fulfilling its GAP.

Soil tillage is carried out to create a suitable growing environment for sugarcane plants from the beginning of growth to harvest so that optimal land is obtained for sugarcane growth. Soil preparation can be done through the Manual System, the Semi-Mechanized System, or the Mechanized System.

Based on the Good Agriculture Practice (GAP) compliance scoring analysis, it is known that GAP processing sugarcane land is only carried out by 64.8% of farmers. At the same time, the rest are not by the GAP, including the absence of drainage channels, such as the unfortunate gutter. This will directly affect sustainability in cultivation, especially during the rainy season. It is bearing in mind that sugarcane plants are not resistant to anaerobic conditions. Good land cultivation with crop management during ratoon increases sugarcane production and yields (Salamah, 2016; Simamora et al., 2015).

Maintenance of sugarcane plants consists of several stages, including irrigation, replanting, fertilizing, tilling the soil and mounds, cloves, drainage arrangements, and pest and disease control. Maintenance can be done with 2 (two) systems, namely the manual and mechanical systems.

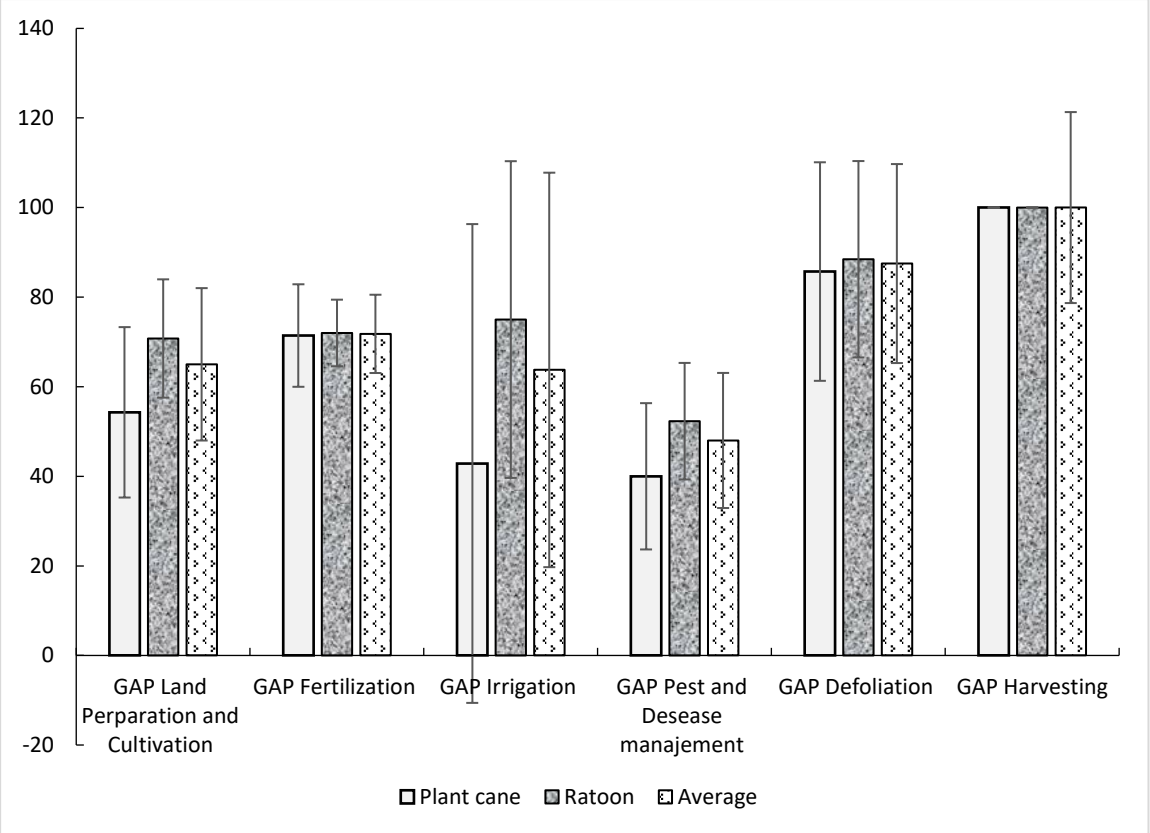


Fig.1. Graph of Fulfillment of Good Agriculture Practices by Farmers

Furthermore, in compliance with the GAP criteria for fertilization, 28% of the area still needs to meet the GAP. The GAP sugarcane fertilization is only done on 71.8% of farmers. The GAP irrigating sugarcane was only carried out in 63.8% of the sugarcane land. The rest was rainfed land that needed to be washed. In the GAP indicator, Pest and disease and weed control were carried out at 0.48% of farmers. The smallest number of the overall results is because pests and diseases have never been controlled, so the GAP is not fulfilled. Then 82% of the sugar cane area was carried out by Defoliation, where most farmers understood the benefits of Defoliation for plant quality and easier harvesting. As well as in practice, Defoliation is done two or more times, while 18% of the area is only done one time.

Slashing, loading, and transport of milled sugarcane are carried out to maximize the potential weight of sugarcane and the yield that has been formed in the plantation to become the raw material for sugar production and to fulfill the planned daily supply of quality raw materials by the milling pattern coordinated by the mill. For the success of Slashing, loading, and transport activities, it is necessary to establish proper management from planning to implementation. The results of this study indicate that 100% of the harvested area can be shipped the same day within 1 x 24 hours.

4.3 Compliance of Good Agriculture Practice with Components of Sugar Cane Products

Based on the research conducted in Figure 2, it shows that even though it comes from ratoons, if the GAP compliance is done correctly, there will not be a significant difference between the average yield % Yield (A), % Brix (B) and % Pol (C) for new sugar cane Plant cane and Ratoon.

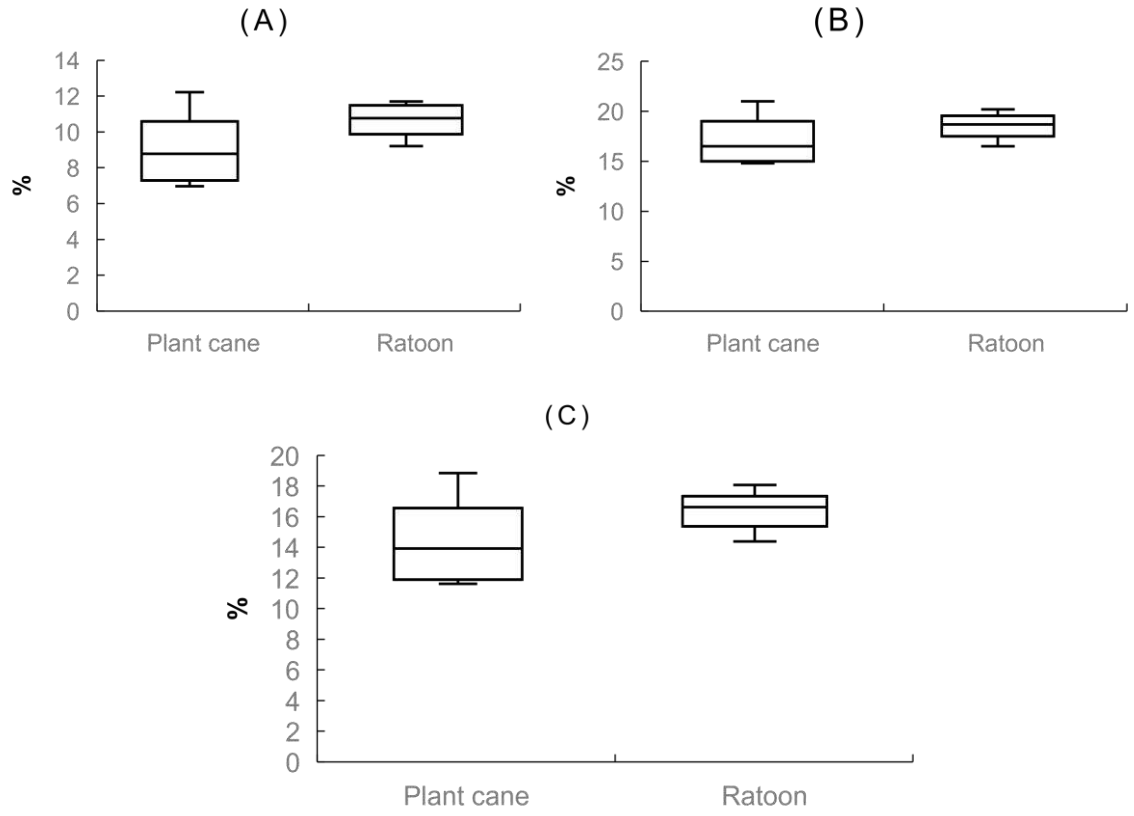
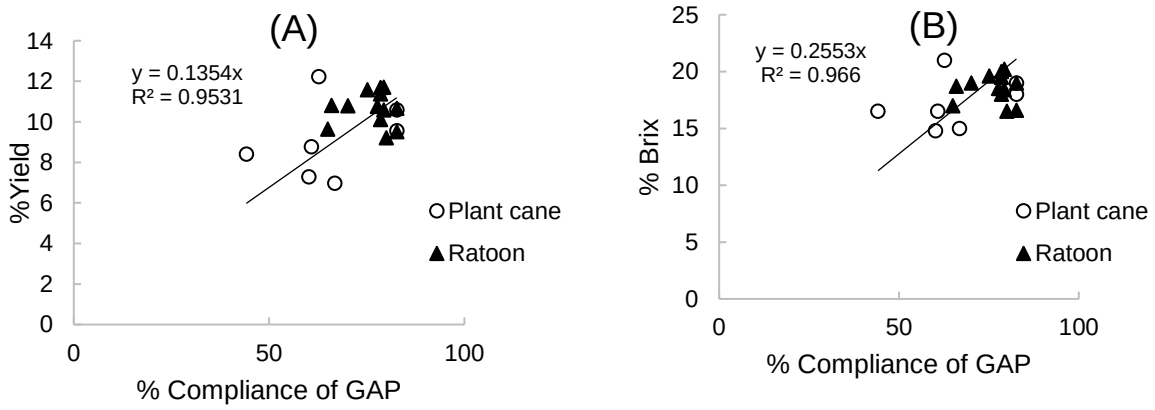


Fig.2. The average % yield (A), % Brix (B), and % Pol (C) for new sugarcane plants and ratoons, the results showed no significant difference between new sugarcane plantings and ratoons based on the Kruskal Wallis Test (P> 0.05)

We also observe whether there is a positive correlation between the average farmer compliance with GAP and the results % Yield (A), % Brix (B), and % Pol (C) for Cane and Ratoon Plants Figure 3. Both cane and ratoon plants show a positive correlation relationship between adherence to implementing GAP with % Yield, % Brix and % Pol.



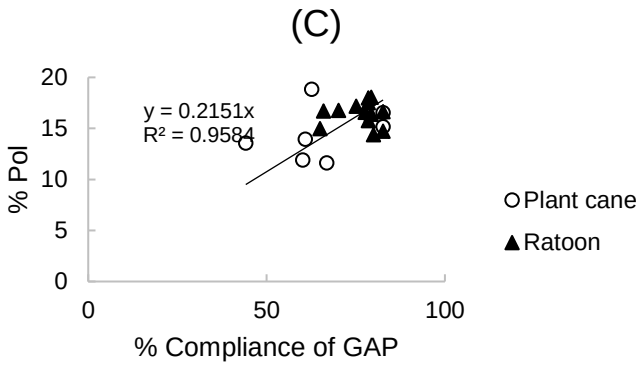
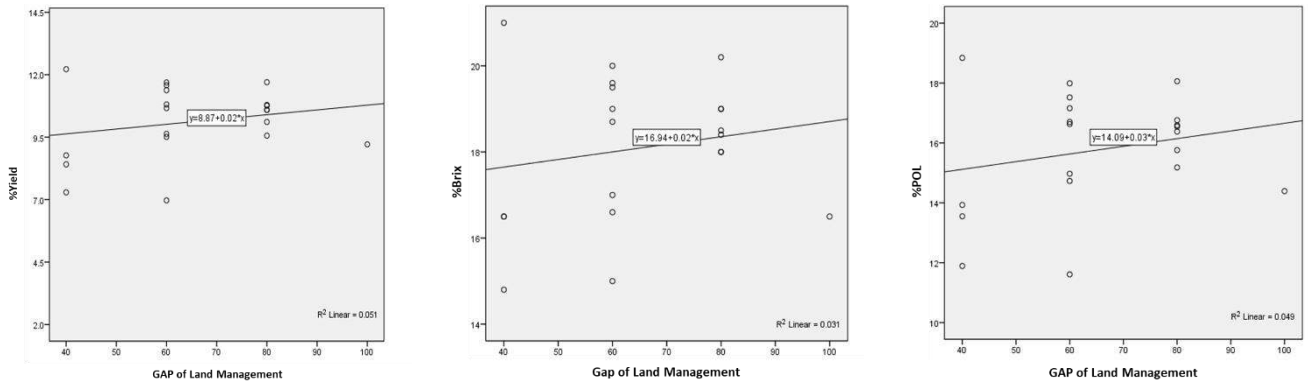


Fig.3. Regression correlation between average farmer compliance with GAP with % Yield (A), % Brix (B), and % Pol (C) for cane and ratoon plants

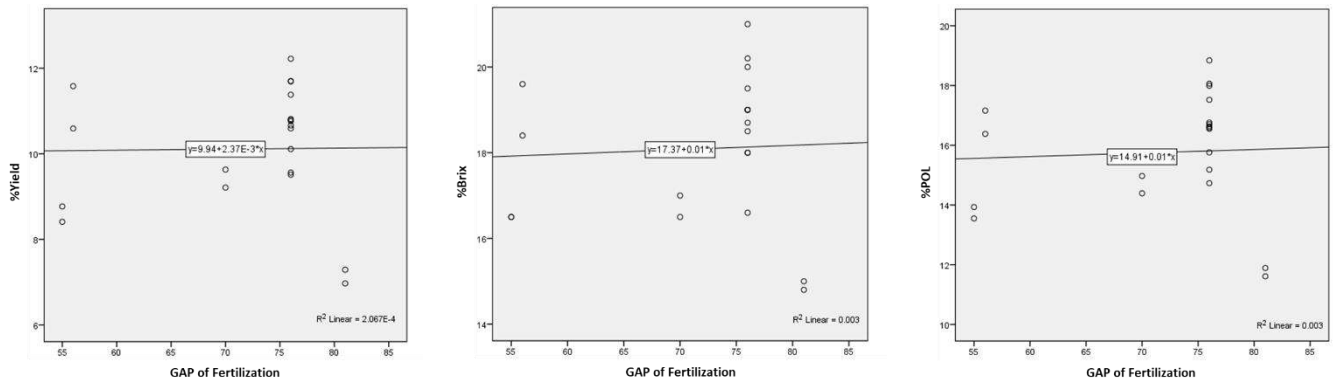
When broken down for each GAP with Spearman correlation analysis, the results show that the GAP of land cultivation with the sugar cane yield component in Figure 4. has a correlation coefficient to % yield of 0.137 and % brix of 0.143 and % pol of 0.149. As for the significance of the correlation, the results obtained between the GAP of land processing with a yield component of 0.564 for % yield, 0.546 for % brix and 0.530 for % pol. Based on these results, it can be seen that the application of GAP in land management carried out by farmers positively correlates with the components of the sugarcane yield obtained.



(A) (B) (C)

Fig.4. Graph of Correlation GAP of Land Processing with Yield Components; A). %Yield, B). %Brix, C). %Pol

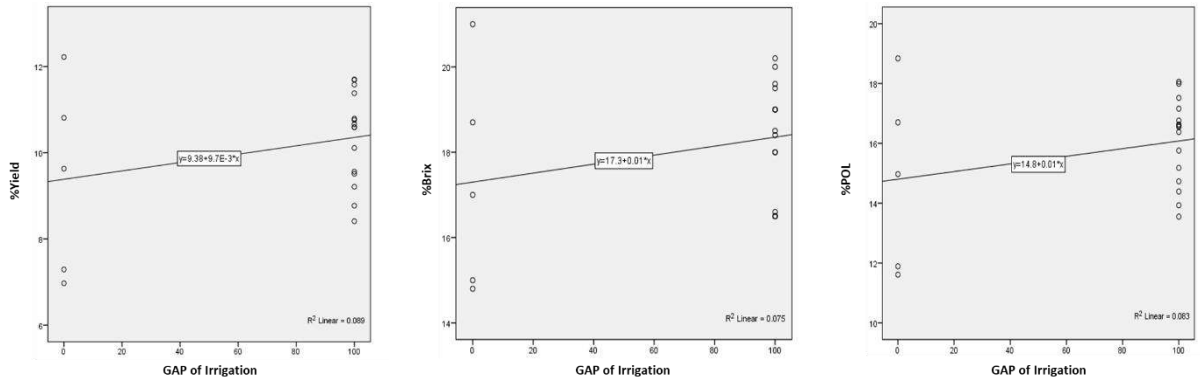
The results of the Spearman correlation analysis show that the fertilization GAP with the sugarcane yield component in Figure 5 has a correlation coefficient on the % yield of 0.043, a %brix of 0.056, and a % pol of 0.078. As for the significance of the correlation, the results obtained between GAP Fertilization with a yield component of 0.856 for % yield, 0.814 for %brix and 0.743 for % pol. Based on these results, it can be seen that the application of fertilization applied by farmers has a positive correlation with the components of the sugar cane yield obtained.



(A) (B) (C)

Fig.5. Graph of Correlation of GAP Fertilization with Yield Components; A). %Yield, B). %Brix, C). %Pol

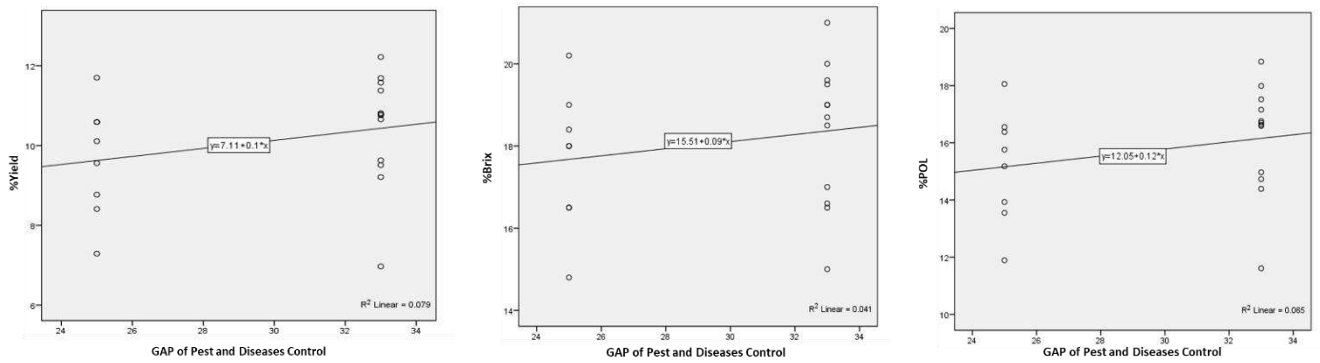
The results of Spearman's correlation analysis showed that GAP Irrigation with components of sugarcane yields had a correlation coefficient for the % yield, % brix and % pol were 0.130, 0.211, and 0.170, respectively. As for the significance of the correlation, the results obtained between GAP Irrigation with a yield component for % yield, % brix and % pol were 0.584, 0.372, and 0.473, respectively. Based on these results, it can be seen that the application of GAP in land irrigation carried out by farmers positively correlates with the components of the sugarcane yield obtained.



(A) (B) (C)

Fig.6. Graph of Correlation of Irrigation GAP with Yield Components; A). %Yield, B). %Brix, C). %Pol

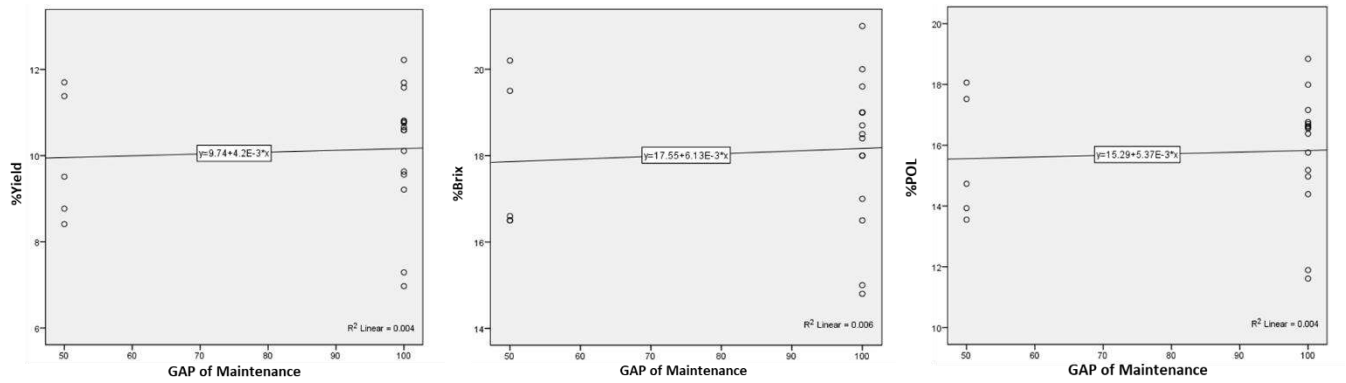
The results of Spearman's correlation analysis show that the GAP of pest control with the sugarcane yield component in Figure 7. has a correlation coefficient to % yield, % brix, and % pol ware 0.336, 0.266, and 0.319, respectively. As for the significance of the correlation, the results obtained between the GAP of pest control and the yield component such as % yield, % brix and % pol were 0.147, 0.256, and 0.171, respectively. Based on these results, it can be seen that the application of GAP in pest control carried out by farmers has a positive correlation with the components of the sugarcane yield obtained.



(A) (B) (C)

Fig.7. Graph of pest and diseases control GAP Correlation with Yield Components; A). %Yield, B). % Brix, C). %Pol

The results of Spearman's correlation analysis show that the maintenance GAP with the sugar cane yield component in Figure 8 has a correlation coefficient to % yield of 0.090 and % brix of 0.070, and % pol of 0.070. As for the significance of the correlation, the results obtained between GAP Maintenance with a yield component of 0.705 for % yield, 0.768 for % brix and 0.769 for % pol. Based on these results, it can be seen that the application of GAP in maintenance carried out by farmers positively correlates with the components of the sugarcane yield obtained.



(A) (B) (C)

Fig.8. Graph of Maintenance GAP Correlation with Yield Components; A). % Yield, B). % Brix, C). % Pol

IV. DISCUSSION

The results of our research show the importance of GAP land management as one of the critical activities in successful cultivation. Land processing is one of the land preparation techniques for planting. This aims to provide seeds and seeds with an optimal place to grow for their growth. In a study by Lubis et al. (2015) and Awe et al. (2020), it was shown that tillage and soil drainage affected increasing plant height, stem diameter and the number of age segments. The highest production estimation was obtained in good drainage treatment with deep knife processing. Deep knife processing produces higher production estimates than shallow knife processing under excellent or poor drainage conditions. This shows how vital the GAP of land management is in sugarcane cultivation.

Applying fertiliser can increase crop yields supported by the correct dosage. In a study conducted by Djajadi et al. (2016), sugarcane production and profit increased with the application of Si fertiliser, a concentration of 30% Si produced cane with the highest weight and yield and sugar rock crystal, which respectively 184.16 tons/ha and 8.36% and 15.37 tons/ha. In addition, according to Magandi (2019), Nitrogen fertilisation through Za fertiliser has a positive correlation with increasing sugarcane production and yield, so the results of this study also provide an accurate picture of the application of GAP to fertilisation, which can increase sugarcane % yield, % brix and % pol.

In fulfilling the GAP for proper irrigation, applying GAP in fertilisation can increase the % yield, % Brix and %pol of sugarcane. In line with Ardiansyah (2015), Yusara et al. (2019) state that providing water can also increase production and product at the proper needs and with the correct administration. Provision of water

according to the needs and growth phase of the plant as well as the harvest or milling period of the sugar factory.

Defoliation is an activity we then include in one of the GAPs for sugarcane cultivation. Defoliation aims to reduce pest and disease attacks. Apart from that, to improve air circulation in the garden and make it easier for sunlight to enter, C4 plants need good circulation to increase their production. Another benefit of Klentek is to prevent roots from coming out of the segments, which can disrupt plant growth and increase the number of tillers of sugarcane. (Arceneaux et al., 1980; Sales et al., 2021), Then according to Islam et al. (2016), yields and yield quality are obtained when defoliation is carried out by 40% of the number of leaves.

Integrated pest and disease control is also a GAP criterion for increasing sugarcane production. Research by Muliasari & Trilaksono (2020); Wicaksono (2012) shows that pest and disease control directly increases sugarcane production. So pest and disease control causes a decrease in sugarcane production by up to 10%, according to Subiyakto (2016). This study illustrates that the right farmers' low compliance with the GAP for pest and disease control can increase sugarcane production.

The GAP harvest and delivery time also determines the quality of the yield. According to Sriwana & Djatna (2012); Yusrina (2016), The obstacle in the distribution of sugarcane is that if the sugarcane is processed after 36 hours from the time of cutting, the yield of sugarcane will decrease (a good work of sugarcane ranges from 8% to 10%). If there are more than 36 hours of accumulation, the sugar cane yield will decrease by 1% to 2% so that productivity can decrease. So it is essential to ensure that the cane is sent immediately after cutting in less than 24 hours. In line with the implementation of this survey and research, which shows that the increase in production and yield of sugarcane is strongly influenced by farmer compliance with GAP from start to finish, including timely delivery.

V. CONCLUSION

The implementation of GAP in sugarcane cultivation determines the success of sugarcane cultivation in Indonesia. From the research results, all indicators of Good Agriculture Practices positively correlate with the sugar yield. This means that better land management by sugar cane farmers significantly increases sugar cane harvest. There is no significant difference between the newly planted cane and ratoons when farmers compliance the GAP. This illustrates how crop production can grow along with improving good cultivation management by GAP. The average yield obtained was 10.11% with % Brix 18.09% and 15.76% Pol while the average GAP compliance by farmers was 72%. This shows that the sugar cane produced by farmers in the sampling is higher than the average national yield of 7%.

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Enhancing maize productivity under abiotic stresses through the combined use of nitrogen, potassium humate, and zinc

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Abstract— *The soils found in the Kharga oasis in Egypt have been identified as having low levels of nitrogen and organic matter, along with high pH and salinity. These conditions make it difficult for the plants to absorb essential micronutrients. In order to address this issue, a study was conducted with the main objective of evaluating the interactive effect of potassium humate, zinc, and nitrogen fertilization on the grain yield of maize and its various components. The study consisted of two field experiments that took place during the growing seasons of 2022 and 2023 at the research station of the Desert Research Center in Kharga, located in the western desert of Egypt. The treatments in the experiments involved three different levels of potassium humate and zinc combinations, including 20 kg/ha of potassium humate, 375 ppm of zinc, and a combination of both, which were compared to a control group. Additionally, four levels of nitrogen fertilization were applied, ranging from 100 to 400 kg/ha. The results of the study revealed that the highest and most significant grain yields were observed in both seasons when potassium humate and zinc were applied together, resulting in yields of 10,436 and 10,590 kg/ha in the first and second seasons, respectively. Furthermore, it was found that the highest significant grain yields in both seasons were achieved by applying 300 kg of nitrogen per hectare, with yields of 9423 and 9196 kg/ha in the first and second seasons, respectively. These findings suggest that the combination of nitrogen fertilization with potassium humate and zinc proves to be effective in maximizing the grain yield of maize. This information is valuable for farmers and researchers in the Kharga oasis, as it provides insights into the optimal fertilization practices that can enhance crop productivity in this specific region with challenging soil conditions.*

Keywords— ***Maize, potassium humate, zinc, nitrogen, yield, Kharga oasis***

I. INTRODUCTION

Maize (*Zea mays* L.) is the second most important cereal crop in Egypt in terms of harvested area, serving as both a major field crop and a staple food source. In 2022, Egypt's maize production reached around 7.5 million metric tons with a slight increase of production of 6.85 % as compared to 2018-2023 production average (**FAO, 2024**). The slight increase in production may be due to the increase in the cultivated area. However, around 38% of Egypt's maize is imported because of the country's insufficient production to meet demand (**Altaie *et al.*, 2022**). The goal of the Egyptian strategy for sustainable agricultural development (2030) was to raise maize production to 18.5 million tons in order to boost 91% of the country's self-sufficiency by 2030 (**El Shamy *et al.*, 2023**). To increase production and productivity, interventions are needed at both the production and policy levels, including the increased use of high-yielding varieties of quality seeds (**Altaie *et al.*, 2022**).

Chemical fertilizers are synthetic inorganic materials used to maintain plant development. Usually, they are rich in nutrients that are vital to plant growth, such as potassium, phosphate, and nitrogen. Chemical fertilizers tend to be less expensive than organic fertilizers

and can be used to quickly address issues with plant growth, optimize soil conditions, and improve crop production and quality. On the other hand, overuse of chemical fertilizers can result in more acidic soil, chemically tainted water sources, damaged plants due to overgrowth, and emissions that contribute to climate change. Several studies have replaced partially or completely the chemical fertilizers especially NPK with organic fertilizers, for example in maize. **Li-Chao *et al.* (2023)** suggested that the kernel position effect in maize can be mitigated by applying both chemical and organic fertilizers together, which will increase post-silking nitrogen uptake and dry matter accumulation. Furthermore, it has been demonstrated that combining biochar with chemical fertilizers improves the physio-biochemical characteristics of the soil and maize production, suggesting a possibility for raising crop productivity and soil fertility (**Wu *et al.*, 2023**). These results emphasize the significance of investigating sustainable farming methods that preserve or increase crop yields while lessening dependency on synthetic fertilizers.

Nitrogen fertilization influences several aspects of plant growth and resource utilization, which is important for maximizing maize grain yield. The main benefits of nitrogen fertilization on maize include increase photosynthesis (**Nasar *et al.*, 2021)**, radiation use efficiency (RUE; **Bonelli *et al.*, 2020**), water use efficiency (WUE; **Guo *et al.*, 2021**), and nitrogen uptake (**Su *et al.*, 2020**. In addition to improve root architecture and the distribution of deep roots, increase height of the plant, ear, and leaf area, increase the number of ears per hectare and grain production per ear, higher total grain yield and nitrogen usage efficiency (NUE) **Ma *et al.*, 2022**. High nitrogen inputs may initially maximize maize yield, but they can lead to a decrease in NUE if excessive amounts are applied. Optimal nitrogen fertilizer rates typically range from 200 to 300 kilograms per hectare (kg/ha) (**Su *et al.*, 2020** and **Geith *et al.*, 2022**). Split applications of nitrogen fertilizer can help maintain NUE and reduce losses due to leaching or denitrification (**Davies *et al.*, 2020**)

The choice of nitrogen fertilizer rate and timing depends on local environmental conditions, soil types, and agronomic practices. **Omar *et al.* (2020)** found that out of the four N treatments, the optimal N application between 50–100 kg N ha^{-1} potentially increased the maize yield, however **Hammad *et al.* (2022)** recommended that for temperature-tolerant hybrid maize, applying 300 kg N ha^{-1} is advised as one of the best agricultural management techniques to get the highest possible grain production.

Foliar micronutrient application is a fast-acting and efficient method to increase crop yields and improve nutrient uptake, especially in conditions when root uptake is compromised, like late crop stages, nutrient-poor soils high soil pH. Studies show that foliar micronutrient treatment increases soil biological activity and increases nutrient-use efficiency (NUE) (**Bana e*t al.*, 2022**), and improves crop bio fortification (**Hao *et al.*, 2021**). Zinc (Zn) is an essential micronutrient that is required by plants, animals and humans for growth. Zinc is required for a large number of enzymes that are involved in protein synthesis, energy transmission, and nitrogen metabolism. Zinc foliar spray can promote the formation of reproductive organs, such as ears, and improve the capacity for grain productivity in zinc-deficient maize plants, resulting in higher growth and yield (**Singh *et al.*, 2020**). While foliar treatments have these advantages, it's crucial to remember that they work better when combined with appropriate soil fertilization techniques that are based on soil and tissue tests. This ensures that all nutrients are managed thoroughly for the best possible plant growth (**Shahrajabian *et al.*, 2022**).

Several benefits in crop productivity were increased when nitrogen fertilizer and zinc foliar treatment were combined. In wheat, **Zhang *et al.*, 2021** stated that, foliar Zn and soil N applications effectively raised wheat grain production and N and Zn concentrations, especially in the endosperm, and this is could be useful approaches to treat Zn insufficiency. In dry conditions, foliar application of zinc either alone or in conjunction with other substances like trehalose can improve maize growth and nutrient status (**Klofac *et al.*, 2023**). While N fertilization increases overall grain yields and nutrient concentrations, there are still some restrictions on the precise advantages of applying N and the foliar application of Zn together. However, in order to maximize the nutritious content of cereal grains, it is still imperative to optimize the time and dosage of these inputs. Consequently, more research is required to identify the best formulations, doses, and application schedules.

Research findings indicate that humic acid may cause mild acid stress responses in maize seedlings, resulting in increased root development and physiological responses (**Baía *et al.*, 2020**). Additionally, combining humic acid with zinc foliar application enhanced sunflower production under drought stress by increasing grain weight, head diameter, and number of seeds per head (**Hammati 2016**). Although there aren't many research studies about maize in the literature, there is a general evidence supporting the idea that applying zinc and humic acid together improves crop health and productivity in a variety of crops (**Manas *et al.*, 2014 and Sharma et al.,**

2023). According to a number of studies, humic acid enhances nitrogen use efficiency (NUE), reduces nitrogen leaching, and increases maize production and nitrogen uptake (**Azeem *et al.*, 2014, Kong *et al.*, 2022 and Brodowska *et al.*, 2023**) . In conclusion, humic acid seems to be a useful tool for improving nitrogen management in maize agrosystems.

The primary aim of this research is to investigate the influence of varying nitrogen fertilization rates, in conjunction with the application of potassium humate to the soil and zinc spray on the leaves, on the yield of maize cultivated in the Kharga oasis of Egypt. The study specifically focuses on the effects of abiotic stress factors such as high temperatures and salinity on the productivity of maize.

II. MATERIALS AND METHODS

1- Experimental Site Description and Soil Properties

A field experiment was performed to evaluate the impact of nitrogen fertilization, potassium humate addition and zinc spray on the yield and its components of maize (Single hybrid; Giza 168) at the Desert Research Center station in Kharga Oasis in the New Valley Governorate (25.52° N and 30.61° E). The experiments were conducted during the two successive seasons of 2022 and 2023. Soil and irrigation water samples were taken from the experimental site before sowing in both study seasons to estimate the physical and chemical properties.

Table 1: Physical and chemical properties of the experimental soil.

Season	Particles (%)			Texture	EC (dS/m)	pH	Soil available nutrients (ppm)		
	Sand	Silt	Clay				N	P	K
2021	77.3	15.4	7.3	sand	8.6	8.1	67	6.35	110
2022	78.5	14.9	6.6		7.8	8.3	84	7.44	123

Table 2: Analysis of irrigation water

Season	pH	E.C. ds/m	S.A.R	Soluble cations (meq/l)				Soluble anions (meq/l)			
				Ca^{++}	Mg^{++}	Na^{+}	K^{+}	$CO_3^{=}$	HCO_3^{-}	$SO4^{=}$	Cl^{-}
2021	7.84	3.38	6.86	13.68	2.74	14.82	0.41	-	5.43	4.37	9.47
2022	7.79	3.32	6.14	15.32	2.93	14.51	0.45	-	5.69	4.76	10.24

Kharga oasis is characterized by a tropical arid climate. The mean annual temperature is 25°, average mean temperature is 12°C and the average maximum temperature is 41°C. The maximum absolute temperature is 52°C which usually occurs in August and the absolute minimum temperature is 2°C which usually occurs in January. August is typically a low-wind month; from November to January, wind speed gradually increases, peaking from March to May and producing the infamous "El-Khamasin" dust storms. The relative humidity has an annual mean value of roughly 39%. In general, atmospheric precipitation in the form of rainfall is incredibly rare (1 mm/year) with heavy rainstorms from time to time.

2. Experimental Design and Layout

The experimental treatments were set up in a split-plot design with four replications. In comparison to the control, the main plots represented the addition of potassium humate (C_9 H_8 K_2 O_4) to the soil at a rate of 20 kg/ha which was added three times with the N fertilization, and the application of chelated zinc spray at a rate of 250 g/100 L of water (in the form of 15% EDTA-$ZnNa_2$ ($C_{10}H_{12}N_2O_8ZnNa_2$), or 375 ppm, at three times i.e., 3,5 and 7 weeks after sowing. Sub-plots were occupied by four N fertilizer levels: 100 kg/ha, 200 kg/ha, 300 kg/ha, and 400 kg/ha. Nitrogen was supplied in three doses within 35 days of sowing, or until the 7–8 leaf stage of maize growth.

Maize was sown under drip irrigation system in ridges with 70 cm apart and 4 m in length (plot area was 28 m^2). Sowing was in hills, on one side of the ridge (the distance between hills was 25 cm)

The sowing was carried out at the first week of August in both seasons, according to the suitable sowing time of the study region. After soil preparation, The conventional crop practices of the region were applied in

both seasons. P was applied in the form of calcium monophosphate (15.5% P_2O_5) at the rate of 350 Kg/ha, while N was applied in the form of Ammonium nitrate (33.5% N) and K was applied in the form of potassium sulfate (48% K_2O) at the rate of 125 Kg /ha. Experimental units were kept weed-free through hand hoeing at early stages and hand pulling at later stages to eliminate the weed effect.

3. Studied Characters and Measurements

The harvesting was performed on the same time in each treatment, where the grains moisture content was between 20% and 30%, according to the conventional harvesting practices in the region . Grain yield (Kg/ha) was determined from four inner guarded ridges of each plot and transformed to yield/ha. The yield components i.e., Plant height (cm), ear height (cm), Ear length (cm), Ear diameter (cm), number of rows per ear, number of grains per row and 100-grains weight (g) were calculated in samples of 10 plants:

4. Statistical Analysis

The data were analyzed using SPSS (version 20), and treatment means were compared using the least significant difference (LSD 0.05) for the individual factors as well as the interaction.

III. RESULTS AND DISCUSSION

1- Impact of Potassium humate and zinc application

Table 3 illustrates the impact of applying Zn and potassium humate on maize yield parameters. Notably, the most noteworthy outcomes in terms of plant height were observed during both the first and second seasons when both potassium humate and Zn were applied together. The plant height values recorded were 233.5 cm and 220.5 cm, respectively, surpassing the results obtained from individual applications. These findings strongly indicate that the combined use of potassium humate and Zn exhibits a synergistic impact on enhancing the growth of maize, specifically in relation to plant height.

Table 3. Effect of Zn and potassium humate application on maize growth parameters in the two studied seasons.

Treatment	Plant height (cm)	Ear height (cm)	Ear length (cm)	Ear diameter (cm)	No. of rows/ear	No. of grains/row	100-grain weight (g)	Grain yield (kg/h)
The first season 2022								
Control	226.9 c	127.3 c	21.25 c	15.17 b	13.83 a	39.33 c	31.17 c	7528 d
Zn	229.5 b	129.8 b	22.21 b	15.63 ab	14.17 a	41.33 b	33.33 b	8773 b
P.H.	229.0 b	128.9 b	21.42 c	15.67 ab	14.58 a	39.92 c	32.33 a	8273 c
P.H.+Zn	233.5 a	132.6 a	23.92 a	15.96 a	14.67 a	43.67 a	33.42 b	10436 a
The second season 2023								
Control	218.4 b	117.5 c	21.77 b	19.23 c	14.08 b	39.25 c	29.33 c	7657 d
Zn	213.8 c	118.0 b	21.10 bc	19.81 b	14.42 ab	40.58 b	31.50 b	8675 b
P.H.	208.0 d	114.9 d	20.63 c	20.00 ab	14.42 ab	39.33 c	30.92 b	8456 c
P.H. +Zn	220.5 a	123.7 a	23.99 a	20.23 a	14.75 a	42.00 a	32.37 a	10590 a

In terms of ear height of maize, the highest significant values in both seasons were achieved by applying both potassium humate and Zn. The ear height in the first season was 132.6 cm, while in the second season it was 123.7 cm. Regarding of ear length, the most significant increase was observed when also applying both potassium humate and Zn in both seasons. During the first season, the ear length measured 23.92 cm, while in the second season it reached 23.99 cm.

During the first season, there was no significant variability in ear diameter when Zn and potassium humate were applied either individually or in combination, with values ranging from 15.63 to 15.96 cm. However, in the second season, the application of potassium humate and a combination of potassium humate and Zn resulted in significantly higher values of 20 and 20.23 cm, respectively. This demonstrates the positive impact of these treatments on ear diameter growth.

There was no consistent trend observed in the number of rows per ear in both seasons. However, there was a clear trend in the number of grains per ear. The application of potassium humate and Zn resulted in the highest significant values of 43.67 and 42.0 in the first and second seasons, respectively. This suggests that the use of

potassium humate and Zn can positively impact the number of grains per ear in maize crops.

In terms of 100-grain weight, the highest significant value was observed in the first season with the application of potassium humate, yielding a value of 32.33 g. Interestingly, in the second season, the combination of potassium humate and Zn application resulted in an even higher significant value of 32.37 g. This suggests that the combined application of potassium humate and Zn may have a synergistic effect on grain weight.

Regarding maize grain yield, the highest significant values were observed in both seasons when potassium humate and Zn were applied together, outperforming individual applications. During the first season, the combined application resulted in a yield of 10,436 kg/ha, while in the second season, the yield reached 10,590 kg/ha.

The results suggest that the synergistic effect of potassium humate and Zn on maize grain yield is particularly strong when applied together. The combined application seems to have a positive impact on plant growth, development, and ultimately yield. Potassium humate, derived from natural organic matter, acts as a soil conditioner, enhancing soil structure and promoting nutrient uptake by the roots (Kumar *et al.*, 2013; Ibrahim and Ali, 2018; Jin *et al.*, 2022). This leads to improved water retention in the soil, increased availability of essential nutrients like nitrogen, phosphorus, and potassium, and enhanced microbial activity (Gatabazi, 2014; Kumar *et al.*, 2022). Zn plays a crucial role in plant development, especially in photosynthesis and enzyme activation (Solanki, 2021; Khan *et al.*, 2022). By applying potassium humate and Zn together, farmers can ensure that their crops receive all the necessary elements for healthy growth and maximal productivity. The improved yield can be attributed to the enhanced nutrient uptake and utilization, as well as the stimulation of plant metabolism by potassium humate and Zn.

2- Impact of nitrogen fertilization

Table 4 presents the impact of N application rates on maize growth parameters. In terms of plant height, the most significant increase in the first season was observed with the application of 400 kg N/ha, resulting in a height of 226.5 cm. During the second season, application rates of 300 and 400 kg N/ha produced statistically similar results, with plant heights of 215.9 cm and 216.9 cm, respectively. These results suggest that higher N application rates can lead to increased plant height in maize. However, there may be a point of diminishing returns, as the difference in plant height between the 300 kg N/ha and 400 kg N/ha treatments was not statistically significant in the second season.

Table 4. Effect of nitrogen application rate on maize growth parameters in the two studied seasons.

N levels	Plant height (cm)	Ear height (cm)	Ear length (cm)	Ear diameter (cm)	No. of rows/ear	No. of grains/row	100-grain weight (g)	Grain yield (kg/h)
The first season 2022								
100 kg	226.7 c	127.9 c	21.13 b	15.29 b	14.58 a	38.17 c	31.33 c	7936 d
200 Kg	231.1 b	130.8 b	22.67 a	15.50 ab	14.33 a	40.83 b	33.08 b	8581 c
300 Kg	234.7 c	132.7 a	22.58 a	15.96 a	14.25 a	43.75 a	31.50 c	9423 a
400 Kg	226.5 a	127.3 c	22.42 a	15.67 ab	14.08 a	41.50 b	34.33 a	9070 b
The second season 2023								
100 kg	214.3 b	117.0 c	21.29 a	19.58 b	14.58 a	38.83 c	30.65 a	8322 d
200 Kg	213.8 b	117.9 c	21.75 a	19.83 ab	14.42 a	40.83 b	31.38 a	8840 c
300 Kg	215.9 a	119.1 b	21.81 a	19.83 a	14.33 a	43.25 a	31.08 a	9196 a
400 Kg	216.9 a	120.8 a	21.69 a	19.85 ab	14.33 a	38.25 c	31.38 a	9020 b

During the first season, the highest significant value for ear height was achieved by applying 300 kg N/ha, resulting in a height of 132.7 cm. In contrast, the highest value in the second season was attained by applying 400 kg N/ha, resulting in a height of 120.8 cm. However, there was no distinctive trend in terms of ear length, ear diameter, and number of rows per ear in both seasons. This suggests that while N application had a clear impact on ear height in both seasons, it did not have a consistent effect on other important maize yield components such as ear length, diameter, and number of rows per ear. This could indicate that factors other than N

levels may be influencing these traits in the maize plants, highlighting the complexity of plant growth and development.

In terms of the number of grains per row, the highest significant values were achieved in both seasons by applying 300 kg N/ha, with values of 43.75 and 43.25, respectively. Additionally, the highest significant value for 100-grain weight in the first season was achieved by applying 400 kg N/ha, with a value of 34.33 grams. However, there was no specific trend observed in the second season. This suggests that the application of 300 kg N/ha is optimal for maximizing the number of grains per row in both seasons. However, when it comes to 100-grain weight, a higher rate of nitrogen application (400 kg N/ha) may be more beneficial in certain circumstances, such as in the first season. The lack of a specific trend in the second season could indicate that other factors may have influenced the results, and further investigation may be needed to determine the most effective nitrogen application rate for that particular season.

The highest significant grain yield in both seasons was achieved by applying 300 kg N/ha, with values of 9423 and 9196 kg/ha in the first and second seasons, respectively. It is noteworthy that this treatment resulted in a higher yield than applying 400 kg N/ha. This may be attributed to the fact that higher doses of N can impair yield through various mechanisms. One possible explanation for the lower yield with 400 kg N/ha could be N toxicity, where excessive N can lead to imbalances in nutrient uptake and negatively impact plant growth (Vitousek *et al.*, 2009; Guo *et al.*, 2019; Aliyu *et al.*, 2021). Additionally, high levels of N can also contribute to increased susceptibility to diseases and pests (Szulc *et al.*, 2008; Huber *et al*., 2012; Veromann *et al.*, 2013; Virla *et al.*, 2023).

Impact of interaction between potassium humate, Zn, and nitrogen application

Table 5 shows the influence of the interaction effect between potassium humate, Zn, and N application rate on maize growth parameters. In terms of plant height, the most significant values were observed in both seasons when applying 300 kg N/ha in conjunction with potassium humate and Zn. The plant height reached 243.7 cm in the first season and 229.7 cm in the second season under these conditions. This suggests that the combination of 300 kg N/ha with potassium humate and Zn had a positive impact on the growth and development of maize plants, resulting in taller plants compared to other treatments. The significant increase in plant height could be attributed to the synergistic effects of nitrogen, potassium humate, and Zn on promoting nutrient uptake, photosynthesis, and overall plant health.

Table 5. Effect of the interaction between potassium humate, Zn, and nitrogen application rate on maize growth parameters in the two studied seasons.

Zn + P.H treatments	N	Plant height (cm)	Ear height (cm)	Ear length (cm)	Ear diameter (cm)	No. of rows/ear	No. of grains/row	100-grain weight (g)	Grain yield (kg/h)
The first season 2022									
Control	100	222.3 g	127.0 e	19.83 i	15.00 bc	13.58 a	33.67 g	30.00 f	6964 i
	200	228.3 de	127.0 e	21.67 efg	14.83 c	13.67 a	40.67 cde	31.67 ef	7305 h
	300	229.6 cde	129.0 cde	21.33 fgh	15.50 abc	13.67 a	41.00 cde	33.00 cde	7954 fg
	400	227.3 def	126.3 e	22.17 cdefg	15.33 abc	13.58 a	42.00 cd	30.00 f	7891 g
Zn	100	227.0 defg	129.3 cde	21.50 fgh	15.67 abc	14.67 a	38.67 ef	33.00 cde	8215 f
	200	229.3 cde	130.0 cde	22.00 defg	15.33 abc	14.58 a	40.67 cde	34.00 bc	8215 f
	300	234.0 bc	132.7 bc	22.17 cdefg	15.83 abc	14.33 a	45.00 ab	35.33 ab	9465 cd
	400	227.7 def	127.3 de	23.17 bcd	15.67 abc	13.67 a	41.00 cde	31.33 ef	9197 d
P.H.	100	226.3 efg	127.3 de	20.17 hi	15.33 abc	15.67 a	37.00 f	31.33 ef	7096 hi
	200	230.3 cde	129.7 cde	22.50 cdef	15.33 abc	13.67 a	39.00 ef	33.00 cde	8020 fg

	300	231.3 cd	131.0 cd	22.17 cdefg	16.17 abc	14.67 a	43.33 abc	33.00 cde	9296 cd
	400	228.0 de	127.7 de	20.83 ghi	15.83 abc	14.33 a	40.33 de	32.00 de	8678 e
P.H.+ Zn	100	231.0 cde	128.0 de	23.00 cde	15.17 abc	14.58 a	42.67 bcd	31.67 ef	9469 c
	200	236.3 b	136.3 ab	24.50 ab	16.33 ab	14.67 a	43.33 abc	33.67 bcd	10784 a
	300	243.7 a	138.0 a	24.67 a	16.50 a	14.33 a	45.67 a	36.00 a	10978 a
	400	223.0 fg	128.0 de	23.50 abc	15.83 abc	14.33 a	43.00 abcd	32.00 de	10512 b
The second season 2023									
Control	100	209.7 ef	113.0 gh	21.58 abcd	18.83 c	14.00 a	36.67 e	28.17 e	6917 g
	200	216.3 d	115.7 fg	21.67 abcd	19.33 bc	14.00 a	41.00 bcd	30.00 cde	7873 ef
	300	221.3 c	121.3 cd	22.00 abcd	19.42 bc	14.00 a	42.33 ab	30.00 cde	7936 ef
	400	226.3 ab	120.0 de	21.83 abcd	19.33 bc	14.33 a	37.00 e	29.17 de	7903 ef
Zn	100	209.7 ef	113.0 gh	20.75 bcd	19.83 ab	14.67 a	39.33 cde	30.83 bcd	8396 d
	200	211.3 ef	118.0 ef	20.83 bcd	19.67 abc	14.33 a	40.67 bcd	31.67 abc	8396 d
	300	221.3 c	122.0 bcd	21.58 abcd	19.92 ab	14.33 a	43.00 ab	32.17 abc	9020 c
	400	213.0 de	122.0 bcd	21.25 abcd	19.83 ab	14.33 a	39.33 cde	31.33 abcd	8889 c
P.H.	100	199.7 g	112.0 h	20.00 d	19.83 ab	15.00 a	37.67 e	30.33 bcde	7870 f
	200	208.0 f	112.0 h	20.33 cd	19.83 ab	13.00 a	38.67 de	30.50 bcd	8330 de
	300	213.0 de	120.3 cde	21.17 abcd	20.25 ab	14.33 a	43.33 ab	31.83 abc	8965 c
	400	212.0 ef	115.3 fg	21.00 abcd	20.08 ab	14.33 a	37.67 e	31.00 abcd	8659 cd
P.H.+ Zn	100	213.0 de	121.0 cde	22.58 abc	19.83 ab	14.67 a	39.00 cde	31.63 abc	10105 b
	200	216.3 d	123.3 abc	23.33 a	20.43 b	15.33 a	43.00 ab	32.25 abc	10762 a
	300	229.7 a	125.3 a	23.42 a	20.50 b	14.67 a	44.33 a	33.17 a	10861 a
	400	223.0 bc	125.0 ab	22.83 ab	20.17 ab	14.33 a	41.67 ab	32.42 ab	10631 a

Significant improvements in ear height were observed when applying either 200 or 300 kg N/ha in combination with potassium humate and Zn. In the first season, ear heights reached 136.3 and 138 cm, while in the second season, they measured 123.3 and 125.3 cm, respectively. Similarly, these treatments also led to the highest significant values in ear length during the first season, with measurements of 24.5 and 24.67 cm. However, no specific trend was observed in the second season. These results suggest that the combination of nitrogen, potassium humate, and Zn can have a positive impact on ear height and length in maize crops. The significant improvements seen in the first season indicate that these treatments can potentially enhance the growth and development of maize plants. However, the lack of a specific trend in the second season suggests that other factors may have influenced the results.

In terms of ear diameter, number of rows per ear, and number of grains per row, no significant trends were observed among the treatments. However, during the first season, the weight of 100 grains showed a notable result with the highest significant value of 36 grams in the treatment involving the application of 300 kg N/ha along with potassium humate and Zn. Conversely, no specific trend was evident in the second season. This suggests that the combination of 300 kg N/ha, potassium humate, and

Zn had a positive impact on grain weight in the first season. However, it is important to note that this trend was not consistent in the second season, indicating that other factors may have influenced the results.

The highest grain yields in the first season were achieved by applying either 200 or 300 kg N/ha along with potassium humate and Zn. Specifically, the grain yields in these treatments were 10,784 and 10,978 kg/ha, respectively. In the second season, applying 200 or 300 kg N/ha along with potassium humate and Zn, as well as applying 400 kg of N/ha, resulted in statistically similar values. The grain yields in these treatments were 10,762, 10,861, and 10,631 kg/ha, respectively.

These results suggest that the combination of 200 or 300 kg N/ha with potassium humate and Zn is effective in maximizing maize grain yields in the first season. However, in the second season, applying 400 kg of N/ha also produced comparable results. This indicates that there may be different optimal nutrient levels depending on the growing conditions and needs of the maize crop. Further research could help determine the most cost-effective and sustainable approach to achieving high grain yields consistently across seasons.

The relationship between potassium humate, Zn, and N is intricate and can significantly influence plant growth. Potassium humate has been shown to boost the absorption and utilization of Zn in plants, resulting in enhanced growth and yield (Ozkutlu *et al.*, 2006; Kumar and Singh, 2017; Rahi *et al.*, 2021). Furthermore, potassium humate can mitigate nutrient imbalances and deficiencies that may arise in the presence of N (Petrus *et al.*, 2010; El-Naqma, 2020; Aljoubory and Al-Yasari, 2023). On the other hand, N plays a role in facilitating the efficient uptake of Zn by plants and utilizing potassium humate for improved growth and development (Nawaz *et al.*, 2012; Kumar and Singh, 2017; Montoya *et al.*, 2020; Aljoubory and Al-Yasari, 2023). This interplay between these elements underscores the importance of understanding their interactions for optimizing plant health and productivity.

IV. CONCLUSION

The impact of potassium humate, zinc spray, and nitrogen fertilization on maize yield and its components in Kharga oasis, Egypt, was found to be significant. The study revealed that the combination of potassium humate and zinc spray resulted in higher yields compared to other treatments. Additionally, the application of 300 kg/ha of nitrogen fertilizer also contributed to increased yields. Interestingly, there was no significant difference in yield when different levels of nitrogen (100, 200, and 300 kg/ha) were combined with potassium humate and zinc spray. These findings suggest that maize plants exhibited a clear response to the applied nutrients. However, it is worth noting that the highest application of nitrogen fertilizer led to a lower grain yield, indicating a potential loss of nitrogen fertilizers.

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The Effect of NPK Fertilizer on the Growth and Yield of Sweet Corn (*Zea mays Saccharata* Sturt.) with Various Planting Arrangements

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Abstract— *Planting layout arrangements greatly influence the sweet corn population and crop yields per unit area. Planting layout arrangements are related to plant spacing, plant spacing that is too wide causes too little population, while plant spacing that is too tight results in competition between plants. Fertilization of macronutrients such as NPK is needed to meet the needs of plants in the maximum population. The research was carried out from May to July 2023 in Jetis Village, Jetis District, Ponorogo Regency, East Java Province, Indonesia. The experiment used a split-plot design with NPK fertilizer dosage as the main plot and planting pattern as a subplot. The NPK dosage consists of three levels, namely 200 kg ha^{-1}, 300 kg ha^{-1} and 400 kg ha^{-1}, while the planting pattern consists of four levels, namely single row, jajar legowo 3:1, jajar legowo 2: 1 and ring pit pattern. The research results show that differences in planting layout influence the microclimate around sweet corn plants which includes temperature, relative humidity and sunlight intensity. The ringpit planting layout shows a higher population and yield compared to other planting patterns. The single row planting pattern showed the lowest growth and yield of sweet corn. NPK fertilization with doses of 300 kg ha^{-1} and 400 kg ha^{-1} can increase the growth and yield of sweet corn plants.*

Keywords— ***Fertilizer, NPK, Planting system, Population density, Sweet corn***

I. INTRODUCTION

Sweet corn (*Zea mays saccharata* Sturt.) is a type of plant that is harvested young and widely cultivated in tropical areas. Not only in tropical regions, sweet corn is a popular vegetable in the USA, Australia and Canada. Sweet corn is very popular among people because it has a 25-30% sweeter taste than normal grain corn (Bhatt, 2012; Ugur and Maden, 2015). Sweet corn is a source of carbohydrates, protein, vitamins, minerals, phenolic acids, carotenoids and fiber. Regular consumption of sweet corn can reduce the severity of chronic diseases such as cardiovascular disease, eye disease, obesity, diabetes and digestive diseases (Sheng *et al*., 2018). Sweet corn also has an important role in meeting the nutritional needs of celiac sufferers, because sweet corn kernels do not contain gluten which is usually provided in the form of corn flour (Baranowska, 2023).

In Indonesia, the average sweet corn production is around 8.31 tons ha^{-1} and has a value that is very far from the potential yield of 14 tons ha^{-1} to 18 tons ha^{-1} (Pratama *et al*., 2022). Low production can be caused by various aspects, including cultivation factors, biotic factors and abiotic factors. Improvement efforts that can be made to increase production are expanding cultivation areas (extensification) as well as increasing land productivity and improving cultivation techniques (intensification) (Magezi *et al*., 2023). Intensification can be done by regulating planting patterns and managing adequate nutrition.

Setting planting patterns is related to plant spacing, plant population per hectare which will then influence the production produced per hectare (Bernhard and Below, 2020). Increasing population in a unit area can increase yields, but plant density that is too high reduces yields and

quality due to competition for environmental resources such as light, temperature, water and nutrients (Xue *et al*., 2016). As a C4 plant, corn is very susceptible to light stress and low temperatures (Bilska and Sowiński, 2010; Bellasio and Griffiths, 2014).

At maximum population conditions, nutrient management is needed so that plant nutrition is sufficient. Macro essential fertilizers that must be met for plant growth are N, P and K (Gul *et al*., 2015). N plays an important role in the synthesis of proteins, enzymes, DNA, RNA and chlorophyll (Hayat *et al*., 2010). P is needed in the formation of proteins and carbohydrates, DNA, RNA, forms ATP as an energy source and is important in the photosynthesis process, while K plays a role in osmoregulation, photosynthesis and induces sucrose content, soluble sugar content and starch content in corn plants (Pettigrew, 2008; Duncan *et al*., 2018; Yang *et al*., 2021). This research aims to obtain a model for proper plant layout and NPK fertilization to increase the productivity of sweet corn plants.

II. MATERIALS AND METHOD

2.1 Experimental detail

The research was carried out from May to July 2023 in Jetis Village, Jetis District, Ponorogo Regency, East Java Province, Indonesia. The research was carried out using a split-plot design, the main plot was the NPK fertilizer dose and the plant layout method was the subplot

The main plot is the NPK fertilizer dose which consists of three levels:

N1: NPK 200 kg ha^{-1}

N2: NPK 300 kg ha^{-1}

N3: NPK 400 kg ha^{-1}

The subplot consists of a plant layout method which consists of four levels:

P1 = Single row (95 plants per plot)

P2 = Jajar legowo 3:1 (246 plants per plot)

P3 = Jajar legowo 2:1 (270 plants per plot)

P4 = Ring pit (280 plants per plot)

This treatment combination was repeated 3 times to obtain 36 experimental plots.

Data were analyzed using ANOVA (Analysis of Variance) and carried out with the F test was at the 5% error level, if there is an effect continued HSD (honest significant difference) test at the 5% error level.

2.2 Field experiment

The experimental site is situated at an altitude of approximately 146 meters above sea level (masl), average air temperature of 21-31°C, and with Alluvial soil type. Planting sweet corn seeds is carried out using 4 plant layout methods according to treatment: a) Single row planting uses spacing 20x70 cm (population of 95 plants per plot), b) Jajar legowo 3:1 with a planting distance of 30x30x90 cm (population 246 plants per plot), c) Jajar legowo 2:1 with a planting distance of 20x30x90 cm (population 270 plants per plot), d) ring pit pattern, in one circle there are 8 planting holes with a planting distance between the circles of 60x70 cm (population 280 plants per plot). An illustration of the plant layout is presented in Figure 1.

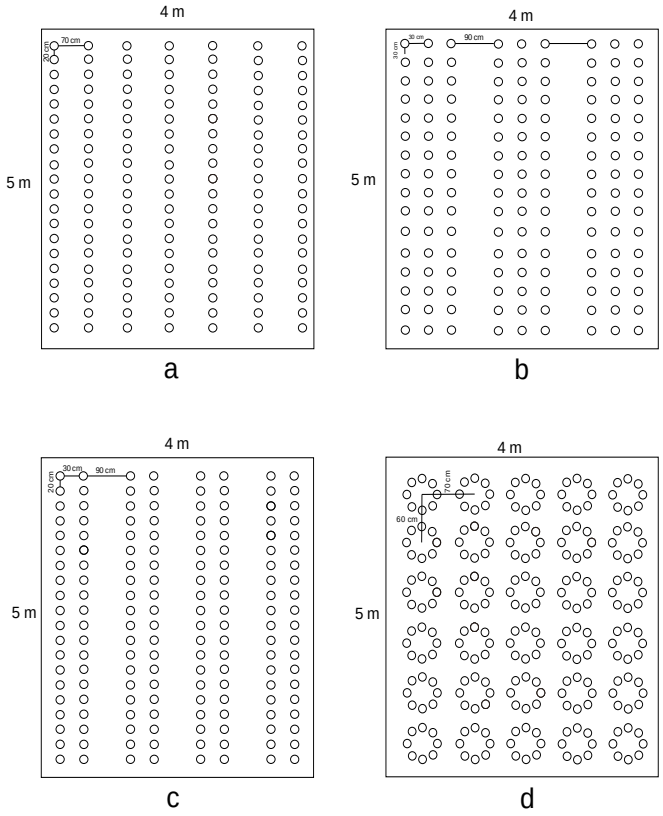

Fig.1: Sweet corn planting pattern: a. Single row, b. Jajar Legowo 3:1, c. Jajar Legowo 2:1, d. Ring pit pattern

Fertilizer application uses compound NPK 16:16:16 according to the dosage of each treatment. Maintenance activities include replanting, irrigation, weeding, hilling and controlling pests and diseases. Sweet corn harvesting is carried out 18-24 days after pollination. The age of a sweet corn plant is between 60-70 days at physiological maturity which is marked by the color of the corn hair starting to turn brown and drying out, the tip of the cob starting to be filled with corn kernels.

2.3 Measurement

The variables were observed in this research include growth variables, microclimate observations and yield variables.

2.3.1 Growth variables observed at age 56 DAP. Growth variables include :

1. Plant height

Observation of plant height is carried out by measuring the height from the base of the stem to the highest growing point of the plant

2. Number of leaves
   Counting the number of leaves is done by counting the leaves that have opened completely
3. Leaf area (cm^2)
   Leaf area was measured using the ALA method (*Average Leaf Area*) which can be done non-destructively (Widaryanto *et al*., 2019). The method used is to find the average estimated leaf area using a formula :

$$\overline{X} = \frac{(\textbf{Leaf area 1} / \varepsilon \textbf{ leaf 1}) + (\textbf{Leaf area 2} / \varepsilon \textbf{ leaf 2})}{\textbf{2}}$$

   To find out the average leaf area per plant use the formula:

$$\text{LA per plant} = \overline{X} \times \text{NL}$$

   Keterangan:
   $\overline{X}$ = Average area per leaf
   $\boldsymbol{\varepsilon}$ = Number of leaves per plant
   LA = Leaf area
   NL = Number of leaves per sample plant
4. Stem diameter
   Measurements were carried out on the second segment from the bottom of the soil using a caliper
5. Fresh weight per plant
   Fresh weight was observed by measuring the total fresh weight of the plant using a digital scale
6. Dry weight per plant
   Dry weight is obtained from the weight of samples that have been oven-treated for 48 hours at 80ºC.

2.3.2 Microclimate observations were observed at 14, 28, 42 and 56 HST. Microclimate observations include :

1. Temperature and relative humidity
   Temperature and relative humidity are measured using a thermohygrometer. Measurements on plants were carried out in the middle of the plant canopy during the day (12.00 am).
2. Intensity of solar radiation
   Measurement of the intensity of solar radiation was carried out at 12.00 am on plants including the top, middle and bottom of the plant canopy using a lightmeter (Sugito., 1999).

2.3.3 Yield variables are observed at harvest time at approximately 60-70 days after planting. Yield variables include :

1. Number of cobs per plot
   The number of cobs was calculated by counting all the corn cobs in each treatment plot with an area of 4 x 5 m
2. Length of the filled cob
   Measurements are made by measuring the length of the cob from the base to the tip of the cob using a tape measure
3. Cob diameter
   Measurements were made at the center of the corn cob using a caliper
4. Weight the cobs with husks
   Measurements were made by weighing the corn cobs and husks using a digital scale
5. Weight of cobs without husks
   The corn husks were peeled and then the weight of the cobs without the husks was weighed using a digital scale
6. Harvest index
   The harvest index is a value that describes the distribution of photosynthesis or plant biomass between the two parts of the plant that have been considered, namely the organs for photosynthesis and the organs of economic value. The harvest index using a formula :

$$\text{Harvest index} = \frac{Weight\ of\ cobs}{Total\ biomass}$$

III. RESULTS

3.1 Growth Observations

The effect of administering NPK fertilizer doses and planting layout methods on sweet corn plant growth variables is presented in Table 1. There is no interaction between the two treatments, but each factor has its own influence.

Table 1. Effect of NPK dosage and planting layout method on sweet corn plant growth components

Treatment	Plant height (cm)	Number of leaves (sheet)	Leaf area (cm^2)	Stem diameter (cm)	Fresh weight (g tan^{-1})	Dry weight (g tan^{-1})
NPK fertilizer dosage						
N1(NPK 200 kg ha^{-1})	181,25 a	11,58 a	1084,40 a	2,48	507,81 a	176,98 a
N2 (NPK 300 kg ha^{-1})	205,56 ab	12,42 b	1128,99 ab	2,56	585,80 ab	220,14 ab
N3 (NPK 400 kg ha^{-1})	223,54 b	12,77 b	1312,83 b	2,59	607,17 b	238,98 b

HSD 5%	35,90	0,803	187,6	ns	95,7	52,72
CV (%)	12,13	4,50	10,97	8,00	11,60	23,32
Planting layout method						
P1 (Single row)	192,50	12,11	994,38 a	2,60	510,35 a	192,26 a
P2 (Jajar legowo 3:1)	206,31	13	1118,42 a	2,58	458,82 a	163,78 a
P3 (Jajar legowo 2:1)	205,14	12,33	1143,79 a	2,48	579,86 ab	188,33 a
P4 (Ring pit)	209,86	12,00	1445,04 b	2,50	718,68 b	303,77 b
HSD 5%	ns	ns	227,6	ns	157,7	57,18
CV (%)	7,20	5,92	14,54	7,37	20,89	22,94

Remarks: The numbers followed by the same letter in the same column are not significantly different in the HSD test with a confidence level of 95%. HSD: Honest Significant Difference CV: Coefficient of variation

The variables of plant height and number of leaves were only influenced by NPK fertilization and were not influenced by differences in planting layout. The variables of leaf area, fresh weight per plant and dry weight per plant were influenced by the two treatments, namely the dose of NPK fertilizer and differences in planting layout. Neither the NPK fertilizer dose nor the planting layout method had any effect on the stem diameter of sweet corn plants. Providing NPK fertilizer at doses of 300 kg ha^{-1} and 400 kg ha^{-1} showed higher growth variables than the NPK dose of 200 kg ha^{-1}.

Different planting layout methods have no effect on plant height, number of leaves and stem diameter of sweet corn plants. The planting layout method influences the variables of leaf area, fresh weight and dry weight of the plant. The planting layout method using ring pits showed higher leaf area and plant dry weight compared to other treatments. In the variable fresh weight per plant, it shows that the 2:1 row legowo planting layout method and row rings show higher yields.

3.2 Microclimate observations

In this research, we studied how the planting layout method influences the microclimate. The components observed in microclimate observations are temperature, humidity and sunlight intensity (Figure 2). Observations of air temperature show that the temperature decreases as the age of the plant increases. Air temperature decreased drastically at the age of 42 WAP and 56 WAP plants. The jajar legowo 3:1 treatment had a higher air temperature compared to the other treatments. Lined ring treatments tend to have lower temperatures. Differences in air temperature between treatments can be related to canopy density which is formed due to differences in planting distance and plant density.

Observations of relative humidity show a graph that is inversely proportional to air temperature. When the temperature is high, the relative humidity is low, and conversely, if the temperature is low, the relative humidity becomes high. Relative humidity decreased at 28 WAP, and then continued to increase at 42 WAP and peaked at 56 WAP. The lined ring layout method tends to have higher relative humidity compared to other treatments. The planting layout method that shows low relative humidity is found in the row legowo 3:1 treatment.

The value of solar radiation intensity is inversely related to the graph of relative humidity observations. The intensity of solar radiation continues to increase until the age of 42 WAP and then decreases significantly at the age of 56 WAP. The jajar legowo 3:1 treatment showed higher intensity compared to other treatments. The lined ring treatment had the lowest intensity at 14 WAP and 56 WAP, but the intensity increased drastically at 28 WAP and 42 WAP. Air temperature and solar radiation intensity have a pattern that is directly proportional, and both have values that are inversely proportional to relative humidity. When the intensity of solar radiation increases, the air temperature will increase and the relative humidity will decrease.

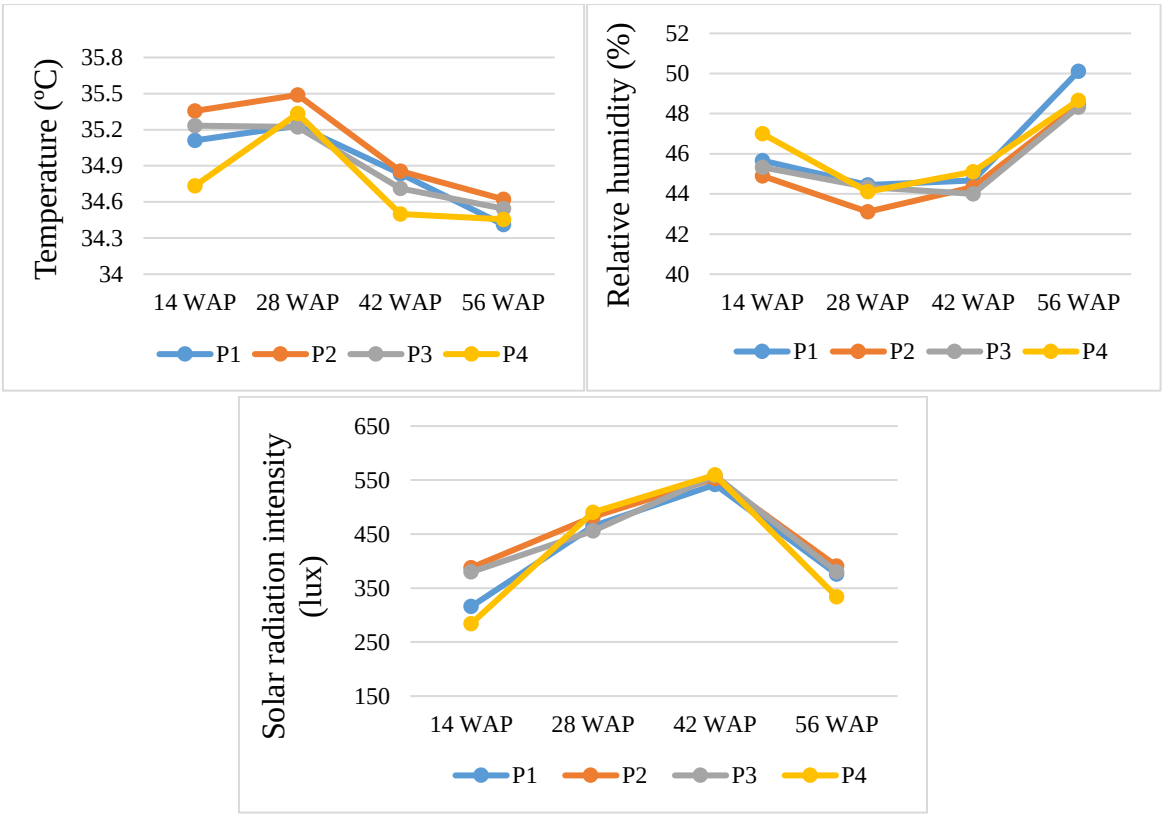

Fig.2: The effect of planting layout methods on air temperature, relative humidity and solar radiation intensity at each observation age

3.3 Yield Observations

The effect of administering NPK fertilizer doses and planting layout methods on sweet corn plants is presented in Table 2. There is no interaction between the two treatments, but each factor has its own influence.

Table 2. Effect of NPK dosage and planting layout method on sweet corn yield observations

Treatment	Number of cobs per plot	Length of cob contains (cm)	Cob diameter (cm)	Cob weight with husk (g)	Cob weight without husk (g)	Harvest index
NPK fertilizer dosage						
N1(NPK 200 kg ha^{-1})	48,92	17,86 a	4,76	257,34 a	177,33 a	0,51
N2 (NPK 300 kg ha^{-1})	50,50	19,38 ab	4,85	298,75 b	196,74 ab	0,50
N3 (NPK 400 kg ha^{-1})	50,08	22,73 b	5,13	295,18 b	203,93 b	0,52
HSD 5%	ns	3,668	ns	34,34	23,60	ns
CV (%)	15,96	12,61	8,05	8,32	8,42	20,83
Planting layout method						
P1 (Single row)	43,78 ab	18,39 a	5,05	245,635 a	176,111 a	0,44 a
P2 (Jajar legowo 3:1)	34,67 a	20,31 b	4,94	295,536 b	200,536 b	0,58 b
P3 (Jajar legowo 2:1)	53,22 b	20,46 b	4,76	278,492 ab	188,948 ab	0,49 ab
P4 (Ring pit)	67,67 c	20,81 b	4,90	315,357 b	205,071 b	0,53 ab
HSD 5%	10,56	1,384	ns	43,75	19,93	9,93
CV (%)	15,91	5,20	6,45	11,57	7,77	14,30

Remarks: The numbers followed by the same letter in the same column are not significantly different in the HSD test with a confidence level of 95%. HSD: Honest Significant Difference CV: Coefficient of variation

The dose of NPK fertilizer affects the yield variables of sweet corn plants which include the length of the filled cob, the weight of the cob with the husk and the weight of the cob without the husk. Differences in NPK fertilizer doses did not affect on the number of cobs per plot, ear diameter and harvest index. Giving NPK at a dose of 300 kg ha^{-1} and 400 kg ha^{-1} showed higher results compared to the NPK dose of 200 kg ha^{-1}.

The planting layout method does not affect the diameter of the cobs, but it does affect the number of cobs per plot, the length of the filled cobs, the weight of the cobs with husks, the weight of the cobs without husks and the harvest index. The planting layout with a lined ring pattern shows a higher number of cobs per plot compared to other planting patterns. Observing the length of the filled cobs, the weight of the cobs with husks, the weight of the cobs without husks and the harvest index showed that the planting patterns of jajar legowo 3:1, jajar legowo 2:1 and lined rings had values that were not significantly different. The pattern of planting row legowo 2:1 and single row had values that were not significantly different when observing the weight of cobs with husks and the weight of cobs without husks. When observing the harvest index of pattern of planting, single row planting, row legowo 2:1 and row rows have results that are not significantly different.

IV. DISCUSSION

Based on the data obtained, there was no interaction between the two treatments, however the use of NPK fertilizer and planting patterns had an effect on the growth and yield of sweet corn plants. In the treatments carried out in the research, the largest population was found in the ring pit planting layout (lined circles) with 280 plants per plot. That way, the yields produced in this layout show the highest. Research conducted on white corn shows that grain yield increases with plant density, double-row planting produces 12-18.22% higher yields compared to single-row (Akbar *et al*., 2016). Plant population greatly influences crop yields in unit area. Research conducted on peanut plants shows that planting patterns and plant density influence crop growth rate and harvest index (Rasekh *et al*., 2010).

Plant layout and plant density influence canopy architecture which then influences radiation interception and absorption of CO_2 which will be converted into carbohydrates. The higher the carbohydrates produced, the higher the harvest will be. The canopy formed due to differences in planting layout will affect the interception and utilization of solar radiation in corn plants, this will result in photosynthesis running less well and reducing grain yield (Valadabadi and Farahani, 2010; Duncan *et al*., 2018). A larger plant population, with appropriate layout arrangements and paying attention to canopy architecture will increase corn grain yield. Corn grain yield is influenced by the number of plants per unit area, the number of cobs per plant, the number of seeds per cob and the weight of seeds per ear (Gozubenli *et al*., 2004).

In a row planting layout, it is still possible for the corn plant canopy to grow freely and receive maximum sunlight. In a single row plant layout, the population produced per unit area is smaller than in other treatments. Apart from population differences, in a single row planting layout the empty space between corn plants is wider, this can result in a greater evapotranspiration rate and more water and nutrient loss. A high population will produce higher yields, but competition between plants for resources such as light, nutrients and water must be considered (Onat *et al*., 2017; Zhang *et al*., 2020).

The dose of NPK fertilizer greatly influences the growth and yield of corn plants. Giving NPK 200 kg ha^{-1} showed the lowest results in all variables. Giving NPK at a dose of 300 kg ha^{-1} and 400 kg ha^{-1} showed results that were not significantly different, therefore giving 300 kg ha^{-1} was more economically profitable to reduce input (cost). One of the nutrients contained in NPK fertilizer is nitrogen. Nitrogen has an important role in plant growth, is a protein constituent and has an important role in the photosynthesis process. Previous research examined the role of nitrogen in corn plants, giving the highest dose of urea, namely 520 kg ha^{-1}, showed higher results in the parameters of total fresh weight, relative growth rate, leaf area index and crop growth rate (Valadabadi and Farahani, 2010). Plant density and nitrogen fertilizer dosage influence the growth and yield of corn plants. Increasing the corn plant population of 88888 ha^{-1} with a fertilizer dose of 161 kg N ha^{-1} can increase grain yield by 65.16% compared to a population of 44444 ha^{-1} with 92 kg N ha^{-1} (Zeleke *et al*., 2018).

Apart from nitrogen, fulfilling the nutrient element phosphorus in corn plants significantly increases the leaf area index, crop growth rate, dry weight and yield of corn plants (Amanullah *et al*., 2010). An increase in the leaf are index value is related to an increase in leaf area, where a wider leaf area will maximize the absorption of PAR (Photosynthetically Active Radiation). Potassium also has an important role in the process of photosynthesis and absorption of CO_2. In conditions of potassium deficiency, photosynthetic activity decreases due to leaf chlorosis due to the accumulation of reactive oxygen species (ROS) (Du *et al*., 2019). Not only growth, potassium plays a role in improving the quality of sweet corn through sugar transportation. Potassium is needed by plants in the process of photosynthesis, ATP production, sugar translocation,

starch production in grains, nitrogen fixation and protein synthesis (Wolde, 2016).

To maximize yield potential and land use efficiency, planting patterns can be carried out using an intercropping system. For further research, it is necessary to study in more depth the sweet corn intercropping system combined with undercrops such as legumes. Setting the planting pattern of corn plants intercropped with soybeans affects the uptake and distribution of nutrients such as nitrogen (N), phosphorus (P) and potassium (K) (Raza *et al*., 2019). Nitrogen fertilization and intercropping of corn with alfalfa showed an increase in photosynthetic characteristics, photosynthetic nitrogen utilization efficiency (PNUE) and yield up to 25% (Nasar *et al*., 2021).

V. CONCLUSION

This research shows that the ring pit planting layout method produces higher growth and yields in sweet corn. In addition, the ring pit planting layout produces the largest population. To meet nutrient needs due to population changes, an NPK fertilizer dose of 300-400 kg ha^{-1} is required.

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Perception of Agroecological Practices by Farmers in the Sub-Prefectures of n'ganon, Niofoin and Sirasso in the Department of Korhogo (Ivory Coast)

Perception des Pratiques Agroécologiques par les Exploitants Agricoles des Sous-Préfectures de n'ganon, Niofoin et Sirasso Dans le Département de Korhogo (Côte D'ivoire)

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Abstract— *A survey was carried out in villages in the sub-prefectures of N'ganon, Niofoin and Sirasso in the north of Côte d'Ivoire in order to understand the perception of agroecological practices by farmers. The target population consists of 150 individuals including 94 men and 56 women. The results of this survey showed that the majority of farmers are illiterate (86.67%). Farmers are mainly men (62.67%□). Then, the farmers surveyed in the three sub-prefectures are 58% engaged in agriculture and 42% practice agriculture and livestock farming. Agroecological practices the most applied by farmers in the three northern study sub-prefectures are: rotational crops (100%), the shrub-crop association (92%) and the sole crop of legumes (78.67%) Indeed, the farmers of the three localities have agricultural practices based on the overexploitation of natural resources, the intensive use of pesticides and chemical fertilizers. These farmers have a lack of knowledge of agroecological practices. As a result, agroecological practices are very important and deserve the particular attention of agricultural policy makers in general, officials in charge of agriculture and the environment, farmers and researchers in agriculture. particular to preserve the environment, food and health security, and for sustainable agriculture.*

Keywords— ***Understanding, Practice, Agroecological, Farmer***

Résumé— *L'agroécologie est au cœur des débats de nos jours pour donner une orientation aux systèmes agricoles qui doivent être performants et durables. En Afrique subsaharienne, des pratiques relevant de l'agroécologie peuvent aider à une agriculture durable dans cette partie de l'Afrique. Une enquête a été réalisée dans des villages des sous-préfectures de N'Ganon, Niofoin et Sirasso au nord de la Côte d'Ivoire*

en vue de comprendre la perception des pratiques agroécologiques par les exploitants agricoles. La population cible est constituée de 150 individus dont 94 hommes et 56 femmes. Les résultats de cette enquête ont montré que les exploitants agricoles sont en majorité analphabètes (86,67 %). Les exploitants agricoles sont essentiellement des hommes (62,67 %). Ensuite, les exploitants enquêtés des trois sous-préfectures s'adonnent à 58 % à l'agriculture seule et 42 % pratiquent l'agriculture et l'élevage. Les pratiques agroécologiques les plus appliquées par les exploitants agricoles dans les trois sous-préfectures d'étude du nord sont : les cultures en rotation (100 %), l'association arbustes-cultures (92 %) et la culture pure de légumineuses (78,67 %). En effet, les exploitants agricoles des trois localités ont des pratiques agricoles fondées sur la surexploitation des ressources naturelles, l'utilisation intensive des pesticides et d'engrais chimiques. Ces exploitants agricoles ont une méconnaissance des pratiques agroécologiques. En conséquent, les pratiques agroécologiques sont très importantes et méritent une attention particulière des décideurs politiques agricoles en général, des responsables en charge de l'agriculture et de l'environnement, des agriculteurs et des chercheurs en particulier pour préserver l'environnement, la sécurité alimentaire et sanitaire, et pour une agriculture durable.

***Mots-clés*— Compréhension, Pratique, Agroécologique, Exploitant agricole**

I. INTRODUCTION

Aujourd'hui, l'agriculture fait face à de nouveaux enjeux : augmentation des besoins de l'alimentation humaine et animale dûe à la croissance démographique d'une part, d'autre part à de nouvelles exigences de la société qui imposent une production agricole garantissant la qualité nutritionnelle et le respect de l'environnement **(Trabelsi, 2018)**.

Durant la dernière décennie, une importante production scientifique et technique a été consacrée à des modèles d'agriculture plus économes en intrants et moins polluante dont les fondements s'inspirent du fonctionnement des écosystèmes naturels **(Wezel *et al.*, 2009).** Les termes ″agroécologie, agriculture de conservation, permaculture, agriculture biologique, agriculture raisonnée et écologisation des pratiques″ sont ainsi rentrés dans le langage commun des agronomes, des décideurs et des agriculteurs. Ces nouveaux modèles de production agricole et de consommation alimentaire cherchent à répondre, pour les prochaines décennies, aux défis de la sécurité alimentaire (quantitative et qualitative) et de la protection de l'environnement **(Dugué *et al.*, 2014)**.

Ainsi, des structures de recherche nationale et Internationale (CNRA, CIRAD AGRISUD, FAO, AFDI...) sont-elles mobilisées pour réduire la vulnérabilité des exploitations agricoles familiales, notamment face aux contraintes environnementales et économique **(Diouf et Dieng, 2015)**. C'est dans cette optique que la FAO a organisé, en 2014, un Symposium international sur l'agroécologie pour la sécurité alimentaire et la nutrition **(FAO, 2014).** L'agroécologie apparait ainsi comme une solution pour une agriculture durable et protéger l'environnement **(Altieri, 2002 ; Francis *et al.*, 2003)**.

Cependant, il se pose toujours le problème du changement climatique dû à une destruction de la faune et de la flore en Côte d'Ivoire précisément dans le département de Korhogo. Alors, des études ont été menées au nord de la Côte d'Ivoire dans le but de connaitre le niveau de connaissance des pratiques agroécologiques par les exploitants agricoles. Dès lors, comment les exploitants agricoles dans le nord de la Côte d'Ivoire perçoivent ils l'usage des pratiques de l'agroécologie ? Et quels sont les systèmes agricoles des exploitants agricoles ?

L'objectif de cette étude est de comprendre le niveau de connaissance des pratiques agroécologiques par les exploitants agricoles dans les sous-préfectures de N'Ganon, Niofoin et Sirasso du département de Korhogo. Plus spécifiquement, il s'agit d'inventorier les pratiques agroécologiques des exploitants agricoles, d'analyser les pratiques agroécologiques des exploitants agricoles et identifier les contraintes liées aux pratiques agroécologiques auxquelles sont confrontés les exploitants agricoles.

II. MATERIEL ET METHODES

2.1.Matériel

Cette étude a été menée auprès de 150 exploitants agricoles dans les sous-préfectures de N'Ganon, Niofoin et Sirasso (département de Korhogo) dont 50 exploitants par sous-préfectures. L'enquête a été menée à l'aide d'un questionnaire élaboré comme outil de travail. Il a permis de collecter des données du terrain auprès des exploitants agricoles des différentes localités. Un appareil photo numérique de marque Sony a été utilisé pour la prise de photos. Le logiciel EXCEL a servi au traitement des données et le tracé des graphiques.

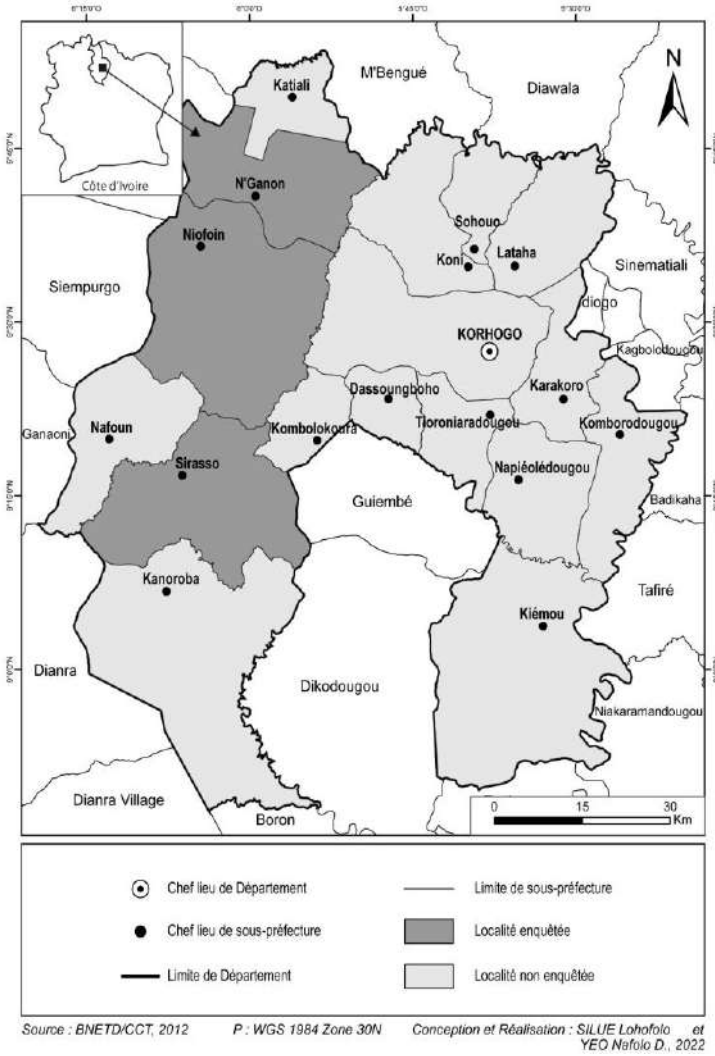

Fig.1 : Carte du département de Korhogo

2.2. Méthodes

Échantillonnage

Ces travaux ont débuté le 29 avril **et** ont pris fin le 9 juillet 2022**.** Les enquêtes ont été menées auprès d'un échantillon de 150 exploitants agricoles, à raison de 50 par localité. Chaque localité comprend plusieurs villages. Par ailleurs, pour atteindre les objectifs visés 11 villages ont été choisis dans la sous-préfecture de Niofoin (Niofoin, M'bia, Kamara, Kamanhan, Pivonhon, Tangafla, Tarato, Yoro, Tolma, Ogari et Djougoiblé), 8 villages dans la sous-préfecture de Sirasso (Sirasso, Lopin, Dokaha, Dagba, Nouhouo, M'balla, Sakpélé et Talléré) et 4 villages dans la sous-préfecture de N'Ganon (N'Ganon, Sakouma, Sindiré et Katchaga).

Ces villages ont été choisis en raison de leur accès facile et des données utiles pour l'article. L'enquête a porté essentiellement sur l'exploitant et son système d'exploitation agricole. Sur l'ensemble des exploitants agricoles des trois sous-préfectures, 94 hommes et 56 femmes ont été interrogés dont 29 hommes et 21 femmes dans la sous-préfecture de Sirasso, 33 hommes et 17 femmes dans la sous-préfecture de Niofoin et 32 hommes et 18 femmes dans la sous-préfecture de N'Ganon **(tableau II)**. Le choix du nombre de personnes par genre s'est fait de manière aléatoire. Ensuite l'interview des exploitants agricoles, a été faite en fonction de leur disponibilité. L'enquête a été conduite sous forme d'interviews individuelles.

Enquête et collecte des données

L'enquête s'est faite en deux phases. La première phase est la pré-enquête qui a consisté à visiter chaque zone d'étude, afin de prendre connaissance avec les différentes autorités administratives et coutumières. La seconde phase est l'enquête proprement dite c'est à dire à soumettre les questionnaires aux différents exploitants agricoles. Le questionnaire porte sur des caractéristiques

sociodémographiques de l'exploitant et son exploitation à savoir : l'identification de l'exploitant (la situation matrimoniale, le genre, l'âge, le niveau d'alphabétisation), son système d'exploitation (surface, système de culture, taille des superficies). La superficie moyenne de l'exploitation a été déterminée avec la formule suivante :

$$X = \frac{1}{N}\sum_{i=1}^{\infty}(xi)$$

N : nombre d'exploitant par sous-préfecture

Xi : superficie par exploitant

Traitement et analyse statistique des données

Après le dépouillement manuel des fiches d'enquête, les résultats ont été analysés à l'aide des statistiques descriptives. Les programmes de bureautique que sont Excel et Word ont été utilisés pour le traitement des données et du texte. Le logiciel Excel a servi à calculer les différentes proportions et à réaliser des représentations graphiques (histogrammes et diagrammes). Le test de Ki2 a permis de comparer les proportions.

Tableau I: Présentation du nombre d'enquêté par genre et par sous-préfectures.

Sous-prefectures	Nombre de villages	Nombre des enquêtés par sous-préfecture		
		Hommes	Femmes	Total des enquêtés
N'ganon	4	32	18	50
Niofoin	11	33	17	50
Sirasso	8	29	21	50
Total	23	94	56	150

III. RÉSULTATS ET DISCUSSION

3.1 Résultats

- Caractéristiques sociodémographiques des exploitants agricoles des trois sous-préfectures

La figure 2 présente la répartition des exploitants agricoles des trois sous-préfectures en fonction du genre. Ces résultats ont montré que sur un échantillon de 150 exploitants agricoles interrogés, 62,67 % sont des hommes et 37,33 % de femmes. Dans la sous-préfecture de N'ganon, un taux de 64 % d'hommes contre 36 % de femmes ; 66 % d'hommes dans la sous-préfecture de Niofoin contre 34% de femmes et 58 % d'hommes contre 42 % de femmes dans la sous-préfecture de Sirasso ont été enregistrés**.** Le tableau III présente la répartition des exploitants par tranche d'âge par sous-préfecture. Ces résultats ont montré que sur 150 exploitants agricoles enquêtés, 78 % ont un âge compris entre 30 à 49 ans. Les exploitants agricoles ayants un âge compris de 18 ans et 29 ans représente 14,66 % de l'échantillon et 7,33 % des exploitants agricoles ont un âge supérieur ou égal à 50 ans.

Les résultats de l'enquête montrent que les exploitants agricoles enquêtés dans les trois sous-préfectures ne sont pas instruits avec des 82 % dans la sous-préfecture de N'ganon, 90 % dans la sous-préfecture de Niofoin et 88 % dans celle de Sirasso. Ceux qui ont un niveau primaire représentent 16 % à N'ganon, 8 % à Niofoin et 10 % à Sirasso. Dans chaque localité ceux qui ont un niveau secondaire représentent 2 %. Aucun agriculteur n'a le niveau supérieur dans chacune des trois localités (figure 3).

La figure 4 présente la répartition des exploitants agricoles des trois sous-préfectures en fonction de la situation matrimoniale. Sur un échantillon de 150 exploitants agricoles, 96,67 % vivent en couple, tandis que 3.33 % des exploitants sont célibataires.

Le tableau IV présente les fréquences des ménages des exploitants agricoles enquêtés dans les trois sous-préfectures. Dans les trois sous-préfectures d'études, la taille moyenne des ménages est de 8 individus.

Sur 150 exploitants agricoles enquêtés, ceux ayant une charge comprise entre 1 et 9 personnes représentent 75,33 % de l'échantillon et 24,67 % ont un ménage compris entre 10 et 20 personnes.

L'analyse des résultats montre que sur les 150 enquêtés, 58 % des exploitants agricoles pratiquent uniquement l'agriculture dans les sous-préfectures de N'ganon, Niofoin et 56 % dans la sous-préfecture de Sirasso ; 42 %des exploitants pratiquent des activités mixes (agriculture-élevage) **(Tableau V)**.

-Taille des surfaces cultivées par les exploitants agricoles des trois sous-préfectures

Les résultats de l'enquête enregistrés sur la taille des surfaces cultivées par les exploitants agricoles dans les trois sous-préfectures d'étude, indiquent que la taille moyenne des surfaces cultivées est de 11,13 hectares. De plus, sur les 150 personnes enquêtées, 52,67 % ont des exploitations agricoles qui vont de 1 à 9 hectares. Toutes les femmes (100 %) enquêtées ont des parcelles comprises

entre 1 et 9 hectares dans toutes les localités et 24,47 % d'hommes enquêtés selon les informations recueillies ; 75,53 % d'hommes ont des exploitations supérieures à 9 hectares selon les exploitants **(tableau VI).**

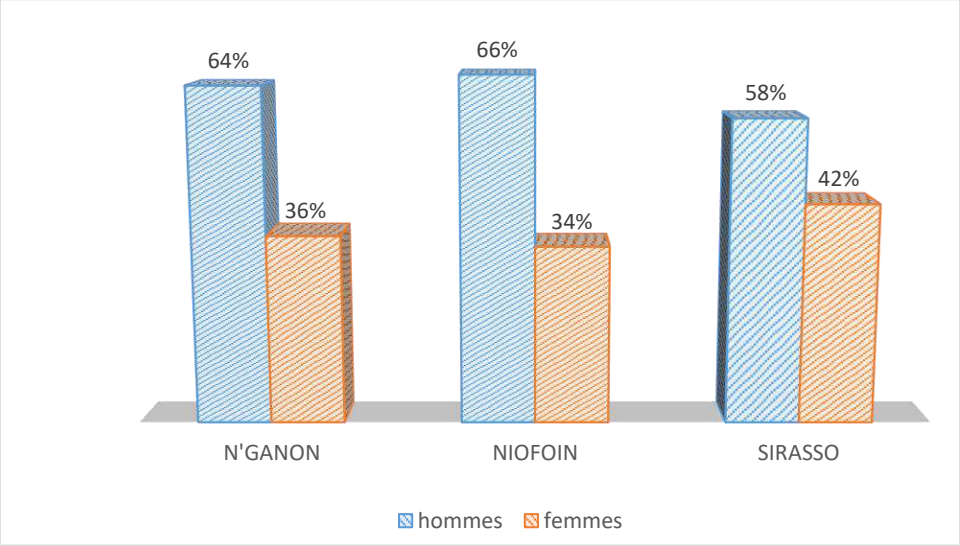

Fig.2: Répartition des exploitants agricoles des trois sous-préfectures en fonction du genre

Tableau II: Répartition des exploitants par tranche d'âge par sous-préfecture

Tranches d'âge (ans)	Proportion par sous-préfecture			
	N'GANON	NIOFOIN	SIRASSO	Total
[18-30[	6	26	12	14,67
[30-50 [	88	66	80	78
[50 et plus [	6	8	8	7,33
Total	100	100	100	100

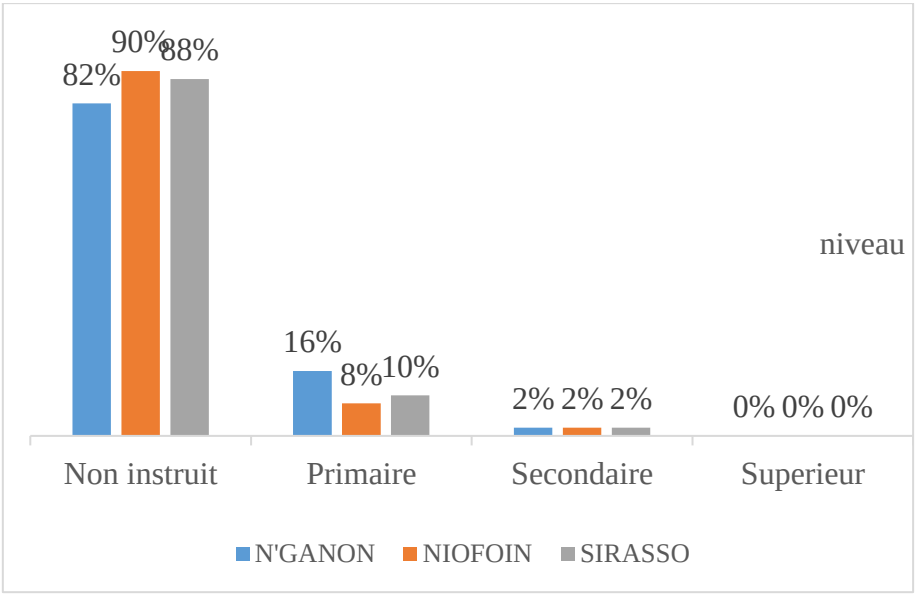

Fig.3 : Répartition des exploitants agricoles enquêtés en fonction du niveau d'alphabétisation

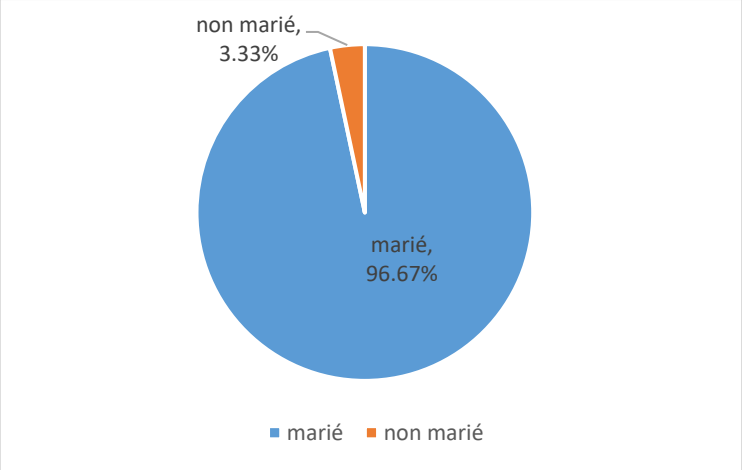

Fig.4 : Répartition des exploitants agricoles enquêtés en fonction la situation matrimoniale

Tableau III: Tailles des ménages des exploitants agricoles enquêtés par sous-préfecture

Taille de ménage	N'ganon	Niofoin	Sirasso	Total	Proportion
[1-10[	36	39	36	113	75,33
]10-20]	14	11	14	37	24,67
Total	50	50	50	150	100

Tableau IV: Répartition des exploitants agricoles en fonction du type d'activité par sous-préfecture

Sous-préfectures	Exploitants agricoles	Proportion (%)
N'ganon	Agriculteurs	58
	Agriculteurs-éleveurs	42
Niofoin	Agriculteurs	58
	Agriculteurs-éleveurs	42
Sirasso	Agriculteurs	56
	Agriculteurs-éleveurs	44

Tableau V: Répartition des parcelles exploitées par genre et par sous-préfecture.

Sous-préfectures	N'ganon		Niofoin		Sirasso		Total
Surfaces (ha)	Hommes	Femmes	Hommes	Femmes	Hommes	Femmes	
1-9	12	18	8	17	3	21	79
10-20	12	0	18	0	12	0	42
21-30	8	0	7	0	14	0	29
Total	32	18	33	17	29	21	150

3.1.3. Quelques exemples des pratiques agroécologiques en application

Les figures 5 et 6 montrent un sol où des résidus de récolte sont enfouies par labour et un sol où la fumure est simplement épandue afin d'améliorer la fertilité du sol dans la sous-préfecture de N'ganon. La culture de laitue et chou sont associées sur une même parcelle (figure 7).

3.1.4. Perception paysanne des pratiques agroécologiques par exploitants agricoles des trois sous-préfectures

Les résultats de l'enquête montrent que tous les exploitants agricoles (100 %) pratiquent la culture en rotation sur la même parcelle d'une saison à une autre ; 95,74 % hommes et 85,71 % de femmes pratiquent l'association arbustes-cultures (cultures pérennes de moins de 4 ans et cultures annuelles). La culture d'arachide uniquement (légumineuses) est réalisée à 79,79 % par les hommes et 76,78 % par les femmes dans les trois sous-préfectures d'étude (annexe). En plus, 91,07 % de femmes appliquent le paillage contre 8,51 % d'hommes. Cependant, 7,44 % d'hommes et 3,57 % associent les cultures annuelles et cultures pérennes (plantes de services) âgées de 4 ans et plus. Aucun exploitant agricole n'utilise des bio-pesticides (feuilles de papayers, neem, piment, ail) et de bande enherbée (planter des rangs d'arbres, herbes pour lutter contre le splash) (tableaux VI). Les tableaux VIII, IX et X montrent des contraintes pour les pratiques agroécologiques dans trois sous-préfectures.

Fig.5: résidus enfouis par labour dans un sol à N'ganon

Fig.6: fumure organique appliquée sur sol à Tolma (s/p Niofoin)

Fig.7: association de laitue et de chou

Tableau VI: Proportions des pratiques agroécologiques par genre par sous-préfecture

Pratiques agroécologiques	N'ganon		Niofoin		Sirasso		TOTAL	
	M	F	M	F	M	Fs	M	F
Culture d'arachide (légumineuses) (%)	81,25	72,22	75,75	70,59	82,76	85,71	79,79	76,78
Association céréales-légumineuses (arachide-maïs) (%)	43,75	83,33	21,21	64,70	89,65	100	50	83,93

Association de cultures pérennes (+ 4 ans) et annuelles (%)	3,12	0	18,18	5,88	0	4,76	7,44	3,57
Association de cultures pérennes (- 4 ans) et annuelles (%)	100	88,88	87,87	82,35	100	85,71	95,74	85,71
Application de fumure organique (%)	37,5	61,11	72,72	52,94	65,51	57,14	58,51	57,14
Paillage (%)	18,75	100	3,03	88,23	3,45	85,71	8,51	91,07
Dispositifs antiérosifs	53,12	100	42,42	5,88	10,34	71,43	36,17	60,71
Bande enherbée (%)	0	0	0	0	0	0	0	0
Jachère (%)	75	5,55	63,63	64,70	20,69	90,47	54,25	55,35
Enfouissement des résidus par labour (%)	78,12	16,16	60,60	52,94	79,31	57,14	72,34	42,86
Utilisation biopesticides (%)	0	0	0	0	0	0	0	0
Culture en rotation (%)	100	100	100	100	100	100	100	100

Tableau VII: Quelques contraintes des pratiques agroécologiques dans les sous-préfectures de N'ganon, Niofoin, Sirasso

Pratiques	N'ganon	Nofoin	Sirasso
Association céréales-légumineuses	Augmentation du travail de préparation du sol et de récoltes; Risque de compétition entre les espèces cultivées, Risque d'amensalisme entre certaines espèces; Augmentation de la complexité de gestion du système.	Augmentation du travail de préparation du sol et de récoltes ; Risque de compétition entre les espèces cultivées, Risque d'amensalisme entre certaines espèces; Augmentation de la complexité de gestion du système ; Méconnaissance des semences diversifiées.	Augmentation du travail de préparation du sol et de récoltes; Risque de compétition entre les espèces cultivées, Risque d'amensalisme entre certaines espèces; Augmentation de la complexité de gestion du système.
Enfouissement des résidus par labour	Nécessite beaucoup de travail pour couper et intégrer les résidus au sol sans machinerie; Connaissances scientifiques et techniques faibles; Performances variables dépendamment des cultures utilisées;	Nécessite beaucoup de travail pour couper et intégrer les résidus au sol sans machinerie ; Connaissances scientifiques et techniques faibles ; Performances variables dépendamment des cultures utilisées; Disponibilité et accès aux semences	Nécessite beaucoup de travail pour couper et intégrer les résidus au sol sans machinerie; Connaissances scientifiques et techniques faibles ; manque d'équipement de travail.

Paillage	Peut contenir une variété de ravageur, de pathogènes; Compétition entre la culture de couverture et la culture principale; Compétition pour les différents usages des paillis (alimentation animale, fourrage, termites); Difficulté d'accès à la paille ou des semences de plantes de couverture.	Augmentation du temps de travail ; Peut contenir une variété de ravageur, de pathogènes ; Compétition entre la culture de couverture et la culture principale ; attire les termites, les rats et autres rongeurs ; Difficulté d'accès à la paille ou des semences de plantes de couverture	Peut contenir une variété de ravageur, de pathogènes; Compétition entre la culture de couverture et la culture principale; Compétition pour les différents usages des paillis (alimentation animale, fourrage, termites); Difficulté d'accès à la paille ou des semences de plantes de couverture
Utilisation bio-pesticides	Manque de connaissances scientifiques et techniques; Disponibilité et accès restreint selon le milieu; Coûts; Potentiels risques d'introduction d'espèces tout dépendant l'état des organismes; Potentiels effets délétère sur des organismes non-ciblés.	Demande beaucoup de gestion et de connaissances de l'agroécosystème local; Disponibilité et accès restreint selon le milieu; Coûts; Potentiels risques d'introduction d'espèces tout dépendant l'état des organismes; Potentiels effets délétère sur des organismes non-ciblés.	Demande beaucoup de gestion et de connaissances de l'agroécosystème local; Disponibilité et accès restreint selon le milieu; Coûts; Potentiels risques d'introduction d'espèces tout dépendant l'état des organismes; Potentiels effets délétère sur des organismes non-ciblés.
Bande enherbée	Risque de créer des habitats pour les ravageurs; Perte d'espace pour les cultures.	Risque de créer des habitats pour les ravageurs; Perte d'espace pour les cultures.	Risque de créer des habitats pour les ravageurs; Perte d'espace pour les cultures.

2.2. Discussion

L'étude de la perception des pratiques agroécologiques a révélé que les exploitants agricoles des trois sous-préfectures sont à 62,67 % d'hommes contre 37,33 % de femmes. Cette même observation a été faite dans chaque sous-préfecture avec une proportion de 64 %, 66 % et 58 % d'hommes respectivement à N'ganon, Niofoin et Sirasso contre 36 %, 34 % et 42 % de femmes. Ces résultats peuvent s'expliquer par le fait qu'en milieu rural en Afrique , le champ appartient à la famille et est placé sous la responsabilité de l'homme. Ces résultats concordent avec ceux obtenus lors d'une étude à Sirasso par **Tuo (2018),** dans laquelle il a révélé que l'agriculture est majoritairement pratiquée par les hommes surtout la culture du coton.

Les résultats de l'étude révèlent que les exploitants agricoles des trois localités d'étude sont des adultes (78 %) et ont un âge compris entre 30 et 49 ans et 14,67 % sont des jeunes ayant un âge qui varie entre 18 et 29 ans. Quant aux plus âgés (50 ans et plus), ils ont une proportion de 7,33 %. Ces mêmes observations ont été faites dans chacune des trois localités. Ces résultats ne concordent pas avec ceux de **Bengaly (2010)** dans lequel il affirme que la plupart de la population rurale est jeune avec une nette domination de la population féminine. Cette différence peut s'expliquer par la politique de scolarisation de toutes les jeunes filles, décidée par l'Etat de Côte d'Ivoire depuis 2011.

Concernant le niveau d'instruction, les résultats de l'étude montrent que les exploitants agricoles ont un niveau d'instruction faible. Les exploitants enquêtés non scolarisés ont une forte proportion soit 86,67 %. Seulement 19,33 % d'entre eux sont instruits dont 17,33 % ont un niveau primaire et 2 % ont un niveau secondaire. Les résultats sont identiques dans les trois sous-préfectures. Cela peut rendre difficile la tâche des agents chargés d'encadrer et de

communiquer des directives techniques à la population rurale. Ces résultats sont conformes à ceux de **Tuo (2018).** En effet, la population est en générale analphabète.

Quant à la situation matrimoniale, la majorité des exploitants agricoles (96,67 %) vivent en couple seuls 3,33 % sont célibataires. Ce qui pourrait entraîner l'accroissement rapide de la population humaine d'où la disparition des ressources naturelles telles que la faune et la flore. Ces résultats ont été affirmés par les paysans qui disent que lorsque tu possèdes de très grandes parcelles, tu pourras prendre en charge toute ta famille.

L'analyse des tailles des ménages a été effectuée en deux tranches. Celle de 1 à 9 individus par famille représente 75,33 % et celle de 10 à 20 membres par famille représente la proportion de 24,67 %. Cela peut s'expliquer par une main d'œuvre familiale potentielle pour l'agriculture. Nos résultats concordent avec ceux obtenu par le ministère de l'Agriculture lors du recensement de l'exploitant et des exploitations agricoles (**2017)** qui a montré que la moyenne nationale des personnes par ménage agricole est de 7,2.

Concernant les activités des exploitants enquêtés des localités de N'ganon et Niofoin, 58 % sont des agriculteurs et 42 % sont à la fois agriculteurs et éleveurs. Dans la sous-préfecture de Sirasso, 56 % des exploitants sont des agriculteurs et 44 % sont des agro-éleveurs. Ces résultats sont identiques dans les trois localités. L'on constate que dans les trois localités d'études, il n'y a pas d'éleveurs proprement dit. Les agriculteurs sont aussi des éleveurs. Leurs animaux sont conduits de façon traditionnelle. Ce résultat concorde avec ceux de **Ouattara (2018**) qui montrent que les éleveurs des petits ruminants de la région du Poro sont à 75 % des agro-éleveurs.

L'étude a dévoilé que la taille moyenne des exploitations agricoles est de 11,13 hectares par personne. Cela peut rendre difficile la mise en œuvre des pratiques agroécologiques. Toutes les femmes enquêtées (100 %) ont des exploitations agricoles comprises entre 1 et 9 hectares. Cela peut s'expliquer par le fait que les femmes éprouvent beaucoup de difficultés pour accéder à la terre. Ces résultats concordent avec ceux de **Kouassi (2019)** qui montrent que l'accès aux ressources de production telle que la terre est souvent difficile aux femmes au nord de la Côte d'Ivoire du fait des exigences culturelles.

Selon cette étude, tous les exploitants agricoles (100 %) appliquent la culture en rotation ; 95,74 % hommes et 85,71 % de femmes associent les cultures pérennes de moins de 4 ans et cultures annuelles dans les trois localités du nord. La culture pure d'arachide est cultivée à 79,79 % par les hommes enquêtés et 76,78 % par les femmes enquêtées. Ce qui peut s'expliquer par le fait que les exploitants agricoles s'adaptent facilement à ces pratiques agricoles.- Ajouter à cela la pénibilité à la mise en œuvre de certaines pratiques agricoles pour des cultures qui nécessite une main d'œuvre masculine en Afrique. En plus, 91,07 % de femmes appliquent le paillage contre 8,51 % d'hommes. Cette pratique est appliquée dans les cultures maraîchères. Ce qui peut s'expliquer le fait que les maraîchers sont cultivés par les femmes.

Cependant, 7,44 % d'hommes et 3,57 % associent les cultures annuelles et cultures pérennes âgées de 4 ans et plus et aucun exploitant agricole n'utilise des biopesticides et de bande enherbée. Ce qui peut s'expliquer par une méconnaissance des pratiques agroécologiques dans les trois sous-préfectures et par la grandeur des surfaces cultivées. Cette méconnaissance est observée dans les trois sous-préfectures.

IV. CONCLUSION

Au terme de cette étude, il ressort que les exploitants agricoles des sous-préfectures de N'ganon, Niofoin et Sirasso dans le département de Korhogo ont des pratiques agricoles fondées sur la surexploitation des ressources naturelles, l'utilisation intensive des pesticides et des engrais chimiques. Ils ont donc une méconnaissance des pratiques agroécologiques. Ils pratiquent encore l'agriculture traditionnelle. Par ailleurs, ils sont confrontés à de nombreuses contraintes liées à la mise en œuvre des pratiques agroécologiques dans toutes les localités.

Pour mieux approfondir cette étude, il sera intéressant d'innover les pratiques agroécologiques, faire des études expérimentales avec les paysans dans le nord de la Côte d'Ivoire afin de les amener à comprendre l'importance des pratiques agroécologiques dans l'agropastoralisme et de continuer la collecte de données dans le district des savanes (nord de la Côte d'Ivoire).

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Effect of salinity on Growth and Secondary Metabolites of Sesbania Grandiflora seedlings: An Analytical Study

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Abstract— *Plants can undergo different types of stresses like biotic and abiotic stresses. Plants develop mechanisms to overcome these threats of biotic and abiotic stresses, which influence secondary metabolite production, enhancing the potential to overproduce useful phytochemicals for varied applications. Salt stress has a major impact on Agriculture. It results in losses of 806.4 billion rupees per year due to low production yield. Salinity affects plants in various aspects including water stress, ion toxicity, nutritional imbalance, oxidative stress, metabolic processes, membrane disorganization, cell division, and genotoxicity. Using in vitro plant cell culture to produce chemicals and pharmaceuticals has made great strides. A soft wooded tree, Sesbania grandiflora has many significant medicinal values that have been proven scientifically. It is known to work as an immune modulator along with the property of nitrogen fixation. A study was conducted to determine the effect of NaCl salinity on growth, morphological analysis, and phytochemical content in Sesbania grandiflora using in vitro method. Seeds of Sesbania grandiflora germinated on 10, 20, and 30mM NaCl salt concentrations for varying durations, and total flavonoid and phenolic content production was calculated to observe the correlation between salt stress and phenolic production. Thus, the salt stress effect and its effect on the phytochemicals were observed. The present study findings provide additional information for the accumulation of phytochemical compounds under salt stress in the plant which might help to explain the increased levels of secondary metabolites in other plant species.*

Keywords— ***Phenolics, Quantitative analysis, Salinity, Salt Stress, Sesbania grandiflora***

I. INTRODUCTION

Salinity is a major factor that affects crop production and agricultural sustainability in many regions of the world. It reduces the value and productivity of the affected land and has been seen for years. It has a major impact on crop production in arid and semi-arid regions and causes an agricultural problem under rain-fed conditions when annual precipitation is not enough to leach excessive salts and prevent salt accumulation in the root zone. (1) Cell and tissue culture systems have been considered relevant for the selection of plant species tolerant to salinity, drought, and other kinds of stresses (2)(3) A Salt environment leads to cellular dehydration which causes osmotic stress and removal of water from the cytoplasm resulting in a reduction of the cytosolic and vacuolar volumes. Salt stress often creates both ionic as well as osmotic stress in plants resulting in the accumulation or decrease of specific secondary metabolites in plants. (4) One of the most important factors affecting plant growth and the production of secondary metabolites is salt stress (5). Sesbania grandiflora, commonly known as Agastya or Heti, is a soft-wooded tree that belongs to the family Leguminosae [17][18]. It is a perennial tree and grows up to a height of 10 m. It is used as human food and animal fodder [19]. Although the exact origin of S. grandiflora is not known, it is widely distributed in parts of West Bengal, Assam, and Karnataka [20]. Since ancient times almost every single part of S. grandiflora has been used as traditional medicine to

treat an array of diseases such as dysentery, stomatitis, fever, smallpox, sore throat, and headache. This plant is also used in Indian traditional systems of medicine, Siddha, and Ayurveda, for the treatment of various acute and chronic diseases. Numerous reports have also mentioned the isolation of certain bioactive constituents (such as sterols, phenols, tannins, flavonoids, etc.) from the leaves, flowers, and aerial parts of the plant [21]. These chemical constituents are well known for their potential health benefits and have been reported to possess valuable biological activities such as antibacterial, antifungal, and antioxidant properties. Other than this,H it can fix atmospheric nitrogen and can be used as a green manure to improve soil conditions. So, the diversity of its use surely demands more research [22]. In various research, it was observed that the high levels of salinization induced a significant decrease in the contents of pigment fractions (chlorophyll a and b) and consequently of the total chlorophyll content as compared with control plants [23]. It appears at the beginning of growth that S. grandiflora seedlings are not deficient in carbohydrates and that the supply of carbon compounds is not limiting their growth. So, after prolonged periods of exposure to salinity, the levels of reserve carbohydrates increased particularly in the leaves [23] Antioxidants are the compounds that inhibit substrate oxidation possibly by acting as a free radical scavenger [24]. Free radicals are any species that contain one or more unpaired electrons that react with other molecules by taking or giving electrons [25]. When the amount of these highly unstable free radicals exceeds in the body, it damages the cells and may lead to several diseases. Thus, there is a need for antioxidants of natural origin to protect the human body from diseases caused by free radicals [25]. These natural antioxidants are present in plants [25] especially medicinally important plants. These plants produce bioactive secondary metabolites (such as phenols, flavonoids, etc.) naturally hence preventing several life-threatening degenerative diseases such as cancer caused by oxidative stress [24]. The oxidative stress leads to the excess production of highly reactive oxygen species (ROS) or reactive nitrogen species (RNS) that are extremely harmful to the cells causing cell death. Therefore, it is necessary to determine plant tissue containing many antioxidants for novel drug discovery purposes [24]. An increase in population and increasing demand for plant products along with illegal trades are causing the depletion of medicinal plants [26]. For this reason, the plant tissue culture technique has proved a potential alternative for the production and enhancement of the desirable bioactive components from the plants, to produce enough of the plant material that is needed and for the conservation of the threatened species [26]. Reduced rates of cell division and cell elongation due to stress are the main causes of reduced growth of plants under stress [27] Synthesis and accumulation of organic osmolytes, enhanced activities of antioxidant enzymes, and efficient compartmentalization of toxic ions into other cellular compartments like vacuoles help plants to avert stress-induced damage.[32] Hence, the present analytical study enlightens the effect of salinity on the growth and secondary metabolites of Sesbania Grandiflora seedlings.

II. MATERIALS AND METHODS

Collection of Seeds: Sesbania grandiflora seeds were procured from Numinous India, a seed distributor from Andhra Pradesh.

Surface Sterilization and Inoculation of seeds for germination: The seeds were surface sterilized to produce contamination-free plants that can be maintained under aseptic in vitro conditions. Seeds were checked for viability and were washed 2-3 times with tap water followed by washing 2 times consequently with distilled water and detergent. Seeds were then transferred to the sterilized laminar cabinet where HgCl2 (0.1%) treatment was given to the seeds for 10 minutes with continuous stirring, which was then rinsed 2-3 times with distilled water followed by soaking it in a sterile tissue paper to remove excess water. Sterilized seeds were inoculated aseptically on Murashige and Skoog media (half strength,1L). 6 flasks were supplemented with different concentrations of salt. The inoculated flasks were then transferred to the culture room with controlled light and temperature.

Preparation of MS media (half strength): The medium was prepared such that for 1 L, 2.1 g of MS media (half strength) was dissolved in 1000 ml of distilled water with the addition of 8 gm agar per 1000 ml. Before autoclaving, the medium was buffered with 1N NaOH and 1N HCl to adjust its pH to 5.6–5.8 [8]. Preparation of MS media (half strength) for giving salt stress: A solution of NaCl was prepared by adding 58.44 gm salt per 1 liter. Salt stress of 10 mM,20 mM, and 30 mM for 15 Days,30 days, and 45 Days respectively was given. MS media solution for each concentration of NaCl i.e., 10 mM, 20mM, and 30 mM molar concentration on each solution was prepared. The dissolved medium was autoclaved at 15 lbs. pressure at 121°C for 20 minutes. Seeds were inoculated in the respective flasks along with control seeds for each treatment.

Determination of Toxicity: Toxicity was determined by visual screening of – In vitro-grown plantlets supplemented with different salt concentrations. Proliferation of seed growth based on salt concentration was observed. This was

determined based on seed germination rate, survival count of germinated plantlets, and plant biomass yield obtained (i. e. fresh weight and dry weight). Plantlets that grow with less proliferation, low survival count, and low biomass yield were referred to be toxic, As compared to control flasks (without salt concentration) germination of those with salt concentration was observed late, Germination of seeds was observed lowest in the highest concentration of salt, which is 30mM. 0 mM salt concentration> 10 mM Salt concentration> 20 mM salt concentration > 30 mM salt concentration. After respective periods of respective salt concentration plantlets were taken out from the growth room and morphological analysis was done. Stem length, root length, leaf lamina, No. of leaves were measured.

Preparation of extract: Enough plantlets (Normal, 10 mM,20 mM,30 mM) obtained were air dried and pulverized into powder using mortar and pestle separately such that each powder owns different concentrations of NaCl i. e. Normal, 10 mM, 20 mM, 30 mM. The drying process is important for the extraction of plant materials, the fresh plant materials have the active enzymes that produce the active constituents' intermediates and metabolic reactions in the plant materials [25]. The initial weight of powdered samples (0mM, 10mM, 20mM, 30mM) was measured and the pre-weight of the round water bottle was also measured. This process was carried out at 40-50°C adding 500mg per 10 mL of powder.

Hot water extraction process - Extraction solvent is taken in the round bottom flask and heated by using a heating mantle. After the Hot water extraction process the extracted material was concentrated by evaporating the solvent thereby reducing it into a semisolid mass [26].

The concentrated extract of different salt concentrations (0mM, 10mM, 20mM, 30mM) was stored at 4°C in the refrigerator until further use. These extracts were then subjected to phytochemical screening for the identification of different phytochemical analyses such as Quantitative and Qualitative analysis.

2.1 PHYTOCHEMICAL SCREENING

Three extracts (0mM, 10mM, 20mM, 30mM) of 15 days,30 days, and 45 days thus obtained were subjected to preliminary phytochemical screening following the standard protocols.

Phenols: 500µl of samples were added in 1 ml of 2% FeCl3. The appearance of green, blue, or black color represents the presence of phenols. The absence of green, blue, or black color represents the absence of phenols

Saponins: 500 µl of the sample were added in 2 mL of Distilled water, after shaking vigorously, if stable soap forms it represents the presence of Saponins.

Steroids: 500 µl of sample added in 2 ml of chloroform then in this solution addition of conc. H2SO4 (drop by drop), the Appearance of Red color in the Chloroform layer represents the presence of Steroids.

Glycosides: SALKOWSKI'S TEST:500µl of sample and 1 ml of chloroform and the same volume of conc. H2SO4 was added in a test tube, the appearance of bluish-red to violet-red represents the presence of Glycosides.

Flavonoids: SHINODA TEST: 4 Pieces of Mg ribbon added into 500 µl of sample and conc.H2SO4 added drop by drop, reddish or orange color represents the presence of Flavonoids.

Terpenoids: 500µl of sample added in 1 ml of Chloroform. And left to evaporate to dryness, after chloroform is completely evaporated 2 ml conc. H2SO4 was added after heating the solution for 2 minutes. The appearance of gray color represents the presence of Terpenoids.

2.2 ANALYSIS OF ANTIOXIDANT ACTIVITY

TOTAL PHENOLIC CONTENT: The total phenolics of the samples i.e. 0mM, 10mM, 20mM, 30mM were determined using Folin and Ciocalteu reagent following the method of Shubhangi et.al [31] with some modifications.

Preparation of solutions:

1. Folin and Ciocalteu reagent- 1.5 ml of Folin and Ciocalteu's reagent (10-fold diluted) was prepared.

2. Sodium Carbonate- 50 ml of 7.5 % of Na2CO3 was prepared such that 3.75 g of Na2CO3 was dissolved in 50 ml of distilled water.

3. Gallic acid solution- the solution was prepared at a concentration of 20-100 µg/ml to obtain a standard curve at 760 nm.

4. Test Samples- 0mM, 10mM, 20mM, 30mM salt concentration sample.

After mixing, the solutions were further incubated for 30 minutes at room temperature. The absorbance of the tubes was measured at 760 nm. All the determination was performed in triplicate.

2.3 TOTAL FLAVONOID CONTENT- The aluminum chloride colorimetric method was used for the determination of the total flavonoid content of the sample i.e-0mM, 10mM, 20mM, 30mM following the method described by Shubhangi et.al [31] with slight modification.

III. RESULTS AND DISCUSSION

Morphological Analysis: The impact of Salt Stress on in vitro-grown plantlets was estimated by visual screening and *in vitro*-grown plantlets supplemented with different salt concentrations.

Fig.1. Systematic picture of grown plantlets in different concentrations.

Remarkable changes were observed in the leaves of plantlets, as compared to the plants of 0mM concentration color of the leaves was fading and they looked faint green, a reduction of chlorophyll is seen in plants with higher salt concentration. From the below data, we can interpret more about the germination rate which is reducing as we are increasing salt conc.

An inverse relationship is observed between salt concentration and seed germination rate when done Annova test with the germination data salt concentration %. With increasing salt concentration germination rate is decreasing. High salt levels in the media create osmotic stress which is making plants difficult to absorb water and nutrients essential for germination. As a result, higher salt concentrations can inhibit germination.

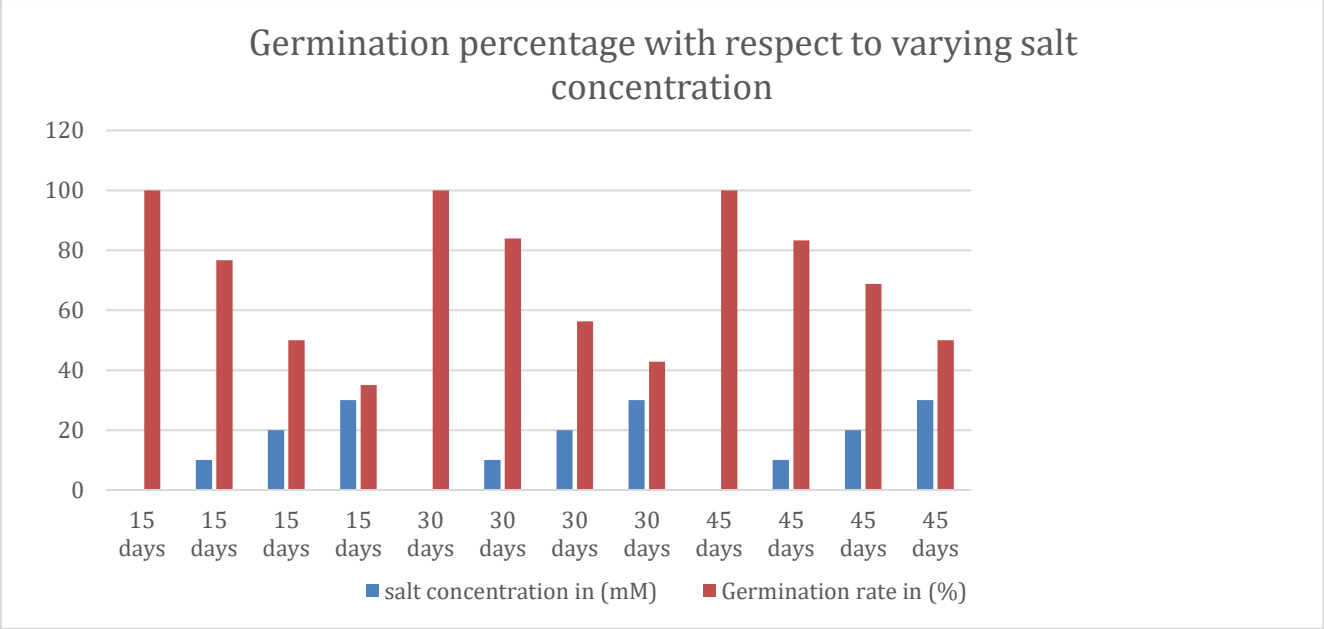


Fig.2: Effect on in vitro grown seedlings with and without Salt stress

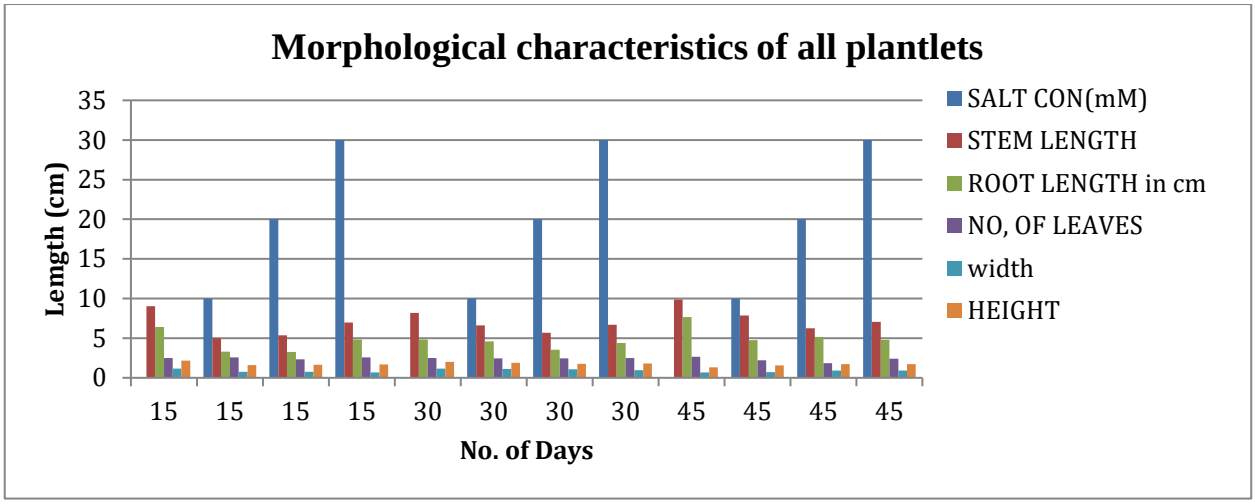


Fig.3. Morphological data of Plantlets in different concentration

It was observed that 0 mM concentration has a normal growth rate as compared to plantlets grown in (10, 20, and 30 mM) salt concentrations. High levels of salinization, (20mM and 30 mM) induce a significant decrease in Stem length, root length, and leaf lamina, It was also noticed that the root length in 45 days increased as compared to those in 30 days and 15 days, so the root length has shown significant growth with the increase in the duration. There is no change in no. of leaves in all concentrations. Overall morphological effect states that salt toxicity affects plants in such a way that they try to complete their life cycle earlier as compared to the control plant.**PHYTOCHEMICAL SCREENING**

Qualitative Analysis: Qualitative phytochemical analysis of plant extract of *Sesbania grandiflora* with concentrations 0mM,10mM,20mM, and 30mM represented below table which represents the presence of phytochemicals such as phenols, saponins, steroids, glycosides, terpenoids in plants with different salt concentration i.e. 10 mM, 20mM, 30mM kept for 15, 30 and 45 days respectively

Table 1. Phytochemical screening of Sesbania grandiflora with and without salt stress

Sr No,	Name of test	Plant Extract											
		15 days salt conc (in mM)				30 days salt conc (in mM)				45 days salt conc (in mM)			
		0	10	20	30	0	10	20	30	0	10	20	30
1	Phenolics	+	+	+	+	+	+	+	+	+	+	+	+
	Saponins	+	+	+	+	+	+	+	+	+	+	+	+
	Terpenoids	+	+	+	+	+	+	+	+	+	+	+	+
	Flavonoids	+	+	+	+	+	+	+	+	+	+	+	+
	Steroids	+	+	+	+	+	+	+	+	+	+	+	+
	Glycosides	+	+	+	+	+	+	+	+	+	+	+	+

(+) Indicates presence; (-) Indicates absence

The preliminary phytochemical analysis revealed the presence of Phenolics, Saponins, Terpenoids, Flavonoids Steroids, and glycosides in plant extract of *Sesbania grandiflora* with different salt concentrations. The addition of optimal concentration of salt did not result in any loss of medicinally active compounds found in the plant.

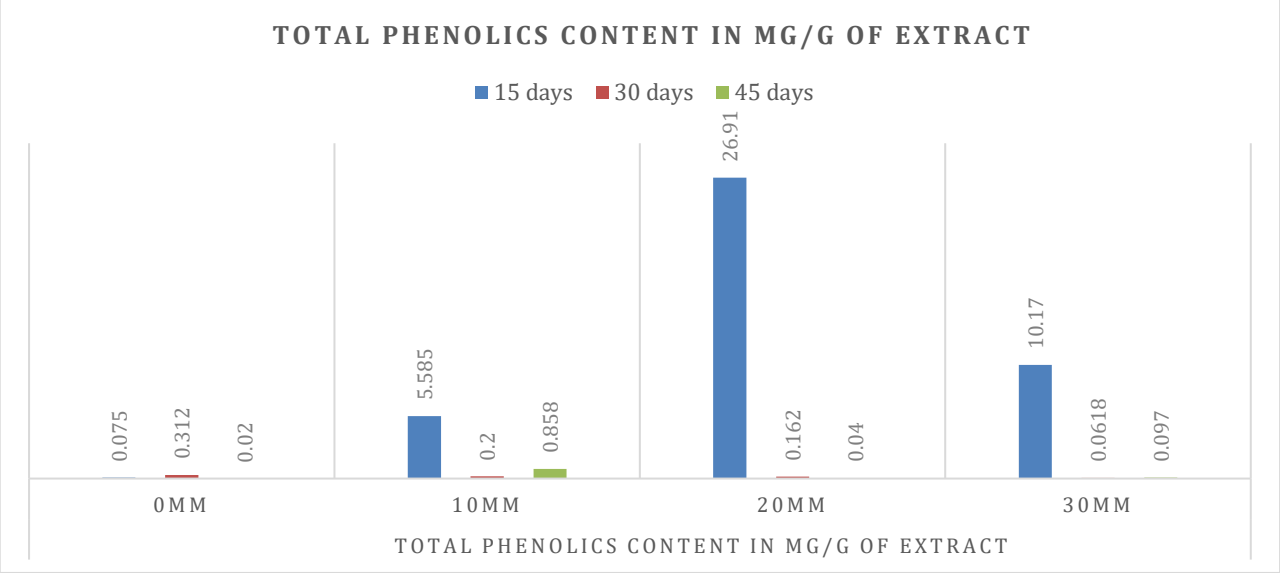
As we know the medicinal uses of secondary metabolites of this sesbania grandiflora plant, the above steroids are present in all salt concentrations, there is no remarkable changes are seen in qualitative analysis, Quantitative analysis of Phenols and Flavonoids is carried out to check the increment or reduction on the quantity of secondary

metabolites.

PHENOLICS CONTENT

Phenolic compounds are important plant constituents with redox properties responsible for antioxidant activity. The hydroxyl groups in plant extracts are responsible for facilitating free radical scavenging [34]. As a basis, phenolic content was measured using Folin- Ciocalteu reagent in each extract C and was depicted in the table given below.

Table 2: Concentration of total phenolic content in the samples



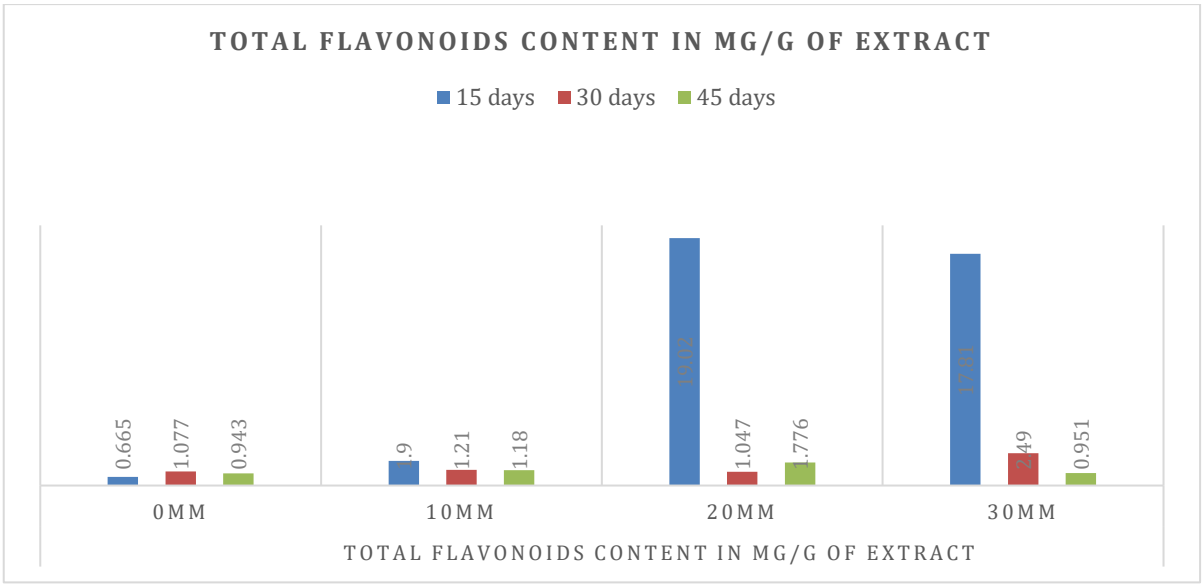
The results were derived from the calibration curve ($Y=7.844x+0.046$, $R^2=0.988$) of gallic acid(20-100 µg/ml) by measuring the absorbance of samples at 765 nm and was expressed in gallic acid equivalents (GAE) per gram dry extract weight.

TOTAL FLAVONOIDS CONTENT

Flavonoids, including flavones, and flavanols are plant secondary metabolites, the antioxidant activity of which depends on the presence of free OH groups, especially 3-OH. Plant flavonoids have antioxidant activity *in vitro* and act as antioxidants *in vivo* [35].

As a basis, Flavonoid content was determined using the aluminum chloride colorimetric method in each extract (0mM, 10mM, 20mM, 30mM) and was depicted in the table given below.

Table 3: Concentration of total flavonoid content in the samples



The Results were derived from a calibration plot (Y=1.6769x-0.0274, R^2=0.9930) of quercetine (50-500 µg/ml) by measuring the absorbance of samples at 415 nm and was expressed in quercetine equivalent (QE) per gram dry extract weight. Annova test conducted on the provided data suggests that there is a significant difference in the total flavonoid content among the various concentrations and periods

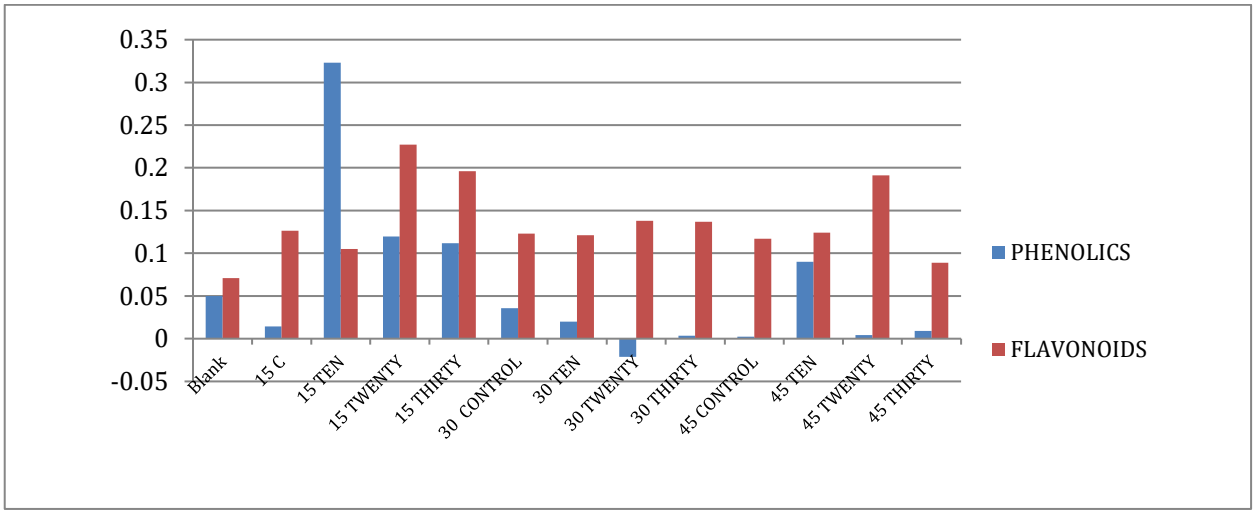


Fig.4. The total phenolic content and total flavonoid content

The table represents the impact of salt concentration on phenolic and flavonoid content in plantlets over different periods. Initially, no salt concentration control plantlets showed lower phenolic content when compared with 10 Mm salt concentration which shows an increase in phenolic levels. AT 20 Mm salt concentration, a decrease in phenolics is observed. Interestingly, a significant growth in phenolic content was noted in 10 Mm concentration after 45 days. Flavonoids showed higher levels of phenolic content in control plants at 15 days compared to 10Mm and 45 days.

Statistical Analysis - The experiment was performed in triplicates and results were represented as mean ± SE. The significance of the results of each experiment was checked by Student's t-test and ANOVAs using Microsoft, Excel. P < 0.05 was considered to suggest statistical significance.

The most significant salt level and duration of salt treatment for maximum phenolic production was found to be 10 Mm salt concentration after 45 days. The remarkable growth in phenolic content was observed at this specific salt level and time point. There is a noticeable increase in the phenolic content compared to other salt levels and time durations.

IV. CONCLUSION

10 Mm salt concentration for a longer duration is sufficient to trigger the secretion of secondary metabolites. This can be helpful for improved metabolite production from this important medicinal plant.

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Novel medium-and long-chain triacylglycerols rich structured lipids enriched in n-3 polysaturated fatty acids encapsulated by spray drying: Characterization and stability

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Abstract— *Novel MLCTs-rich SLs, enriched with n-3 PUFAs, were synthesized to combine the benefits of docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), and medium-chain FAs (MCFAs). However, these SLs were susceptible to oxidative degradation. To solve this issue, the MLCT-rich SLs were microencapsulated using different wall materials through spray drying technique. Initially, in this study three formulations of different wall materials were designed: modified starch/maltodextrin [MS/MD (2:1)], gum Arabic/maltodextrin [GA/MD (2:1)], and mixed [MIX (MS:GA:MD, 1:1:1)]. Subsequently, all the three formulations were analysed. The analysis included encapsulation yield, encapsulation efficiency, moisture content, hygroscopicity, water activity, density properties, Carr's index, cohesiveness, flowability, porosity, wettability, solubility, color, and relative crystallinity. Fourier transform infrared spectroscopy (FTIR) was employed to identify the chemical structure of the microcapsule powder. The moisture content ranged from 1.71% to 3.45%, while water activity ranged from 0.17 to 0.31 of all formulations, indicating suitability for long-term storage. Additionally, the highest microencapsulation yield (93.23%) and microencapsulation efficiency (93.74%) were achieved with GA/MD formulation. GA/MD also exhibited the highest relative crystallinity (32.50%). Moreover, FTIR analysis confirmed the successful encapsulation of the oil in the microcapsules. Furthermore, scanning electron microscopy (SEM) images revealed spherical shapes without any visible cracking on the surfaces of the microcapsules. Besides, GA/MD formulation demonstrated better results in density properties, flowability, porosity, and wettability. During the oxidative stability, GA/MD microencapsulation provided the best protection against lipid oxidation. These findings highlight the effectiveness of using GA/MD formulation for the production of microcapsule powders containing MLCTs-rich SLs.*

Keywords— ***Medium and Long-Chain Triacylglycerols; n-3 Polyunsaturated Fatty Acids; Bio-imprinted Lipase; Interesterification; Microencapsulation***

I. INTRODUCTION

Structured lipids (SLs) can be produced enzymatically or chemically via interesterification, acidolysis, and/or esterification processes from the conventional fats/oils in order to improve their nutritional and functional properties (Lu et al., 2017). SLs are available in a number of

commercial zero- or low- calorie food ingredients, for example salatrim (Nabisco, USA) and diacylglycerol (Kao Cooperation, Japan). Medium and long-chain triacylglycerols (MLCTs) are a special kind of SLs found in food products like Resetta™ (Nisshin Oillio Group Ltd., Japan) that have been found with many physiological benefits, such as anti-obesity effect and the maintenance of good cholesterol (Lee et al., 2015). Furthermore, MLCTs possess the capability to provide the body with quick energy and essential fatty acids such as arachidonic acid (ARA) by an easy way, thus they are very useful for humans, in particular for infants (Korma et al., 2018). Polyunsaturated fatty acids (PUFAs) are FAs with more than one double bond. Omega-3 (n-3) PUFAs include eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA) and alpha-linolenic acid (ALA). The health benefits related to n-3 PUFAs are reported in several studies (Calder, 2020; Khan et al., 2023). The consumption of EPA and DHA helps in the treatment and prevention of inflammatory, neurological, and cardiovascular diseases (Jiang et al., 2022; Khan et al., 2023). Fish oil is abundant in EPA and DHA but contains very low concentration of ALA. Examples of fish oils with the highest concentrations of EPA and DHA include tuna, anchovy, sardine, cod, and salmon, among others. Nevertheless, the reported concentration of DHA in different fish oils ranges from 10% to 30%. The derivation of DHA and EPA from fish oils has been demonstrated through both chemical and enzymatic processes (Ackman & Burgher, 1963). Therefore, it is important to reduce the oxidation of ARA, in order to improve their quality in food applications.

In the encapsulation method, the wall materials have an excellent influence on the physical and functional properties of the microencapsulated products (Premi & Sharma, 2017). Maltodextrin (MD), gum arabic (GA), and whey protein isolate (WPI) are the most frequently applied wall materials for microencapsulation of oils (Chew et al., 2018; Korma et al., 2019). Maltodextrin is the most commonly used wall material due to its low cost, low viscosity at a high solid concentration, neutral taste, and well protection against oxidation with low capacity for emulsifying (de Barros Fernandes et al., 2014). Thus, the MD preferred only in various combinations with other carrier agents such as WPI and GA. GA exhibits low viscosity, high solubility, and good emulsifying characteristics that make it an appropriate wall material (Korma et al., 2019). The utilization of WPI as an encapsulating agent in the food industry is very appealing due its extensive nutritional benefit. Besides, the presence of hydrophilic and hydrophobic amino acids of whey protein smoothens the encapsulation of compounds of the hydrophobic nature (Premi & Sharma, 2017). In recent decades, researchers applied microencapsulation with different carriers and techniques to improve the quality and stability of oils (Bakry et al., 2016). Consequently, the stability, nutritional quality, and health properties of several vegetables, marine, and seed oils have been improved by microencapsulation (Bakry et al., 2016).

In order to improve the applicability of MLCTs-rich SLs rich in n-3 PUFAs as lipid ingredient in nutritional and functional food applications, MLCTs-rich SLs could be transformed into powder and encapsulated on food products by spray drying technology. Hence, the aims of this study were to investigate the development of a combination of the gum arabic, maltodextrin, and modified starch, as wall materials for microencapsulation of the MLCTs-rich SLs by spray drying, and to evaluate the physicochemical characteristics and oxidative stability of the encapsulated MCLTs-rich SLs during storage.

II. MATERIALS AND METHODS

2.1. Materials

MS was purchased from Hilmar ingredients (Tianjin Milky way import & export Co., Ltd., China). GA was procured from Sinopharm (Chemical Reagent Co., Ltd., China). MD was provided by Shyuanye Co. China. OMAX 1812 fish oil (a mixture of anchovy and sardine oils) was purchased from NovoSana Co., Ltd (China). MCTs containing 60% caprylic acid and 40% capric acid were obtained from J&K Scientific Ltd. Under controlled conditions in sealed bags. In all experiments, the high purity and analytical grade chemicals were used.

2.2. Production and purification of MLCTs-rich SLs

The best conditions achieved the highest yield of MLCTs-rich SLs were selected for the scale production of MLCTs-rich SLs according to our previous study. The solvent-free interesterification reaction was performed in a 500 mL stirred batch reactor at 70 °C for 3 h with a substrate's mole ratio of ARASCO: MCTs (1:1.5 mol/mol), 4% (w/ w) of bio-imprinted NS 40086 lipase, and constant stirring at 600 rpm. The reactor was covered with foil to decrease exposure to light. At the end of the reaction, the resulting of MLCTs-rich SLs was collected after removal of the enzyme by filtration. Purification of MLCTs-rich SLs product was performed according to the method described by (Korma et al., 2018). The free fatty acids of MLCTs-rich SLs were reduced from 2.7 to 0.13 %, and determined by the method reported by Firestone (2009). Purified MLCTs-rich SLs were stored in an airtight amber container under nitrogen at -20 °C. The major fatty acids in the synthesized SLs were EPA, DHA, caprylic, and capric acids.

2.3. MLCTs rich SLs emulsion preparation

Three formulations were used based on the method outlined by Chew et al. [275], with some modifications. These formulations consisted of (GA/MD, 2:1), (MS/MD, 2:1), and mixed (MIX) (GA/MS/MD, 1:1:1) wall materials (**Table 1**). Firstly, the wall materials were dissolved in distilled water on the day before emulsification and kept overnight at room temperature to ensure the full hydration of the polymer molecules of the materials. Then the MLCTs-rich SLs were then dispersed in these solutions using a high-speed homogenizer (Ultra-Turrax IKA T18 basic, Wilmington, NC, USA) at 22,000 rpm for 4 min at room temperature. A fine emulsion was then formed by passing the dispersion through an ATS AH2100 high pressure homogenizer (AH- 2010, ATS Engineering Inc., Canada) operated at 40 MPa with 4 processing cycles. The resulting emulsions were stored at 4 °C with 0.02% (w/v) sodium azide as an antimicrobial agent until further analysis.

Table 1. Description of formulation.

Treatments	Wall materials (g/100g)			Core materials (g/100g)	Wall to the core ratio	Total solid to the water ratio
	MD	MS	GA	MLTs rich SLs		
MS/MD	33.33 g	66.67 g	-	33.33 g	3:1	30:70
GA/MD	33.33 g	-	66.67 g	33.33 g	3:1	30:70
MIX	33.33 g	33.33 g	33.33 g	33.33 g	3:1	30:70

Abbreviations: MS = modified starch; MD = maltodextrin; GA = gum Arabic; MIX = mixed

2.4. Microencapsulation by spray drying

The microcapsules containing MLCTs-rich SLs were produced by drying the homogenized MLCTs-rich SLs emulsions using a GEA Niro spray dryer (model Mobile MinorTM, Soborg, Denmark). The emulsions were added into the drying chamber through a peristaltic pump at a feed flow rate of 14 - 15 mL min^{-1}. Operating parameters for the spray dryer included inlet and outlet temperatures set at 180 °C ± 5 °C and 80 °C ± 5 °C, respectively, with an airflow rate of 300 NL min^{-1}. The resulting dried powder was then collected and stored in sealed plastic bags at -20 °C for further analysis.

2.5. Characterization of spray dried microcapsules

2.5.1. Microencapsulation yield

The encapsulation yield post-spray drying was determined as a percentage, calculated by dividing the total weight of the microcapsule powders by the weight of the carrier agents, according to Equation (1).

$$\text{Encapsulation yield (\%)} = \frac{\text{weight (g) of the collected powder}}{\text{weight (g) of the carrier agents}} \times 100 \quad (1)$$

2.5.2. Microencapsulation efficiency of total oil (TO), and surface oil (SO)

The TO content within the particles was determined through cold extraction with an organic solvent. Initially, 8 millilitres of distilled water were added to 2.5 grams of powder in a glass tube equipped with a screw cap, which was then placed in an ultrasound bath for 5 minutes at room temperature. Following this, 10 mL of chloroform and 20 mL of methanol were added into the tubes, following the methodology outlined by (Bligh & Dyer, 1959). The tubes were sealed with tape and the lids, and manually shaken by inversion of 50 times. After adding an additional 10 mL of chloroform, the tubes were shaken for an additional 2 minutes and then left undisturbed for 30 minutes to allow for phase separation. The upper hydrophilic phase (containing methanol and water) was discarded. The remaining sample in the lower chloroform phase was filtered through a Whatman filter paper no. 1. A 5 mL aliquot of the filtered solution was transferred to an oven at 100°C for 1 hour to completely evaporate the solvent. The remaining mass in the beaker was considered as the total oil content present in the particles.

The SO was evaluated following a method described by (Bae & Lee, 2008), with certain modifications. Initially, 1.5 grams of powder were washed with 15 millilitres of hexane in a glass tube with a screw cap for 2 minutes at room temperature. The resulting mixture was then filtered through a Whatman filter paper no. 1. The powder collected on the filter was rinsed three times with 20 mL of hexane. Subsequently, the solvent was allowed to evaporate at 60°C until a constant weight was achieved. The non-encapsulated oil was determined by the difference in mass between the initial clean flask and the one containing the extracted oil residue, following methods outlined by (Carneiro et al., 2013).

Both TO and SO determinations were used to calculate the microencapsulation efficiency (M. E. E) using Equation (2) as described by (Bae & Lee, 2008).

$$\text{M.E. E} = \left(\frac{\text{TO-SO}}{\text{TO}}\right) \times 100 \quad (2)$$

2.5.3. Moisture content

The moisture content of the microcapsule powders was assessed using the air oven method, as detailed in reference (Kang et al., 2019). The moisture content was determined by measuring the weight loss after drying the sample in a drying oven at 105˚C until a constant weight was achieved. This process is represented by Equation (3):

$$\text{Moisture content (\%)} = \frac{\text{wet sample weight (g)-dry sample weight (g)}}{\text{wet sample weight (g)}} \times 100 \quad (3)$$

2.5.4. Water activity (aw)

The water activity of the microcapsule powders was determined using an electronic water activity meter (Labmaster-aw, Novasina AG, Neuheimstrasse, Switzerland) following stabilization of the microcapsule powder samples at 25˚C for 15 minutes, as described by (Santhalakshmy et al., 2015).

2.5.5. Hygroscopicity

The hygroscopicity of the microencapsulates was evaluated following the method outlined by Cai and Corke (Cai & Corke, 2000), with minor modifications. Initially, approximately 0.5 grams of each sample were placed in a preweighed petri dish. These dishes were then placed in a desiccator filled with a saturated NaCl solution (75% relative humidity) at 25°C. After one week, the samples were removed from the desiccator and reweighed. The hygroscopicity of the samples was determined based on the change in weight observed over the test period.

2.5.6. Wettability

The wettability of each sample was evaluated by determining the time (minutes) needed to immerse 1 gram of microcapsule powder on the surface of 400 mL of distilled water at a temperature of 25˚C (Mahdi et al., 2020).

2.5.7. Solubility

The solubility of the microcapsule powders was evaluated following the method outlined by (Santhalakshmy et al., 2015). Specifically, 1 gram of microcapsule powder was mixed and transferred into 100 mL of distilled water. The mixture was then stirred using a magnetic stirrer at 500 rpm for 30 minutes at 25˚C, followed by centrifugation at 3000 rpm for 5 minutes. An aliquot of 25 mL of the supernatant was dried at 105 ˚C. Solubility of the microcapsule's powders were then calculated using equation (4):

$$\text{Solubility (\%)} = \frac{\text{weight of the powder (g) in the supernatent}}{\text{weight of the powder (g) in the solution}} \times 100 \quad (4)$$

2.5.8. Bulk density (BD)

The BD of the sample powders was analysed following the procedure outlined by (Saifullah et al., 2016). In summary, 2.0 grams of the powder sample were added into a 10 mL graduated measuring glass cylinder. The bulk density was subsequently calculated using equation (5):

$$\text{BD (g/cm}^3\text{)} = \frac{\text{sample weight (g)}}{\text{sample volume (cm}^3\text{)}} \quad (5)$$

2.5.9. Tapped density (TD)

The TD was evaluated following the procedure described by (Saifullah et al., 2016). To measure the tapped volume of the samples, 2 grams of the powder were placed in a 10 mL graduated measuring glass cylinder. The cylinder was then tapped gently 100 times onto a rubber mat from a height of 20 cm. Subsequently, the tapped density was calculated using equation (6):

$$\text{TD (g/cm}^3\text{)} = \frac{\text{sample weight (g)}}{\text{sample volume (cm}^3\text{)}} \quad (6)$$

2.5.10. Particle density (PD)

PD was determined in accordance with the protocol outlined by (Santhalakshmy et al., 2015). In summary, 1.0 gram of powder was transferred into a 10 mL measuring cylinder equipped with a glass stopper. Approximately 5.0 mL of petroleum ether was then added, and the mixture was shaken for 30 seconds. Subsequently, the walls of the cylinder were rinsed with 2 mL of petroleum ether, and the total volume was recorded. The PD was calculated using equation (7):

$$\text{TD (g/cm}^3\text{)} = \frac{\text{sample weight (g)}}{\text{total volume of petroleum ether and suspended particles (cm}^3\text{) -7}} \quad (7)$$

2.5.11. Flowability

The Flowability was estimated based on the Carr index (CI) and Hausner ratio (HR) (Saifullah et al., 2016). CI was used to expresses the flowability of the sample powders based on tapped and bulk densities (Saifullah et al., 2016). The CI of the microcapsules powders was calculated by using equation (8);

$$\text{carr index (\%)} = \frac{\text{(TD-BD)}}{\text{TD}} \times 100 \quad (8)$$

Where TD is tapped density and BD is bulk density.

The cohesiveness of the samples was estimated using HR based on tapped and bulk densities (Saifullah et al., 2016). The HR was calculated using equation (9);

$$\text{hausner ratio} = \frac{\text{TD}}{\text{BD}} \quad (9)$$

where TD is tapped density and BD is bulk density.

2.5.12. Bulk porosity (BP)

The BP of the samples was determined according to the method reported by (Saifullah et al., 2016), based on PD and TD. BP was estimated using equation (10);

$$\text{bulk porosity (\%)} = \frac{(\text{PD-TD})}{\text{PD}} \times 100 \quad (10)$$

Where TD is tapped density and PD is particle density.

2.5.13. Color (L*, a*, and b*) values

The color of the microcapsule powders was assessed using a colorimeter (HunterLab UltraScan PRO Spectrophotometer, Hunter Associates Laboratory, Inc., Virginia, USA). Before measurement, the instrument was calibrated to standard white and black tiles. About 5 g of microcapsule powder was placed in a transparent plastic bag, and the color parameters [L* (light/dark), a* (red/green), and b* (yellow/blue)] were measured on three randomly selected surfaces of each sample (AL-Ansi et al., 2019).

2.5.14. Scanning electron microscope

The morphology of the microcapsules was examined using a scanning electron microscope (SEM), specifically the Hitachi High-Technologies Corp. SEM SU 1510 (Tokyo, Japan). Samples were fixed onto metal stubs using double-sided sticky tape, with dimensions of 1 cm in diameter and 1 cm in height. Prior to imaging, the samples were gold-coated using a sputter coater (Hitachi, E-1010) within a high-vacuum evaporator. Scanning was performed at an accelerating beam voltage of 3.0 kV and a magnification of 1000x.

2.5.15. Fourier transform infrared (FTIR)

The chemical structure was identified using the FT-IR spectrophotometer (Nicolet iS10, Thermo Fisher Scientific Co. Ltd., Waltham, Massachusetts, USA) according to the established method by (Hu et al., 2018). The sample powder was blended with KBr powder in a ceramic mortar and pressed into pellets. The FT-IR spectrum of the samples was measured at a resolution of 4 cm^{-1} in transmission mode from 500 to 4000 cm^{-1} wave length range.

2.5.16. Crystallinity (X-ray diffraction)

The crystallinity was evaluated using an X-ray diffractometer (D2PHASER, Bruker AXS Co. Ltd., Karlsruhe, Germany) (Li et al., 2018). The scanning region of the diffraction angle (2θ) ranged from 5° to 80°. X-ray diffraction patterns of samples were analysed using software (MDI Jade 6) and calculated as relative crystallinity (%) according to the following equation;

$$\text{RC (\%)} = \frac{\text{sum of total crystalline peak areas}}{\text{sum of the total crystalline and amorphous peak areas}} \times 100 \quad (11)$$

2.5.17. Oxidative stability under accelerated storage

The powder samples were subjected to accelerated storage conditions, maintained at 60°C for a duration of 28 days. At specific intervals of 0, 7, 14, 21, and 28 days, samples were collected from the storage and evaluated for peroxide value (PV) (Firestone, 1989).

2.6. Statistical evaluation

The experiments and sample measurements were replicated three times. Data analysis was performed using Microsoft Excel 2021 and Origin 9.9.0 software, and the results were presented as mean values accompanied by standard deviations. ANOVA (Analysis of Variance) was carried out using the Statistical Analysis System software (SAS, SPSS). A significance level of $\alpha = 0.05$ was used, and any differences with a P-value < 0.05 were considered significant.

III. RESULTS AND DISCUSSION

3.1. Encapsulation yield

The yields of microencapsulated MLCTs rich in SLs are presented in **Table 2**. During this study, the microencapsulation yield ranged from 85.34% to 93.23%, a range considered satisfactory based on the drying standard reported by Du et al. (Du et al., 2014). The results indicated that the GA/MD (93.23%) formulation exhibited the highest microencapsulation yield. Interestingly, there were no significant differences ($p \leq 0.05$) observed between GA/MD and MIX formulations. This lack of difference may be attributed to the presence of GA in both formulations, as GA possesses a strong capability for film-forming. Conversely, the MS/MD (85.34%) formulation demonstrated the lowest microencapsulation yield, may be due to the absence of GA in its composition.

3.2. Microencapsulation efficiency

Microencapsulation efficiency (M.E.E) assists as a critical parameter for confirming the effectiveness of an encapsulation process, regardless of the methodology or core materials used (de Souza et al., 2018). It holds particular significance in evaluating the quality of oil encapsulation products, especially in the context of spray drying. The M.E.E values reported in this study were 90.34%, 93.74%, and 91.65% for MS/MD, GA/MD, and MIX microcapsules, respectively (**Table 2**). The spray-drying technique is preferred for producing powders with high M.E.E values, typically around 90% (Carneiro et al., 2013; Ng et al., 2013). In this study, all three formulations demonstrated excellent retention of MLCTs rich SLs, with M.E.E values exceeding 90%. GA/MD exhibited the highest M.E.E, followed by MIX. These outcomes suggest

that the polysaccharides utilized (GA and MD) enhanced M.E.E in this investigation. Similar findings were reported in other studies regarding microcapsules of refined kenaf (Hibiscus cannabinus L.) seed oil (Chew et al., 2018).

3.3. Moisture content and water activity

Moisture content and water activity are crucial indicators of powder quality and storage stability. The results revealed moisture contents of 3.45%, 1.71%, and 2.66% for microencapsulated MLCTs rich SLs in the MS/MD, GA/MD, and MIX treatments, respectively (**Table 2**). MS/MD exhibited a relatively higher moisture content (3.45%) compared to GA/MD (1.71%), attributed to the higher water-holding capacity of proteins in an amorphous state. Consequently, an increase in MS proportion resulted in higher moisture content in MS/MD. Conversely, in the GA/MD formulation, the incorporation of GA and MD in a 2:1 ratio led to a reduction in moisture content, enabling robust wall formation. Overall, all results indicated moisture contents below 4%, a desirable characteristic for powders, as the maximum moisture content for most powders used in the food industry is typically 4% (Karaca et al., 2013; Korma et al., 2019).

Water activity values for MS/MD, GA/MD, and MIX formulations are also presented in **Table 2**. Similar to the moisture content findings, the addition of GA and MD in a 2:1 ratio in the GA/MD formulation resulted in decreased water activity. Powders with water activity levels below 0.3 are deemed to possess high shelf-life stability against microbial growth (Drusch & Schwarz, 2006; Korma et al., 2019).

3.4. Hygroscopicity

Hygroscopicity refers to a material's ability to absorb moisture from the atmosphere, which can lead to fat oxidation in powders, affecting their nutritional value and flow. The hygroscopicity of MS/MD was significantly lower ($p < 0.05$) compared to GA/MD and MIX formulations, with values of 6.75 g/100 g, 7.44 g/100 g, and 8.67 g/100 g, respectively (**Table 2**). It was previously suggested that samples with lower moisture contents would exhibit increased hygroscopicity, as the ability to absorb moisture is influenced by the water concentration gradient between the powder products and the atmosphere (Tonon et al., 2011). However, the results revealed that MS/MD possessed both the lowest moisture content and the lowest hygroscopicity. The larger particle size of MS/MD compared to other powders may reduce the exposure area of the particles to atmospheric moisture, contributing to its lower hygroscopicity. In contrast, moisture absorption by GA/MD is attributed to the presence of hydrogen (H_2) in water molecules and the hydroxyl groups present in GA and MD. In this study, the hygroscopicity values of microencapsulated gurum seed oil were lower than those reported in previous studies using GA as the wall material, such as Gagaita fruit extract (14.8–18.8%) (Daza et al., 2016), and rosemary essential oil (15.9–18.9%) (de Barros Fernandes et al., 2014). Similar findings were reported by Chew et al. (Chew et al., 2018), for microencapsulated refined kenaf seed oil (7.8–10.1%).

3.5. Wettability

Wettability refers to a powder's ability to absorb a liquid due to capillary forces, with shorter dissolution times indicating better physical properties and potential applications in the food industry (Chew et al., 2018). **Table 2** presents the wettability data for all freshly microencapsulated MLCTs rich SLs. The results indicated that MS/MD exhibited a wettability of 575 s, whereas GA/MD and MIX showed wettability times of 290 s and 744 s, respectively. Notably, the use of GA and MD in a 2:1 ratio reduced the wettability time compared to MS/MD and MIX formulations. This finding aligns with Dima et al.'s observation that decreasing particle size increases wettability time, consistent with the results of this study (Dima et al., 2016).

3.6. Solubility

Solubility marks the last stage of particle dissolution and holds significant importance in assessing food quality (de Barros Fernandes et al., 2014). Powders with poor solubility can lead to processing challenges and economic losses. The results indicated that all powders exhibited solubility, ranging from 86.27% to 92.84% (**Table 2**). MLCTs rich SLs are typically insoluble in water at room temperature. However, the wall materials utilized for encapsulating the MLCTs rich SLs contributed to enhancing their solubility.

Table 2. Encapsulation yield, M.E.E., moisture content, water activity, hygroscopicity, wettability and solubility.

Treatments	MS/MD	GA/MD	MIX
Encapsulation yield (%)	85.34±0.05^{c}	93.23±0.03^{a}	89.88±0.03^{b}
M.E.E (%)	90.34±0.04^{c}	93.74±0.03^{a}	91.65±0.05^{b}
Moisture content (%)	3.45±0.05^{a}	1.71±0.04^{c}	2.66±0.03^{b}
Water activity	0.31±0.01^{a}	0.17±0.01^{c}	0.24±0.02^{b}
Hygroscopicity (g/100g)	6.75±0.01^{c}	7.44±0.04^{b}	8.67±0.05^{a}

Wettability (s)	575.33±4.51[b]	290.67±4.04[c]	744±4[a]
Solubility (%)	88.72±0.03[b]	92.84±0.04[a]	86.27±0.03[c]

Abbreviations: MS/MD = modified starch/ maltodextrin; GA/MD = gum Arabic/ maltodextrin; MIX = mixed; M.E.E = microencapsulation efficiency. Data presented as means of triplicate ±SD. Data with the different superscript letters (within a column, a,b,c) are significantly different ($p < 0.05$).

3.7. Bulk density (BD)

The bulk density of microencapsulated MLCTs rich SLs varied from 0.15 to 0.25 g.cm^{-3} for MS/MD, GA/MD, and MIX formulations (**Table 3**). These results fell within the expected range for bulk density in microencapsulated powders, as reported in previous models (Carneiro et al., 2013). Notably, MS/MD exhibited the lowest bulk density among the GA/MD and MIX microencapsulated MLCTs rich SLs. This difference underscores the significant influence of the wall material on the bulk density of microencapsulated MLCTs rich SLs. The lower bulk density observed in MS/MD formulations may be attributed to the spongy nature of the microcapsule wall produced by MS/MD. This characteristic is likely influenced by the higher proportion of MS in the MS/MD model, which has a distinct impact on bulk density. Additionally, the larger particle size of MS/MD microencapsulated MLCTs rich SLs contributes to lower particle density, as density decreases with an increase in volume for a fixed mass product (Goyal et al., 2015). Our study observed a similar trend, where bulk density decreased with increasing particle size. Conversely, the higher solid content in GA/MD and MIX microencapsulated formulations would contribute to increased bulk density. When compared to microencapsulated flaxseed oil using MD mixed with whey protein concentrate (WPC) and MS, GA mixed with MD tended to produce denser particles (Carneiro et al., 2013).

3.8. Tapped density (TD) and particle density (PD)

Tapped density is a critical factor, affecting the packaging, transportation, and commercialization of powders. In the case of microencapsulated MLCTs rich SLs, the tapped density ranged from 0.33 to 0.43 g.cm^{-3} for MS/MD, GA/MD, and MIX formulations (**Table 3**). In our study, GA/MD microencapsulated MLCTs rich SLs exhibited the highest tapped density, followed by the MIX microencapsulated formulation. The ratio of the wall material significantly influenced the tapped density, with the use of MS together with MD resulting in lower tapped density in MS/MD-microencapsulated MLCTs rich SLs. Powders generated through spray-drying typically exhibit high tapped density, which is advantageous for storing them in small containers conveniently. Moreover, higher bulk density indicates a lesser amount of air in the powders, which can help prevent fat oxidation during storage (Carneiro et al., 2013).

The particle density of microencapsulated MLCTs rich SLs ranged from 0.81 to 1.25 g.cm^{-3} in this study. MS/MD microencapsulated MLCTs rich SLs demonstrated significantly lower particle density ($p < 0.05$) compared to GA/MD and MIX microencapsulated formulations. This observation can be attributed to MS/MD contributing to lower PD compared to GA. The particle density of GA/MD and MIX microcapsules was similar to that reported by Fernandes et al. (de Barros Fernandes et al., 2014), who used GA to microencapsulate rosemary essential oil (1.0-1.3 g mL^{-1}).

3.9. Flowability, cohesiveness, and bulk porosity

The Carr's Index (CI) evaluates the free-flowing characteristic, while the Hausner ratio (HR) evaluates the cohesiveness of the powders. In this study, the results showed that the CI was in the range of 41.86-54.55, indicating awful flowability (**Table 4**). The HR was in the range of 1.72-2.20 for the developed microencapsulated MLCTs rich SLs. The higher HR indicates that the powders are more cohesive and have lower ability to flowability. GA/MD-microencapsulated MLCTs rich SLs displayed significantly lowest ($p < 0.05$) CI and HR values among the MS/MD and MIX-microencapsulated MLCTs rich SLs. This showed that the GA could contribute better flowability than the MS. Anyway, there were previous studies reported awful flowability of the powders with high CI values (33.7-48.7) in flaxseed oil microcapsules (Carneiro et al., 2013), in sunflower oil microcapsules (Goyal et al., 2015), and fish oil microcapsules (Kagami et al., 2003).

Bulk porosity (BP) is an important property in food processing, especially in the reconstitution of dry powder. In this study, the results showed that the bulk porosity of MIX-microencapsulated MLCTs rich SLs was significantly different ($p < 0.05$) than that of MS/MD and GA/MD-microencapsulated MLCTs rich SLs. MS and MD contributed to the lowest bulk porosity of MS/MD microencapsulated MLCTs rich SLs between the microcapsules produced. However, the bulk porosity values in this study were lower than the values (70.0-74.5%) reported by Jinapong et al. (Jinapong et al., 2008), in soy milk powders. The large contents of bulk porosity indicate the presence of a big number of wipers among the particles,

which holding O2 that causes degradation reactions (Santhalakshmy et al., 2015).

Table 3. Bulk density, tapped density, particle density, Carr index (CI), Hausner ratio (HR), and flowability of the gurum seed oil microcapsule powders.

Treatments	MS/MD	GA/MD	MIX
Bulk density (g cm^{-3})	0.15±0.01^{c}	0.25±0.03^{a}	0.23±0.02^{b}
Tapped density (g cm^{-3})	0.33±0.04^{c}	0.43±0.03^{a}	0.41±0.01^{b}
Particle density (g cm^{-3})	0.81±0.02^{c}	1.25±0.03^{a}	1.02±0.01^{b}
Bulk porosity (%)	48.94±0.04^{c}	65.03±0.02^{b}	66.1±0.03^{a}
Carr Index (%)	54.55±0.05^{a}	41.86±0.03^{c}	43.9±0.03^{b}
Hausner Ratio (HR)	2.20±0.05^{a}	1.72±0.04 ^{c}	1.78±0.03^{b}
Flowability	Awful	Awful	Awful

Abbreviations: MS/MD = modified starch/ maltodextrin; GA/MD = gum Arabic/ maltodextrin; MIX = mixed; M.E.E = microencapsulation efficiency. Data presented as means of triplicate ±SD. Data with the different superscript letters (within a column, a,b,c) are significantly different ($p < 0.05$).

Table 4. Flowability and cohesiveness from the Carr's index and Hausner ratio (HR).

Flowability	Carr index (%)	Hausner ratio (HR)
Excellent	0-10	1.00–1.11
Good	11–15	1.12–1.18
Fair	16-20	1.19–1.25
Passable	21-25	1.26–1.34
Poor	26–31	1.35–1.45
Very poor	32–37	1.46–1.59
Awful	>38	>1.60

3.10. Color

The color parameters, which include luminosity (L* representing the degree of lightness on a scale from 0 to 100 where 0 is black and 100 is white), a* (representing the degree of redness (+) to greenness (-)], and b* (representing the degree of yellowness (+) to blueness (-)], are presented in **Table 5** (Korma et al., 2019).

Table 5. Color parameters of the MLCTs rich SLs microencapsulated powder.

Parameters	L*	a*	b*
MS/MD	94.89±0.04^{b}	-0.78±0.02^{a}	9.02±0.04^{a}
GA/MD	95.60±0.02^{a}	-0.52±0.03^{c}	7.25±0.03^{c}
MIX	94.75±0.05^{c}	-0.70±0.04^{b}	8.16±0.03^{b}

L* (luminosity degree), a* (degree of redness to greenness), b* (degree of yellowness to blueness). The results are expressed as the mean value ± standard deviation (n=3). Different letters in the same column indicate significant differences ($p \leq 0.05$) between the data.

As can be seen in **Table 5**, there were no significant differences ($p \leq 0.05$) observed among the mean values of lightness (L*) for the microcapsule powder formulations investigated in this study. The lightness mean values (L*) ranged from 94.75 to 95.60, suggesting that all samples tended to be of white color.

The mean values of a* ranged from -0.52 to -0.78, indicating a tendency towards green color for all samples.

Notably, the GA/MD formulation exhibited a significantly lower ($p \leq 0.05$) mean value of a* compared to other formulations.

Regarding the mean values of b*, they ranged from 7.25 to 9.02, suggesting a tendency towards yellow color for all samples. The GA/MD formulation showed a significantly lower ($p \leq 0.05$) mean value of b* compared to other formulations. This difference could be attributed to its high whey protein content.

3.11. Crystallinity (X-ray diffraction)

Determining whether encapsulated substances possess crystalline or amorphous structures is crucial for understanding their stability, often analyzed through XRD. The XRD patterns of microencapsulated MLCT-rich SLs powders are depicted in **Fig 1**. The relative crystallinity observed in MS/MD, GA/MD, and MIX microencapsulated MLCT-rich SLs ranged from 30.28% to 32.50%. Among these, GA/MD-microencapsulated MLCT-rich SLs exhibited the highest relative crystallinity, followed by MS/MD microencapsulated MLCT-rich SLs. The results suggest that the MIX model, incorporating MS, GA, and MD in a 1:1:1 ratio, exhibits a slight inclination towards crystallization. Additionally, the GA/MD model may offer enhanced storage stability due to its higher relative crystallinity compared to other models. Generally, amorphous samples tend to be hygroscopic, absorbing more water during storage. This propensity for water absorption can lead to weight gain, nutrient degradation, breakdown of microstructure, and increased microbiological instability, ultimately compromising sample quality during storage (Borrmann et al., 2013).

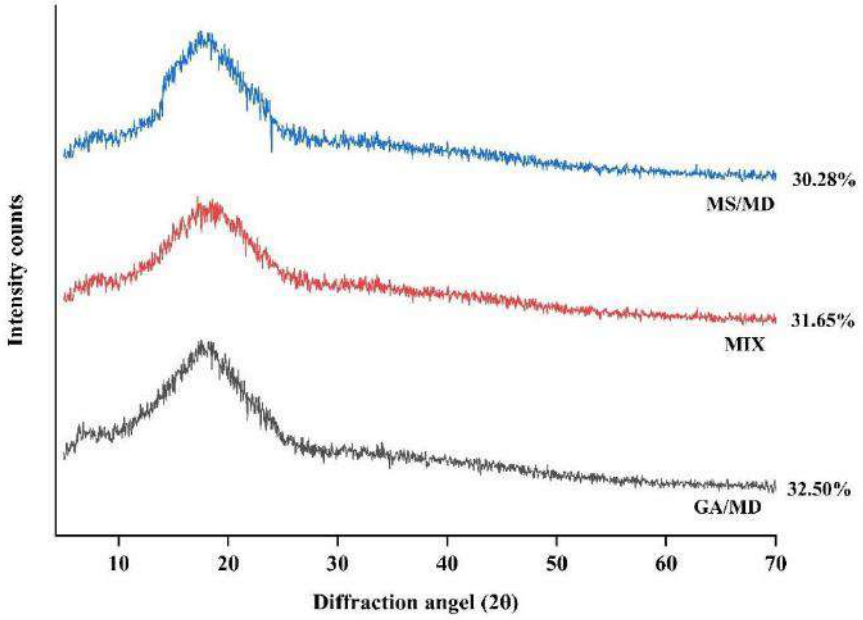

Fig. 1. Relative crystallinity by X-ray diffraction patterns of MLCTs rich SLs rich microcapsule powders, MS/MD: modified starch/maltodextrin; GA/MD: gum Arabic/ maltodextrin; MIX: Mixed

3.12. Fourier transform infrared spectroscopy (FTIR)

FTIR analysis was used to investigate the characteristics of MS/MD, GA/MD, and MIX microcapsules containing MLCTs rich SLs. The spectra displayed in **Fig. 2** included fish oil, interesterified product (MLCTs rich SLs), MD, GA, MS, and the microcapsules (MS/MD, GA/MD, and MIX). Fish oil and MLCT-rich SLs exhibited absorbance at 720 and 2853 cm^{-1}, indicating the presence of DHA in unencapsulated fish oil and MLCTs rich SLs. The spectral range spanning from 2923 to 2852 cm^{-1} indicated the presence of PUFA, while wavelengths falling between 1743 and 1746 cm^{-1} suggested the presence of ester carbonyl (Bekhit et al., 2014; Sinclair et al., 1952). Additionally, the regions around 720 cm^{-1} indicated the presence of FAs with cis double bonds in unencapsulated fish oil and MLCTs rich SLs (Karthik & Anandharamakrishnan, 2013). Additionally, fish oil showed a flat O-H stretching at 3425 cm^{-1}, whereas MLCTs rich SLs showed a slight bend at the same wavelength. The band observed at 1158 cm^{-1} in both fish oil and MLCTs rich SLs corresponded to the C-O-C stretching (Bhandari & Howes, 1999; de Barros Fernandes et al., 2016). The MD spectrum showed absorption bands at 3290 cm^{-1} (O-H

stretching), 2928 cm^{-1} (C-H stretching from the carboxylic group), 1644 cm^{-1} (C=O stretching), 1355 cm^{-1} (O-H bending), and 1155, 1079, 1007 cm^{-1} (C-O stretching and C-O-H bending). These findings are similar with those reported by Chew et al. (Chew et al., 2018). Similarly, GA displayed absorption bands at 3278 cm^{-1} (O-H stretching), 2918 cm^{-1} (C-H stretching from the carboxylic group), 1604 cm^{-1} (C=O stretching or N-H bending), 1410 cm^{-1} (CH3 bending and C-H bending), and 1020 cm^{-1} (C-O stretching), similar results were reported by Chew et al. (Chew et al., 2018). The FTIR spectra revealed absorption bands of MS at specific wavenumbers: 3275 cm^{-1} (O-H stretching), 2938 cm^{-1} (C-H stretching from the carboxylic group), 1641 cm^{-1} (C=O stretching), 1335 cm^{-1} (CH3 bending), and 1145 cm^{-1}, 1074 cm^{-1}, and 993 cm^{-1} (C-O stretching and C-O-H bending). These findings were similar with previous studies (Chew et al., 2018; Hosseinnia et al., 2017), indicating consistency in the observed spectral characteristics of MS.

The presence of bands at 1743, 1158, 2853, and 2922 cm^{-1} in fish oil and MLCTs rich SLs was notably reduced in MS/MD, GA/MD and MIX-microencapsulated samples, suggesting successful encapsulation of the oil into the microcapsules (Hu et al., 2016). In this study, all microcapsule powders exhibited prominent hydroxyl peaks (O-H stretching) at 3288 cm^{-1}, as well as carboxylic group peaks (C-H stretching) at 2925 cm^{-1}. The occurrence of C=O stretching and C-O-H bending at 1155, 1079, and 1007 cm^{-1} was observed in all formulations (Chew et al., 2018; Hosseinnia et al., 2017; Mahdi et al., 2020). MS/MD and MIX-based microcapsules exhibited profiles closely resembling gum Arabic, with gum Arabic being the predominant component in these formulations. The absence of amide groups were observed in all the formulations it may be due to the absence of whey protein (Mahdi et al., 2020).

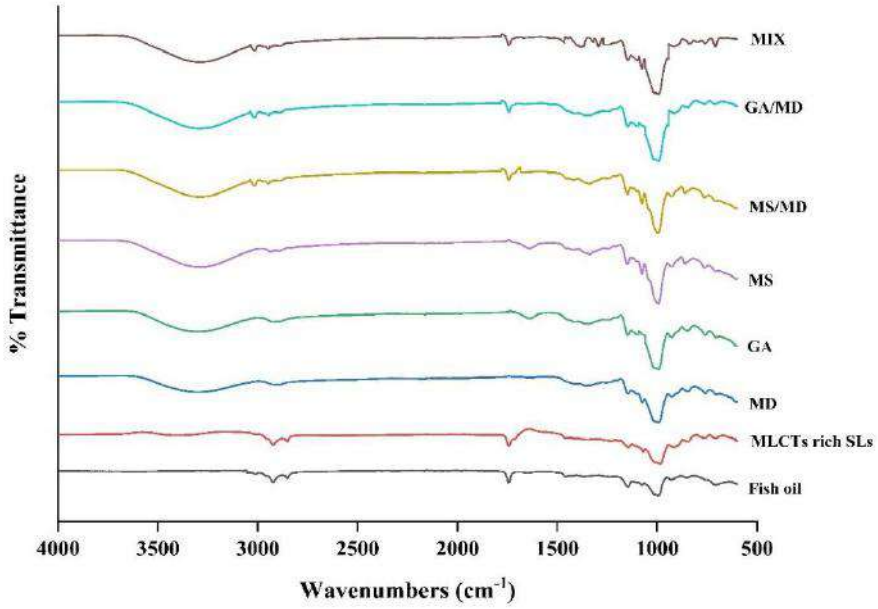

Fig. 2. FTIR spectra of raw materials (gum Arabic (GA), maltodextrin (MD), modified starch (MS), the microcapsules (MS/MD, GA/MD, MIX) MLCTs rich SLs and fish oil..

3.13. Scanning electron microscope (SEM)

SEM analysis was conducted on the microencapsulated MLCTs rich SLs powders, as depicted in **Fig. 3**. The purpose was to examine for any potential defects like fractures or cracks that could compromise the coherence of the encapsulated material, certain that such defects could lead to degradation and oxidation (Guadarrama-Lezama et al., 2012). The SEM micrographs revealed spherical and semi-spherical shapes of the microcapsules, although spray-dried particles typically exhibit a spherical shape with a mean size ranging from 10 to 100 mm (Fang & Bhandari, 2010). Variations in morphologies and irregular surfaces were likely influenced by differences in feeding ratio, droplet size, and drying temperature during the process. Shrinkage followed by potential expansion could change particle size and lead to broken shells. Moreover, surface irregularities on the microcapsules could enhance dispersibility and rehydration of the powders (Guadarrama-Lezama et al., 2012). MLCTs rich SLs microencapsulated with MD/MS, GA/MD, and MIX did not exhibit any fissures or cracking. This suggests that microencapsulation

of MLCTs rich SLs with these wall materials could yield microencapsulated products with improved retention and stability. Hollow spheres, characteristic of spray-dried powders, were observed in MIX microcapsules (Hu et al., 2016). These findings are similar with previous studies using GA as carriers, they also observed spherical shapes and irregularities due to shrinkage in various SEM studies (Chew et al., 2018; Daza et al., 2016; de Barros Fernandes et al., 2016). Similar morphological characteristics in all microcapsules indicate a uniform drying process, while surface shrinkage may result from the drying process.

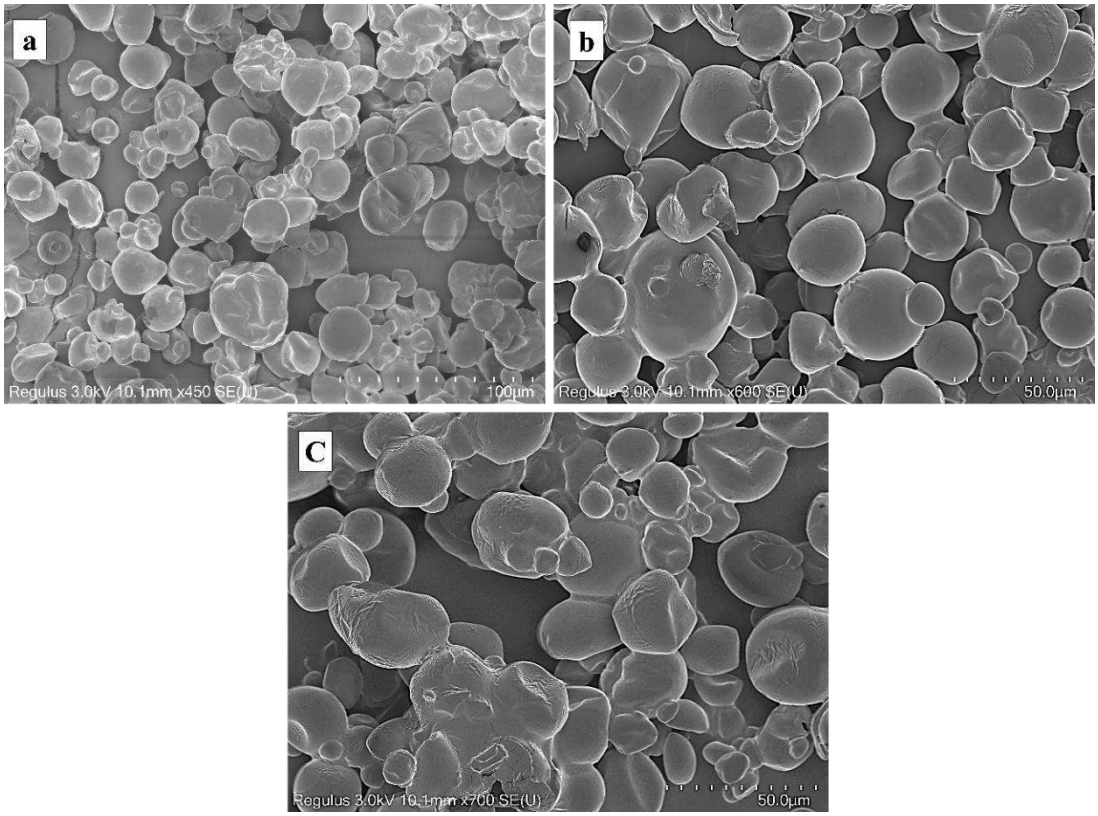

Fig. 3. SEM of MLCTs rich SLs microcapsule powders, (a) MS/MD, (b) GA/MD, (c) MIX: Mixed.

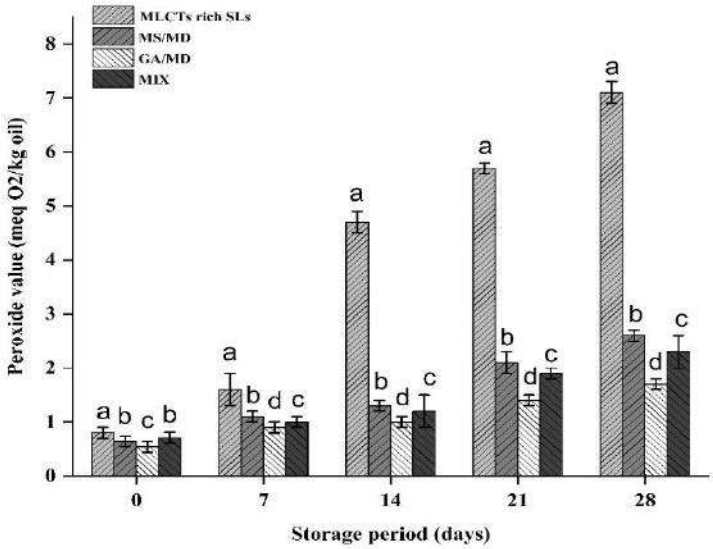

Fig 4. Peroxide values of microencapsulated MLCTs rich SLs and non-encapsulated SLs upon accelerated storage at 60 ˚C. MS/MD, GA/MD and MIX: Mixed

3.14. Oxidative stability under accelerated storage

The primary oxidative by-product of fat and oil is hydroperoxide, typically measured through peroxide value (PV) determination (Korma et al., 2019). The progression of peroxide values in both non-encapsulated MLCTs rich SLs and microencapsulated samples over seven-day intervals of storage at 60˚C is illustrated in **Fig 4**. At the beginning of storage, MLCTs rich SLs exhibited a PV of 0.81 meq O2/kg, which did not significantly differ ($p > 0.05$) from the PVs of microencapsulated oils, measuring 0.64, 0.55, and 0.72 meq O2/kg oil for MS/MD, GA/MD, and MIX microcapsules, respectively. These findings indicate that the applied spray drying method did not adversely affect the peroxidation of MLCTs rich SLs. However, the PV increased more rapidly in raw MLCTs rich SLs than in encapsulated samples over 14 days of storage. While slight increases in PV were observed in MS/MD and MIX microcapsules after 14 days, no significant variation was noted in GA/MD microcapsules. By day 28 of storage at 60˚C, the PV of MLCTs rich SLs reached 7.21 meq O2/kg oil, whereas those of MS/MD and MIX microcapsules were 2.62 and 2.33 meq O2/kg oil, respectively. These findings reveal that GA/MD microcapsules exhibited superior oxidative stability compared to MS/MD and MIX microcapsules. These results are consistent with those reported by Premi et al. (Premi & Sharma, 2017), who observed high oil stability of encapsulated drumstick oil powder (EDOP) with MD and GA. MLCTs rich SLs, being rich in unsaturated fatty acids (UFA), is susceptible to oxidative degradation, leading to the formation of free radicals (hydroperoxides) during processing and storage. This susceptibility may account for the increasing peroxide value of non-encapsulated MLCTs rich SLs during storage. Overall, the microencapsulation of MLCTs rich SLs improved its oxidative stability, with the greatest enhancement observed when carbohydrate-based materials were used.

IV. CONCLUSIONS

The MLCTs rich SLs were microencapsulated and spray-dried into a powder. Three treatments including MS/MD (2:1), GA/MD (2:1), and MIX (1:1:1) were used as wall material combinations, and all of them exhibited well visible microcapsules. The microcapsules produced using GA/MD were selected as the optimal encapsulation process due to their dominance across various properties. They exhibited the highest encapsulation yield and efficiency. Additionally, GA/MD microcapsules demonstrated the highest bulk, tapped, and particle density, indicating excellent storage stability. Conversely, they had the lowest porosity and required a shorter time for wettability, suggesting a strong ability to reconstitute the powders. Moreover, X-ray diffraction analysis revealed that GA/MD microcapsules exhibited the highest relative crystallinity, indicating higher storage stability compared to other formulations. Overall, GA/MD microencapsulation offers an efficient approach for developing MLCTs rich SLs microcapsules with enhanced stability, making them potentially valuable as functional food and pharmaceutical ingredients.

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The Effect Combination of Liquid Organic Fertilizer and Inorganic Fertilizer on Potato (*Solanum tuberosum* L.) var. Granola G2

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Abstract— *The potato (Solanum tuberosum L.) is a crucial food source highly demanded in Indonesian society. The national potato crop productivity in 2018 was approximately 1,284,773 tons. Enhancing potato availability necessitates increasing productivity by expanding cultivation areas and employing improved cultivation techniques. However, excessive use of inorganic fertilizers may adversely affect soil fertility and quality. Therefore, it is imperative to enhance soil fertility by augmenting organic matter content using organic fertilizers, such as liquid organic fertilizers derived from processed sugar cane residue. Consequently, this study aimed to ascertain the optimal dosage of liquid organic fertilizer for enhancing the growth and yield of potato plants. The research was conducted from June to October 2022 in Nongkojajar Village, Tutur District, Pasuruan Regency. Various tools were utilized, including hand tractors, nameplates, bamboo pegs, gembor, tape measures, a 150 L drum, a stirrer drill, 5 L and 250 mL measuring cups, waring, and a camera. Potato seeds of the granola variety, liquid organic fertilizer, inorganic fertilizers (Urea, SP-36, and KCL), as well as soil and water samples, constituted the materials for the research. The experimental research method was employed, utilizing an environmental randomized block design (RBD) with seven treatments repeated four times. Parameters observed encompassed plant height, number of leaves, leaf area, dry weight, plant growth rate, soil chemical analysis, nutrient absorption, and harvest analysis. The results revealed that the treatment of liquid organic fertilizer at a 100% dose and the combined treatment of liquid organic fertilizer at doses of 50%, 100%, 150%, and 200% + 80% inorganic fertilizer exhibited superior productivity in potato plants compared to the control treatment without fertilizer. Additionally, these treatments demonstrated comparable potato plant productivity to that achieved with the 100% standard dose of inorganic fertilizer.*

Keywords— ***Doses, Combination, Liquid organic fertilizer, Inorganic fertilizer***

I. INTRODUCTION

Potato are a staple food crop highly demanded in Indonesian society. Potatoes are typically cultivated in highland areas or at the foothills of mountains due to the specific growth requirements distinct from other food crops. Over time, potatoes have become a viable alternative commodity after rice, leading to an increasing demand for potato plants each year. This necessitates farmers to innovate to ensure that potato cultivation meets consumer needs [1].

Increasing potato plant production can be achieved, among other methods, by employing organic fertilizers to reduce the use of inorganic fertilizers. Organic fertilizers are divided into two types: solid organic fertilizers and liquid organic fertilizers. Liquid organic fertilizers are typically made from beneficial waste, such as agricultural or

industrial waste. One industrial waste that can be used to produce fertilizer is molasses, a by-product of the sugar cane industry. East Java is the province with the highest sugar cane production in Indonesia, yielding 1,164,691 tons, particularly in Malang with a production of 2,774 tons. This leads to a significant amount of sugar cane drip waste being generated. Molasses is a by-product created during the sugar manufacturing process [2]. Molasses is also utilized as a medium for bread yeast growth because it still contains sugar to support the fermentation process. Waste that has been used as a fermentation medium contains N, P, and K content that can be utilized as liquid organic fertilizer [3]. Liquid organic fertilizers rich in organic matter content can benefit potato plants. The use of liquid organic fertilizers during cultivation can help improve the three natural soil properties: physical, chemical, and biological. Therefore, this study aims to utilize liquid organic fertilizers at various doses during potato cultivation to determine the most effective dosage for potato plant cultivation.

II. MATERIALS AND METHODS

This research was conducted from June 2022 to October 2022 in Wonosari Village, Tutur District, Pasuruan Regency. The tools used in this study included a hoe, measuring cups, Leaf Area Meter (LAM), analytical balance, oven, ruler, and camera. The materials used in this research were potato seeds of the Granola variety, liquid organic fertilizer (molasses), inorganic fertilizers (Urea, SP-36, and KCl). The research utilized a Randomized Block Design (RBD) with a single factor, namely the combination of liquid organic fertilizer and inorganic fertilizer, comprising 7 treatments and 4 replications. The treatments used were as follows: T1: Control, T2: 100% Inorganic fertilizer, T3: 100% Liquid organic fertilizer + 80% Inorganic fertilizer, T4: 50% Liquid organic fertilizer + 80% Inorganic fertilizer, T5: 150% Liquid organic fertilizer + 80% Inorganic fertilizer, T6: 200% Liquid organic fertilizer + 80% Inorganic fertilizer, T7: 100% Liquid organic fertilizer.

The research involved two observed variables: growth and yield. Growth variables included plant height, number of leaves, leaf area, and dry weight of plants. Yield variables included the number of tubers per plant, total tuber weight per plot harvest, and production per hectare. The obtained observation data were analyzed using analysis of variance (ANOVA) at a significance level of 5%. If there was a significant effect, a comparison between treatments was conducted using Duncan's Multiple Range Test (DMRT) at a significance level of 5%. Relative Agronomic Effectiveness (RAE) is also calculated to determine the efficiency of the combination treatment of liquid organic and inorganic fertilizers compared to the farmer's standard treatment (1).

$$\text{RAE} = \frac{\text{Treatment} - \text{Control (T1)}}{\text{Farmer's Practice (T2)} - \text{Control (T1)}} \text{x } 100 \text{ (1)}$$

III. RESULTS AND DISCUSSION

Plant Height

Analysis of variance showed that at 4 and 6 weeks after planting (WAP), the organic fertilizer treatment did not significantly affect the mean height of potato plants. However, the organic fertilizer treatment and combinations with inorganic fertilizer showed significant effects at 8 and 10 WAP. Table 1 presents the analysis of variance for the mean plant height at 8 WAP. The combination treatment of 200% inorganic fertilizer without potassium, combined with 100% liquid organic fertilizer and 80% inorganic fertilizer, exhibited a significantly different mean plant height compared to treatments without fertilizer, 100% liquid organic fertilizer + 80% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, 150% liquid organic fertilizer + 80% inorganic fertilizer, and 100% liquid organic fertilizer. Additionally, the treatment of 200% liquid organic fertilizer + 80% inorganic fertilizer showed a greater mean plant height compared to the treatment of 100% liquid organic fertilizer. At 10 WAP, the combination treatment of liquid organic fertilizer with inorganic fertilizer without potassium (100% liquid organic fertilizer + 80% inorganic fertilizer and 200% liquid organic fertilizer + 80% inorganic fertilizer) exhibited a significantly different mean plant height compared to treatments without fertilizer and 100% liquid organic fertilizer but did not significantly differ from the 100% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, and 150% liquid organic fertilizer + 80% inorganic fertilizer treatments. The treatments of 100% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, and 150% liquid organic fertilizer + 80% inorganic fertilizer showed a significantly different mean plant height compared to treatments without fertilizer but did not significantly differ from the 100% liquid organic fertilizer treatment. This is because the nutrient availability is fulfilled through the application of inorganic fertilizers as well as the role of liquid organic fertilizers that can maximize nutrient absorption and availability in the soil by improving the physical, chemical, and biological properties of the soil [4]. The application of organic fertilizer can increase the soil's organic matter content, making it more porous so that plant roots will be better and more efficient in nutrient absorption [5].

Number of Leaves

Table 1. Potato Plant Height in Combination of Liquid Organic Fertilizer and Inorganic Fertilizer

No	Treatment	Plant Height (cm)			
		4 WAP	6 WAP	8 WAP	10 WAP
1	T1	1.96	10.13	33.60 a	42.92 a
2	T2	4.29	14.96	42.70 bc	60.05 bc
3	T3	2.98	16.68	38.92 ab	60.58 c
4	T4	2.38	14.16	39.31 ab	58.25 bc
5	T5	2.37	13.95	37.81 ab	56.83 bc
6	T6	3.00	14.28	48.07 c	61.09 c
7	T7	4.42	13.80	36.78 ab	55.66 b

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

Analysis of variance in Table 2 showed that at 4 WAP, treatments without fertilizer, 100% liquid organic fertilizer + 80% inorganic fertilizer, 100% liquid organic fertilizer + 80% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, 200% liquid organic fertilizer + 80% inorganic fertilizer, and 100% liquid organic fertilizer exhibited mean leaf numbers that did not significantly differ from the 100% inorganic fertilizer treatment. At 6 WAP, the treatments of 100% liquid organic fertilizer + 80% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, and 200% liquid organic fertilizer + 80% inorganic fertilizer showed mean leaf numbers that significantly differed from treatments without fertilizer, 100% inorganic fertilizer, and 100% liquid organic fertilizer. However, the treatments of 100% liquid organic fertilizer + 80% inorganic fertilizer and 50% liquid organic fertilizer + 80% inorganic fertilizer exhibited lower mean leaf numbers compared to higher doses of organic fertilizer (150% liquid organic fertilizer + 80% inorganic fertilizer and 200% liquid organic fertilizer + 80% inorganic fertilizer). At 8 WAP, treatments of 100% liquid organic fertilizer + 80% inorganic fertilizer, 50% liquid organic fertilizer + 80% inorganic fertilizer, 150% liquid organic fertilizer + 80% inorganic fertilizer, 200% liquid organic fertilizer + 80% inorganic fertilizer, and 100% liquid organic fertilizer showed mean leaf numbers that did not significantly differ from the 100% inorganic fertilizer treatment. At the final observation time, 10 WAP, the combination treatment of 200% liquid organic fertilizer + 80% inorganic fertilizer showed a significantly different mean leaf number compared to other treatments. The treatment of 100% liquid organic fertilizer + 80% inorganic fertilizer exhibited a significantly different mean leaf number compared to treatments without fertilizer, 100% inorganic fertilizer, and 100% liquid organic fertilizer. The treatments of 50% liquid organic fertilizer + 80% inorganic fertilizer and 150% liquid organic fertilizer + 80% inorganic fertilizer did not significantly differ from the 100% liquid organic fertilizer and 100% inorganic fertilizer treatments. The treatments of 50% liquid organic fertilizer + 80% inorganic fertilizer and 150% liquid organic fertilizer + 80% inorganic fertilizer significantly differed from the treatment without fertilizer but did not significantly differ from the 100% liquid organic fertilizer treatment. At the final observation time, the combination treatment of 200% liquid organic fertilizer + 80% inorganic fertilizer showed a greater mean leaf number compared to other treatments. The application of liquid organic fertilizer can enhance soil organic matter content, which has various benefits for soil fertility. High soil organic matter content benefits plants by providing available nutrients and improving soil physical properties, such as structure, aggregation, infiltration, and water retention [6].

Table 2. Number of Potato Leaves in Combination of Liquid Organic Fertilizer and Inorganic Fertilizer

No	Treatment	Number of Leaves			
		4 WAP	6 WAP	8 WAP	10 WAP
1	T1	3.00 a	7.28 a	15.88 a	22.12 a
2	T2	3.67 ab	9.78 b	31.67 b	50.67 b

3	T3	3.54 ab	12.01 c	28.24 b	53.79 c
4	T4	4.00 b	13.94 d	26.06 b	49.30 b
5	T5	4.12 b	16.46 e	27.56 b	51.62 bc
6	T6	4.12 b	17.08 e	31.63 b	60.00 d
7	T7	3.64 ab	9.94 b	25.91 b	49.04 b

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

Table 3. Potato Leaf Area in Combination of Liquid Organic Fertilizer and Inorganic Fertilizer

No	Treatment	Leaf Area (cm^2 $plant^{-1}$)			
		4 WAP	6 WAP	8 WAP	10 WAP
1	T1	138.68 a	380.65 a	1289.28 a	1853.54 a
2	T2	182.06 bc	734.42 b	2447.37 c	4979.15 c
3	T3	176.24 bc	946.01 c	2371.71 c	4904.44 c
4	T4	184.57 bc	1076.33 c	2106.46 bc	4386.68 bc
5	T5	201.62 c	1340.61 d	2095.49 bc	4822.84 bc
6	T6	199.67 c	1340.58 d	2107.93 bc	5997.62 d
7	T7	151.46 ab	682.82 b	1917.88 b	4081.38 b

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

Dry Weight of Plants

Based on the analysis of variance, there were differences in the dry weight of potato plants at 4-10 WAP, as presented in Table 4. At 4 WAP, the treatment combination of liquid organic fertilizer and inorganic fertilizer at 50% LOC + 80% inorganic-K and 200% LOC + 80% inorganic-K showed significantly different mean dry weights compared to the treatment without fertilizer and 100% LOC. At 6 WAP, the 200% LOC + 80% inorganic-K treatment exhibited significant differences compared to the treatment without fertilizer, 100% LOC + 80% inorganic-K, 50% LOC + 80% inorganic-K, 150% LOC + 80% inorganic-K, and 100% LOC, but did not differ significantly from the 100% inorganic treatment. At 8 WAP, the treatments 100% LOC + 80% inorganic-K and 200% LOC + 80% inorganic-K showed significantly different mean dry weights compared to the treatment without fertilizer and the 100% LOC treatment. At 10 WAP, the 200% LOC + 80% inorganic-K treatment showed significantly different mean dry weights compared to the treatment without fertilizer, 100% inorganic, and 100% LOC treatments.The application of organic fertilizer in soil can also aid in nutrient absorption, especially nitrogen, by reducing mineral leaching [7]. Nitrogen plays the most crucial role in various physiological processes such as imparting green color to plants, increasing leaf and stem numbers, as well as the growth and development of other vegetative parts [8].

Plant Growth Rate

Based on the analysis of variance, the growth rate of potato plants at 4-10 WAP showed significant differences. At 4 WAP, the treatments 100% LOC + 80% inorganic-K and 200% LOC + 80% inorganic-K exhibited significantly different mean growth rates compared to the treatment without fertilizer, 100% inorganic, 50% LOC + 80% inorganic-K, 150% LOC + 80% inorganic-K, and 100% LOC. At 8-10 WAP, the combination treatments of 150% LOC + 80% showed significantly different plant growth rates compared to the other treatments. Treatments without fertilizer, 100% LOC + 80%, 50% LOC + 80%, 200% LOC + 80%, and 100% LOC showed no significant difference in plant growth rate compared to the 100% inorganic treatment. The application of liquid organic fertilizer affects potassium uptake by plants and increases the potassium content in the soil. High potassium uptake indicates that the

application of liquid organic fertilizer can help fulfill potassium nutrient requirements for plants [9].

Yield Components

Based on the analysis of variance, the fertilizer treatments showed significant effects on the number of tubers per plot, tuber weight per plot, and tuber yield per hectare. Table 6 shows that the combination treatment of liquid organic fertilizer and inorganic fertilizer at 200% LOC + 80% inorganic-K significantly differed in mean tuber number compared to the treatment without fertilizer, 100% inorganic, and 100% LOC. For tuber weight and tuber yield variables, the treatments 100% LOC + 80% inorganic-K, 50% LOC + 80% inorganic-K, 150% LOC + 80% inorganic-K, and 200% LOC + 80% inorganic-K showed significantly different production results compared to the treatment without fertilizer but did not significantly differ from the 100% inorganic and 100% LOC treatments. Potassium is a vital nutrient for potato plants in the growth process and supports higher plant yields. [10] explain that potatoes are high-starch-yielding crops, so a high amount of potassium is needed during the relatively short period of tuber growth and development, indicating that the more potassium available, the better the potato tuber yield. For the RAE value, treatments of 100% LOC + 80% inorganic-K, 50% LOC + 80% inorganic-K, 150% LOC + 80% inorganic-K, and 200% LOC + 80% inorganic-K are effective in replacing some doses of inorganic fertilizer, as indicated by Relative agronomic effectiveness (RAE) values of over 95%. The use of liquid organic fertilizer can provide several benefits for plant growth, including reducing dependence on inorganic fertilizers and increasing crop productivity [11]. Liquid organic fertilizers are generally made from natural materials such as compost, manure fertilizers, or fermented crop residues [12]. The use of liquid organic fertilizer can enhance soil fertility, increase nutrient availability to plants, and improve beneficial soil microbe activity [13]. Research by [14] states that the effect of liquid organic fertilizer on potato plant growth and yield. The results indicate that the use of liquid organic fertilizer can significantly reduce the use of inorganic fertilizers without reducing crop yields. Furthermore, [15] state the use of liquid organic fertilizer in potato production and its impact on soil properties. The results show that liquid organic fertilizer can increase potato plant productivity and reduce dependence on inorganic fertilizers.

Table 4. Dry Weight of Potatoes in Combination with Liquid Organic and Inorganic Fertilizers

No	Treatment	Dry Weight (g plant^{-1})			
		2 WAP	4 WAP	8 WAP	6 WAP
1	T1	0.63 a	3.39 a	20.83 a	54.76 a
2	T2	1.66 b	13.21 de	44.94 ab	90.29 cd
3	T3	1.28 ab	8.84 bc	51.36 b	83.09 c
4	T4	1.95 b	8.99 bc	41.94 ab	89.81 cd
5	T5	0.80 a	10.90 cd	26.18 a	99.13 de
6	T6	1.73 b	13.89 e	59.98 b	106.14 e
7	T7	0.75 a	7.18 b	22.55 a	67.84 b

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

Table 5. Growth Rate of Potato Plants in Combination with Liquid Organic and Inorganic Fertilizers

No	Treatment	Crop Growth Rate (g m^{-2} week^{-1})		
		2-4 WAP	4-6 WAP	6-8 WAP
1	T1	0.82 a	5.19 a	10.10 ab
2	T2	2.94 c	9.44 b	13.50 ab
3	T3	2.27 b	12.66 c	9.44 a
4	T4	2.10 b	10.06 b	14.50 b
5	T5	3.00 cd	4.55 a	21.71 c

6	T6	3.62 d	13.72 c	13.74 ab
7	T7	1.91 b	4.33 a	13.47 ab

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

Table 6. Potato Yield in Combination with Liquid Organic and Inorganic Fertilizers

No	Treatment	Number of Tubers per Harvest Plot	Yield (ton ha^{-1})	RAE (%)
1	T1	110.50 a	39.25 a	-
2	T2	150.29 bc	50.41 c	-
3	T3	160.50 cd	51.58 c	110.49
4	T4	157.89 bcd	51.54 c	110.06
5	T5	168.00 cd	52.73 c	120.77
6	T6	175.75 d	53.03 c	123.46
7	T7	135.50 b	46.78 b	67.46

Note: T1 (Control), T2 (100% Inorganic), T3 (100% LOC + 80% Inorganic-K), T4 (50% LOC + 80% Inorganic-K), T5 (150% LOC + 80% Inorganic-K), T6 (200% LOC + 80% Inorganic-K), T7 (100% LOC). Mean values followed by the same letter in the same column indicate no significant difference at DMRT 5%.

IV. CONCLUSION

The addition of liquid organic fertilizer doses of 50%, 100%, 150%, and 200% with 80% inorganic fertilizer was able to provide results similar to the use of 100% inorganic fertilizer and significantly influences various aspects of potato plant growth and yield. The application of 100% liquid organic fertilizer alone without inorganic fertilizer did not able to achieve the same productivity as 100% inorganic fertilizer. The use of liquid organic fertilizer at a 50% dose is the optimum dosage because it demonstrates the same efficiency as higher doses (100%, 150%, and 200%) in replacing the use of inorganic fertilizer by 20%, thus reducing the amount of fertilizer used to 80% of the total required dose.

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Analyzing of Agrotourism Potential in Malang City

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Abstract— *Agrotourism, in addition to being a tourist and learning place, also functions as generate income for the Regional Original Revenue. Malang City which most of its area is settlement, can be studied as an agro-tourism based on urban agriculture. The research was conducted in 4 Sub District of Kedungkandang District with microclimate characteristic as follow temperature (22.7- 25.1 °C) and humidity (79 - 86%) support the agro-tourism sector. Research was started from May - September 2022. The strong potentials of the selected locations are (1) Land owned by the Malang City government so that the possibility of using it for agro-tourism areas is more open; (2) At the moment a little paddy-based agro-tourism is integrated with sheep farming and fisheries; (3) There is a tour of the Masked Village in the Northeast aerial photo taking location; (4) There are many potential educational locations to be targeted for marketing agrotourism-based edutourism; and (5) Enthusiastic community. The method of survey was direct observation and then analyzed using SWOT method to find high potential agrotourism areas. The conclusion obtained was the design of agrotourism development in Tlogowaru Sub District is divided into two functions, namely (1) lodging and trade functions, (2) agriculture, animal husbandry and fishery education areas.*

Keywords— ***Agro-tourism, Agriculture, Mapping, Paddy Field***

I. INTRODUCTION

Malang is the most potential city in East Java for agro-tourism because of its flourishing biosphere, climate and supportive astronomy position based on "Kota Malang dalam Angka 2018". Malang City has total of 2,742 ha of agricultural land, both paddy fields and non-paddy fields [1]. It is located at 112.06° - 112.07° East Longitude and 7.06° - 8.02° South Latitude, yield a tropical city with a wide variety of agricultural product. Surrounded by inactive mountains on its geographical location, namely Mount Arjuna-Welirang in the north, Mount Semeru in the east, Mount Kawi, Butak and Panderman in the west and Mount Kelud in the south (active), makes Malang a chill, beautiful natural charm to be used as a spotlight. According to [6] in the current era of globalization, tourism is one of the activities that has a strategic role in supporting national economic development and agro-tourism is included.

In agro-tourism sector, Malang is well-known for its experiential tourism based on agro- production, such as plant-nurseries, planting, harvesting crops and fruits, as well as fish- nurseries, dairy process, and others. According to [2] agritourism is any custom developed on agricultural land that has the aim of engaging tourists. The agrotourism model leads to learning about producing in addition to selling its own products. Apart from being a tourist spot, agro-tourism is a place of learning as well as a place for activities that can generate income for Regional Original Revenue. Agritourism is an existing opportunity for farmers to secure assets and the National Gross Domestic Product because it becomes an additional source of income and employment opportunities for traditional farmers. Malang, which is mostly residential area, can be studied as urban agriculture-based agrotourism. This research was conducted with the aim of conserving rice fields by using it as agro- tourism, increasing the income of farmers and communities around the agro-tourism location, and increasing agritourism in Malang City. The target in this research is farmer groups and agricultural land in Malang City..

II. METHOD ALSO CALLED MATERIALS AND METHODS OR EXPERIMENTAL METHODS

This research was conducted in Malang City, which is located at 440 - 667 meters above sea level and one of the tourist destination in East Java. It is located in the middle of Malang Regency, geographically it is located at 112,06° – 112,07° East Longitude and 7,06° – 8,02° South Latitude, with the northern boundary are Singosari and Karangploso District, Malang Regency, to the east is Pakis Subdistrict and Tumpang District, Malang Regency, to the south with Kerajinan District and Pakisaji District, Malang Regency and to the West with Wagir District and Dau District, Malang Regency. The image of Malang City area is presented in Figure 1.

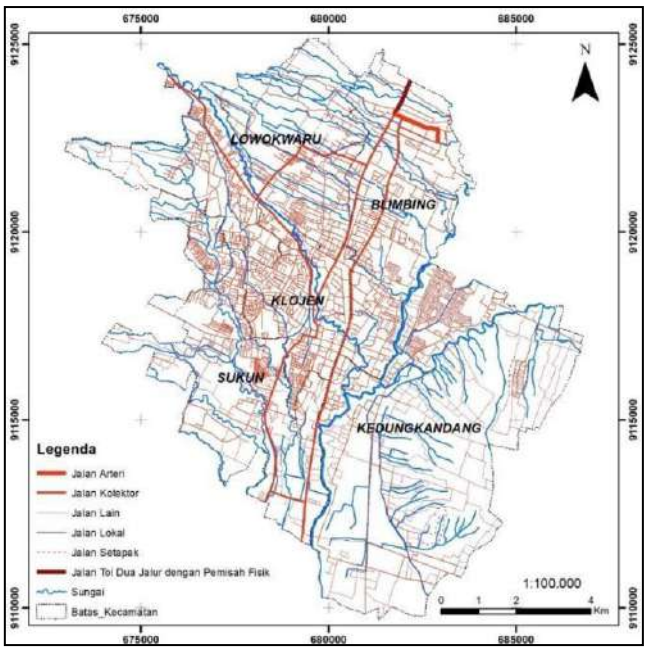

Fig.1. Administration Map of Malang City, with a scale of 1: 100.000

The location of groundcheck and sampling is determined by a map from digitizing google earth. The survey aims to collect primary data from direct-observations, including agricultural land conditions (including food crops, livestock, horticulture and plantations), socio-economic conditions of farming communities, conditions of agricultural supporting infrastructure, and other data in accordance with the terms and criteria. Researchers used SWOT analysis techniques to find out the strengths, weaknesses, threats and opportunities in the formation of agro-tourism in Malang. Analysis of the internal and external environment is an essential task in agritourism development. Furthermore, to formulate a tourism village model, the researcher rests on the data from the mapping of the potential and the formulation of the strategy which is elaborated into a more operational concept scheme.

III. RESULTS AND DISCUSSION

3.1 SWOT Analysis in Determining Potential Areas of Agro-tourism

There are 4 areas that have the potential for agro-tourism. The land that has the potential to be developed into an agro-tourism area is located in Kedungkandang District (Bumiayu, Cemorokembang, Arjowinangun and Tlogowaru Sub District). The selection of Kedungkandang District to be the research location begins with a SWOT analysis in 5 districts, namely Kedungkandang, Sukun, Lowokwaru, Blimbing and Klojen.

Fig.2. Bumiayu Sub District

Bumiayu Sub District has an agricultural stretch of 134 ha with a shape extending from north to south along the Mayjend Sungkono Road. In the western part there is a residential area and the southeast agricultural area in this district is colored yellow, which means it's included in the settlement plan (Figure 2).

Table 1. SWOT Analysis of Bumiayu Sub District

Strength	• There are 134 ha of active rice fields • The area located close to main roads as well as trade and service areas • There is land with attractive contours along the river body
Weaknes s	• Based on the RDTRK (Detailed City Spatial Plan) map, the agricultural area in this village is included in the residential zone • View of agricultural area covered by buildings • The agricultural area enters the river boundary so that high buildings cannot be made
Opportun ities	• Agricultural areas along the river body so that agrotourism areas can be designed with attractive contours and views

	• An agrotourism area that is close to settlements so that it can facilitate access
Threat	• The large number of settlement zoning in this district can lead to isolated and undeveloped agrotourism areas • The large number of residential areas can cause pollution of irrigation channels that irrigate agrotourism areas

Cemorokembang Sub District is also an area that has the potential of agrotourism. It has an agricultural area of 90.8 ha located in the Southeast part of Cemorokandang sub-district. In the southwest part of the village, there is an exit from the Malang Surabaya toll road. In the Malang Spatial Pattern Plan, agricultural land in Cemorokandang is included in the Residential Area Plan (Figure 3).

Fig.3. Cemorokembang Sub District

Table 2. SWOT Analysis of Cemorokembang Sub District

Strength	• There is an active paddy field of 90.8 ha • There is toll road access • Close access to Sawojajar village which is densely populated
Weaknes s	• Based on RDTRK, there are many zoning for settlements in this sub- district • Access near toll roads could cause congestion • Lack of natural landscape views
Opportun ities	• Because it is close to a densely populated area (Sawojajar), the agro-tourism area in this district can invite many visitors • Toll road access makes it easier for out-of-town visitors to come
Threat	• The scarcity of agricultural land due to development plans in this district • The existence of toll road access can cause congestion and make it difficult to enter the agrotourism area • Overcrowding can cause pollution to the agrotourism area

Arjowinangun Sub District is a village that is included in the potential for agro-tourism based on the results of the SWOT that has been carried out. has an agricultural area of 101 ha located in the southern part of the Arjowinangun sub-district. In the Spatial Pattern Plan in southeast Malang, the agricultural area of Arjowinangun sub-district is included in the residential area plan. In addition, there is also an Industrial area plan (Gray Color) which can pollute the surrounding agricultural area (Figure 4).

Fig.4. Arjowinangun Sub District

Table 3. SWOT Analysis of Arjowinangun Sub District

Strength	• There is 101 ha of active rice fields • There is land with attractive contours along the tidal mouth of the river • Many access from residential areas • There is a trade and service area on the main road
Weakness	• Based on RDTRK, there are many zoning for settlements in this Sub District • There is an industrial area zoning in this Sub

	District • The density is quite high in this area
Opportunities	• The location is close to the river so that an attractive area can be created with the tidal/estuary concept • The number of trade and service areas so that the supply of agro-tourism needs is quite easy to access
Threat	• The existence of an industrial area plan can provide a large enough pollution for agro-tourism areas • The large number of settlement zoning in this Sub District can lead to isolated and undeveloped agro-tourism areas

Tlogowaru Sub District has 128 ha of agricultural land spread across the Northeast and the Southern part of the it. In the Southeast Malang Spatial Pattern Plan, there are still several agricultural zone plans in this area. In addition, there is an fish hatchery research institue that has the potential to be developed into an agrotourism area complex (Figure 5).

Fig.5. Tlogowaru Sub District

Table 4. SWOT Analysis of Tlogowaru Sub District

Strength	• There is 128 ha of active rice fields • There is a 13 hectare stretch of agricultural land in the middle of this Sub District • There is an fish hatchery research institue (UPT Pembenihan Ikan) which is well known and used for various activities • There is Agricultural Extension Center (BPP) which supports agriculture in the Tlogowaru village
Weaknes s	• Based on RDTRK, the zoning of agricultural areas is still quite large in this Sub District • The location is far from urban areas and borders the district • The location is quite far from the main road
Opportunities	• The location of the land in the form of a large expanse can provide a rural atmosphere in the agro-tourism area • The existence of fish hatchery research institue can support agro-tourism areas in the fisheries sector • The contour landscape so that the scenery in this area is quite interesting • There is Agricultural Extension Center Tlogowaru which can assist in the extension process of the development of the agrotourism area
Threat	• The location is far enough from the main road so that it requires special attraction for the area • Settlement zones that surround agrotourism areas can isolate the area

3.2 Agrotourism Development Plan

The result of this study show that Tlogowaru Sub District is the most potential as an agrotourism area. The land belonging to residents around this area is also designed to become a well-integrated agro-tourism area. The potentials characteristic of the selected locations are (1) The land is belongs to the Malang City Government so that the use for agrotourism areas is more flexible; (2) Currently there are not many lowland rice-based agrotourism which is integrated with sheep farming and fisheries such as in Tlogowaru Sub District; (3) There is a tour of Topeng Village to the Northeast of the location for taking aerial photographs; (4) There are some schools that are potential to be targeted as agrotourism market; and (5) Enthusiastic society. [8] state that the area to be managed for agro-tourism should have unique areas and support from the surrounding community. Ownership of land to be used as an agro-tourism area also adds value because it is owned by the government. [11] states that location and material sources are internal factors of the competitive factors of a tourist area.

Main commodity is paddy which are planted 2-3 times a year. According to [10], agricultural commodities can be used as agrotourism objects, for example pre and

post-harvest can be an attractive activities. Paddy field in Tlogowaru has been integrated with fisheries and according to [3], it is necessary to combine agriculture with hobby, such as combination of rice field with jogging tract, fishing pond, integrated rice and fish farming, fish pond, flowers and horticultural crops. If this area will be developed as agrotourism, there is support from several nearby tourist locations such as Watu Menyan Nature Tourism and Jenon Water Resources in the southeast; Tumpang Valley and Topeng Village Tourism in the Northeast; Buring Education Tour, Coban Amprong, Keramat Village, Tridi Village and several other tours in the Northwest; Lastly Bonderland on the Southwest.

The development of this area has potency to lead to agrotourism due to the large amount of infrastructure that is very supportive at the specified location. There are Tlogowaru Fish Seed Center and also rice fields surrounding the selected location. According to [9] the attractiveness of a tourist object is based on: 1) The existence of resources that can cause a sense of pleasure, beauty and comfort, 2) High accessibility to be able to visit, 3) The existence of special characteristics/rare specifications, 4) Supporting facilities/infrastructure to serve tourists, 5) Natural attractions have high attractiveness because of their appearance and beauty and 6) Cultural tourism objects have high attractiveness. Therefore, the development of this area leads to agrotourism with the concept of rice field-based tourism and educational tourism on livestock and fisheries. Several areas have implemented agro-tourism development based on rice fields, one of which is in Sukorame Village, Lamongan. Tourism does not have to be in the form of natural objects, but innovation in various agricultural products can be a support for increasing tourist visits [7].

Although there are weaknesses and threats like the location far from the main road, this can also serve as an opportunity to increase the value of the destination. The location away from the city can provide a relaxing and enjoyable feel for travellers who want to take a break. According to [12] one of the most popular reasons for destination selection is a relaxing and pleasant destination with good weather. It can provide a relaxed, comfortable feel and relieve fatigue after work activities. Therefore, Tlogowaru Sub-district can utilise this weakness as an opportunity to become a tourist attraction.

3.3 Social Aspects

The social aspects in the study and design of the agrotourism potential of Malang City are the basis for determining how the carrying capacity of the community in the area is. The results obtained from distributing questionnaires to the community around Tlogowaru. The result show that the respondents are divided almost evenly with 52% women and 48% men who mostly work as farmers around the area. The people already know a lot about agrotourism planning in Malang with a percentage of 90% knowing and 10% not knowing.

Response to the facilities from community is 36% of the people choose a culinary center, then a swimming pool 24%, a gathering room / gazebo by 18%, a kiosk selling local products 16%, a hotel / villa 4% and a flower garden 2%. From these results, agrotourism design is prioritized for culinary areas that can accommodate local residents and tourists from outside. The community's suggestions for the government are to be more concerned with farmers and often held meetings with the rate of 77%, then 13% held competitions with the theme of agriculture or fisheries, then socialization about environmental cleanliness 7%, and 3% suggest repairing waterways that often leak. In developing this agrotourism object requires cooperation between the Malang City Government and the surrounding community. This is in line with [5] statement which argues that tourism / agrotourism development is all activities and efforts that are coordinated to attract tourists, providing all the necessary infrastructure and facilities, goods and services to serve the needs of future tourists. As for [5] which states that agrotourism development efforts in general cover aspects of human resource development, natural resources, promotion of support for facilities and institutions. The presence of various consumers / tourists will also be determined by the convenience created, starting from good service, easy access to accommodation and transportation to the awareness of the surrounding community.

Malang government is also planning to develop agro-tourism locations through several aspect such as provision of training related to proper cultivation techniques for the development of agricultural production, agribusiness for marketing agricultural products and funding for the repair and construction of facilities in Tlogowaru Sub-district. Government also plan to develop the tourism facilities and activities such as hotels, cafes, and restaurants that have a calm village atmosphere. Establishment of agricultural educational tourism activities such as livestock care, planting and harvesting agricultural products and fish farming. Expanding product offerings by omproving the relationship between agritourism and other economic sectors, applying the principles of sustainable development and agriculture and also developing agricultural products of Tlogowaru Sub-district. Marketing of agrotourism destination will be done by cooperating with the tourism department of Malang City to make Tlogowaru Sub-district known on a national scale, marketing through the internet and social media to promote the beauty of Tlogowaru

District agritourism also organising agricultural events to engage tourists.

IV. CONCLUSIONS

It can be concluded that Tlogowaru Sub District has potency to be established as rice field agrotourism. The development of this area has great potential due to the large amount of infrastructure that is very support at a predetermined location. The development of this area leads to agrotourism with the concept of rice field-based tourism and educational tourism on livestock and fisheries. The role of the community is also very influential on the successful development and potential of agrotourism in Tlogowaru Sub District, Malang City.

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Productivity of Arabica Coffee in Brawijaya University's Agroforestry

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Abstract— *Agroforestry is an agricultural system that combines annual crops with various woody or annual plants. The research carried out in arabica coffee agroforestry of Brawijaya University's forest with pines as shading plant. Differences in management mechanisms result in differences in arabica coffee production and environmental conditions in the four pine-arabica coffee agroforestry management models in UB forests. The research's purpose is to analyze production and biodiversity of arabica coffee in UB forests. Based on the results, it can be concluded that different levels of cultivation management have a significant influence on productive branches, number of unproductive branches, branch length, number of clusters each branch, and harvest yield. The average harvest yield BMP treatment was 3473.39 g each plant. The total diversity index value of understorey plants in UB Forest's coffee and pine agroforestry ranges from 1.4 – 2 (the diversity index values obtained for each plot from LC, MC, HC, and BMP management were (1.4), (1.52), (1.73), and (2.02)) with the value of H'= 1 – 3.322 (Shannon-Wiener diversity index). This result indicated that vegetation especially understorey diversity is classified as moderate.*

Keywords— ***coffee management, ecological services, understorey biodiversity***

I. INTRODUCTION

Indonesia's forests have abundant biodiversity. Apart from direct benefits of forest products (wood, rattan, etc.), other benefits that can be obtained are forests as ecosystem services. Ecosystem services are services received by humans from ecosystems (Agroforestri, 2013). Agroforestry is an agricultural farming system, which is a model that integrates annual crops with a variety of woody or annual plants.

Indonesia is the fourth largest coffee producing country in the world, after Brazil, Vietnam and Ethiopia (Dihni, 2020). Based on data from BPS (2021), coffee plant production in Indonesia in 2021 was 774.6 thousand tons. It is hoped that coffee cultivation using agroforestry can increased coffee production and better forest management. Forestry plants provide shelter and coffee plants become an additional source of income from non-timber plants that farmers can obtain. Coffee plants do not require direct light (100%), but the light intensity required is around 40 to 70% (Muschler, 2004). Shade trees also provide income for coffee farmers as a wood, firewood, fruit and others. Coffee and its shade trees contribute to ecosystem preservation, such as creating an environment for the conservation of certain bird species.

Agroforestry provides many important ecosystem services in supporting food security. Some of its roles are reducing run-off and erosion, relatively safer from the risk of harvest failure, and more stable against market fluctuations and the effects of climate change (Noordwijk *et al*., 2016). In East Java, non-forestry plants in agroforestry are very diverse, for example coffee, elephant yam, cocoa, corn, and vegetables. If the income from managing agroforestry is profitable, then the community's opportunity to convert forest land will be smaller. The ability to act as a buffer against biophysical, economic and social changes is

a key requirement for efforts to preserve and sustainably utilize agroforestry (Sulastri *et al*., 2022).

The aim of this research is to evaluate the productivity and environmental condition of pine-coffee agroforestry in UB forests. The four pine-coffee agroforestry management in the UB forest, Karangploso have differences in fertilizer management and shade tree management. In general, assessing ecosystem environmental services in coffee agroforestry can be carried out on 4 main services, namely provision services, regulatory services, supporting services and cultural services. (Abdul *et al*., 2015).

II. MATERIALS AND METHODS

The research was conducted in the UB Forest of Boro Sumbersari Hamlet, Tawangargo Village, Karangploso District, Malang Regency, East Java. Research was conducted on 4 pine-coffee agroforestry management (Low Coffee Management (LC), Medium Coffee Management (MC), High Coffee Management (HC), and Best Management Practice (BMP)). Observations used a nested design by comparing 4 agroforestry managements then tested with LSD (Least Significant Difference). Observation variables include coffee morphology and harvest (provider services) and environmental aspects (supporting services).

2.1 Morphology and Coffee Harvest Observation (Provider Services)

Morphological observations were carried out on plant height, number of leaves, leaf area, number of productive branches, number of unproductive branches, branch length, number of segments each branch, number of clusters each branch, and harvest.

2.2 Environmental Aspects (Support Services)

Observations of understorey plant carried out with determining the area and number of sample plots to be used. Furthermore, observations of understorey plant biodiversity were analyzed using the Sum Dominance Ratio (SDR) (Widaryanto *et al*., 2019) with the formula:

a. Density

- Absolute Density

$$KM = \frac{\text{Number of species discovered}}{\text{Number of plots}}$$

- Relative Density

$$KN = \frac{\text{Absolute density of that type}}{\text{Sum of the absolute densities of all types}} x100\%$$

b. Frequency

- Absolute Frequency

$$FM = \frac{\text{Plots where the species is found}}{\text{Total number of plots}}$$

- Relative Frequency

$$FN = \frac{\text{FM value}}{\text{Number of FM of all species}} x100\%$$

c. Dominance

- Absolute Dominance

$$DM = \frac{\text{Biomass value of the species found}}{\text{Total biomass of the entire sample area}}$$

- Relative Dominance

$$DN = \frac{\text{DM value}}{\text{Number of DM of all species}} x100\%$$

d. Important value index Index

$$IVI = KN+FN+DN$$

e. Summed Dominance Ratio (SDR)

$$SDRs = \frac{IVI}{3}$$

f. Shannon-Wiener diversity index (H ')

$$(H') = -\sum Pi \ln (Pi), \text{ where } Pi = \frac{ni}{N}$$

Information:

H ' = Diversity index

ni = Number of important value indices for each type

N = Total number of important value indices for all types

ln = Natural logarithm (natural number)

The value H ' < 1 indicates low species diversity, H ' = 1 to 3,322 indicates medium species diversity, H ' > 3,322 indicates high species diversity.

g. Simpson dominance index (C)

$$C = \sum \left(\frac{ni}{N}\right)^2$$

Information:

C = Simpson's dominance index

ni = Importance value index of each nth species

N = Total importance index of all species

The dominance index ranges from 0 – 1, if C = 0, it means that there are no species that dominate other species or the community structure is in a stable state. C = 1, meaning there is a species that dominates other species or the community structure is stable due to ecological pressure.

III. RESULTS AND DISCUSSION

3.1 Coffee Morphology and Harvest

Table 1. Arabica coffee morphology and harvest in agroforestry

P	PH	LA	PC	UPC	BL	S	BC	H
LC	381.00b	83.28a	30.83 b	26.09 c	68.68 a	21.73	6.31 a	831.56 a
MC	395.21b	80.07a	36.58 c	8.33 b	88.93 b	18,20	7.51 a	1460.26 b
H.C	405.81b	79.92a	37.04 c	9.88 b	79.82 b	19.09	6.34 a	1804.31 b
BMP	133.97a	100.37b	25.46 a	0.33 a	109.73 c	20.18	9.50 b	3473.39 c
BNT 5%	38.04	9.07	4.77	2.66	10.36	ns	1.53	484.70
KK	11.12	10.15	14.13	22.94	11.48	15.50	19.85	24.63

Information: P (treatment), PH (plant height, cm), LA (leaf area cm^2), PC (number of productive branches), UPC (number of unproductive branches), BL (branch length), S (number of segments per branch), BC (number of branch clusters), H (harvest, g $plant^{-1}$). Numbers accompanied by the same letter in the same column indicate that they are not significantly different based on the BNT test at the 5% level; ns = not significantly different.

The results show that different levels of management in coffee plant cultivation result in different growth and development of coffee plants, thus having an impact on differences in coffee plant production. Differences in management levels have an influence on almost all observed variables, including plant height, leaf area, number of productive branches, number of unproductive branches, and branch length. The influence of branch growth on coffee plants have an impact on the development of coffee plants. Different levels of management in coffee cultivation also have a significant effect on the development of coffee plants. Variables that show the influence of the level of management on the development of coffee plants include the results of the number of clusters each branch and the number of fruit each plant. The influence level of management on the development coffee plants has an impact on harvest yield or coffee fruit production each plant.

Different levels of management have a significant effect on yield each plant. The harvest results in the BMP treatment were able to provide higher yields compared to other treatments. BMP implementation is able to produce high yields each plant because the coffee plants in the BMP treatment are managed according to good coffee plant cultivation. Coffee plants in the BMP treatment were pruned so that coffee plants in the BMP treatment had an average plant height of around 150 cm. Pruning carried out on coffee plants can cause photosynthate results to accumulate in the development of flowers and fruit on coffee plants so that the number of coffee plant fruits in the BMP treatment is higher than in all treatments so that the comparison is straight with the harvest results obtained. Pruning carried out on coffee plants can redistribute the results of photosynthesis in the form of photosynthate leading to a large number of fruit each clusters which has an impact on higher coffee fruit production (Dufour *et al*., 2019).

Pruning coffee plants is useful in regulating the height of coffee plants, making it easier to maintenance and harvesting, forming new production branches for coffee plants, removing unwanted branches such as old branches, facilitating the interception of sunlight, improving air flow in the canopy, making it easier to control pests and diseases, reduce erratic changes in coffee crop yields (fluctuating), as well as reducing excessive fertilization (Hulupi and Martini, 2013). The research from Mulyono *et al*. (2016), shows that pruning coffee plants can produce a higher average percentage of normal beans and has a very significant effect on red fruit weight, green bean weight, normal bean percentage, round bean percentage, empty bean percentage, and coffee bean yield.

3.1 Environmental aspects

Table 2. Understorey biodiversity found on pine-coffee agroforestry

No	Types of Understorey	Plant classification	KM	KN (%)	FM	FN (%)	INP (%)
		LC PLOT					
1	*Ageratina riparia*	Ornamental plant	23.49	79.60	1.00	22.82	102.42
2	*Diplazium esculentum*	Vegetable crop	3.01	10.82	1.00	22.82	33.64
3	*Davallia denticulata*	Ornamental plant	1.03	3.70	0.83	18.84	22.55

4	*Histiopteris incisa*	Vegetable crops	1.03	3.64	0.75	16.85	20.49
5	*Cyperus melanospermus elatus*	Grass	1.24	3.61	0.75	17.92	21.54
		MC PLOT					
1	*Ageratina riparia*	Ornamental plant	14.40	59.60	1.00	24.87	84.48
2	*Rubia cordifolia*	Ornamental plant	9.88	34.36	0.75	17.21	51.58
3	*Diplazium esculentum*	Vegetable crops	1.98	8.49	0.83	20.48	28.97
4	*Histiopteris incisa*	Vegetable crops	0.58	2.48	0.68	17.83	20.32
5	*Cyperus melanospermus elatus*	Grass	2.00	8.23	0.40	10.32	18.56
		HC PLOT					
1	*Ageratina riparia*	Ornamental plant	3.83	29.28	0.83	18.42	47.70
2	*Diplazium esculentum*	Vegetable crops	4.91	40.45	1.00	23.18	63.64
3	*Davallia denticulata*	Ornamental plant	0.51	4.48	0.62	15.47	19.96
4	*Histiopteris incisa*	Vegetable crops	1.91	14.21	0.66	14.21	28.42
5	*Lantana spp.*	Ornamental plant	0.80	6.84	0.58	12.25	19.09
		BMP PLOT					
1	*Ageratina riparia*	Ornamental plant	9.35	52.56	1.00	16.82	69.39
2	*Diplazium esculentum*	Vegetable crops	1.27	7.76	0.91	15.09	22.85
3	*Urtica diocia*	Ornamental plant	1.63	12.87	0.56	9.98	22.95
4	*Centella asiatica*	Herbaceous plants	1.99	9.94	0.62	9.32	19.26
5	*Colocasia esculenta*	Tubers	0.59	3.51	1.00	14.83	18.34

Information: KM= absolute density, KN= relative density, FM= absolute frequency, FN: relative frequency, INP= important value index

Based on biodiversity result, the species that dominates in all agroforestry management plots is *Ageratina riparia* from the *Asteraceae* with an important value index of 102.42% in LC plots. This is also found in all existing management plots. This can be influenced by environmental factors such as temperature, light intensity and humidity of the surrounding air. Moenandir (1990) stated that what influences the number of species living in a community is light, where light greatly influences the type and the number of individuals that can grow in that place.

The other side, this can also be influenced by shading factors that exist on coffee and pine agroforestry and also influenced by the plant species themselves, such as in the process of seed dispersal. This is supported by Zulharman (2017), who gives the opinion that *Ageratina riparia* is a species found in shaded areas. *Ageratina riparia* is also a plant species that is usually found in mountainous areas of secondary forest and open or semi-open areas. Adult *Ageratina riparia* can produce 10,000 to 100,000 seeds each year. Usually species in the *Asteraceae* family are spread via wind and water.

1. Shannon-Wiener Diversity Index

Table 1. Shannon Wiener diversity index value (H')

PLOT	INDEX H'			
	M-1	M-2	M-3	M-4
LC	1.39	1.40	1.40	1.40
MC	1.49	1.52	1.53	1.52
HC	1.70	1.74	1.73	1.75
B MP	1.97	2.03	2.04	2.02

The diversity index values from each plot of LC, MC, HC, and BMP management was 1.4, 1.52, 1.73, and 2.02. From this value shows that the diversity of species on agroforestry is still in the medium category. This is in accordance with Indrawan *et al*. (2013), who explained that if the value of H' = 1-3.322 indicates that species diversity is classified as moderate.

2. Simpson Dominance Index

Table 2. Simpson dominance index value (C)

PLOT	INDEX C			
	M-1	M-2	M-3	M-4
LC	1	1	1	1
MC	1	1	1	1
H.C	1	1	1	1
B MP	1	1	1	1

Based on Table 3, the Simpson dominance index value (C) in all observation plots shows 1. Its mean that there is a dominant species in each plot. This can be seen in the LC, MC, HC and BMP management plots from the first to the last week of observation. This value means that there is species that dominate other species, or the community structure is stable due to ecological pressure (Fachrul *et al*., 2005).

The environmental conditions is the key to building up the understorey plant community that has grown. Environental variables in this research are intensity of sun light. Sun light was received by understorey plants can influence growth and can dominate understorey plant species in sample plots and in each different management plot. The results of the correlation between the Simpson dominance index show that there is no close relationship between understorey dominance and sun light intensity. This is not in accordance with the opinion of Indriyani *et al*. (2017), who explained that one of the environmental conditions that most influences the growth of plants are sunlight and shade.

IV. CONCLUSIONS

Based on the results, it can be concluded that the total diversity index value of understorey plants in UB Forest's coffee and pine agroforestry ranges from 1.4 – 2. Because of the value of H'= 1 – 3.322, its shows that the understorey community at the research location has medium level in vegetation diversity. The results of this study show that there is a diversity of different species in different agroforestry management, but the level of diversity is still the same or moderate.

The differences in cultivation management carried out in each treatment had a real influence on almost all observed variables. Different levels of cultivation management have a significant influence on the variables number of productive branches, number of unproductive branches, branch length, number of clusters each branch, and harvest yield. Different levels of cultivation management do not have a significant effect on the variable number of segments each branch on coffee plants. The results of coffee plant production can be seen in the yield each plant. The yield each plant in the BMP treatment gave higher yields than other treatments. The average harvest yield in BMP treatment was 3473.39 g each plant. BMP treatment is the best treatment that can be used as a recommendation for coffee cultivation management, especially in the UB Forest area.

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Impact of Climate Variation on Potato (*Solanum tuberosum* L.) based on Climate Projections until 2100

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Abstract— One of the challenges faced in cultivating potatoes is global warming, which has the potential to result in an increase in atmospheric temperatures, as well as changes in various climate variabilities that can affect the planting season, growth, and crop yields. This research aims to project an increase in atmospheric temperature until 2100 in two production center areas based on climate projection models in three Representative Concentration Pathways (RCP) scenarios, as well as projecting potato crop productivity in these two locations based on climate projection results. This research was carried out by examining climate variations at two location points, namely Tosari District and Poncokusumo District, and was conducted in May-October 2023. The tools used in this research included Microsoft Excel 2013, CORDEX, ArcMap 10.8, and NASA POWER software. The materials used include historical data on daily rainfall, daily minimum temperature, and daily maximum temperature, as well as potato productivity data from Tosari District and Poncokusumo District. Data analysis carried out in this research consisted of: 1) validation test of the climate projection model used, 2) projection of average rainfall and temperature, 3) multiple linear regression analysis to project potato crop productivity. The research results show that in the RCP 2.6 scenario, there is no pattern of increase in average atmospheric temperature until 2100, while there is a pattern of temperature increase of 0.41-1.21°C and 1.26-2.23°C in the RCP 4.5 scenario and RCP 8.5. There is an increase in potato plant productivity in Tosari District and Poncokusumo District based on productivity projections until 2100. Meanwhile, there is a decrease in potato plant productivity in Poncokusumo District in the RCP 2.6 and RCP 4.5 scenarios. Based on the RCP 8.5 scenario, there is a pattern of higher productivity increase in Poncokusumo District compared to Tosari District.

***Keywords*— Climate Projection, Potatoes, Productivity, RCP**

I. INTRODUCTION

Potatoes (*Solanum tuberosum* L.) are one of the horticultural commodities that serve as an alternative carbohydrate source and are needed by society as one of the essential food ingredients. Along with the increasing population in Indonesia, the demand for potatoes in society continues to increase. In 2022, the demand for potatoes reached 874.25 thousand tons (an increase of about 13.32% compared to the previous year) (Irijayanti et al., 2023). The increasing demand for potatoes in society is also accompanied by an increase in the volume of fresh potato imports from several countries, such as Belgium, the United States, and the Netherlands. The volume of fresh potato imports in Indonesia was 40,493 tons in 2020, 52,286 tons in 2021, and 74,438 tons in 2022. Meanwhile, the volume of fresh potato exports continues to decline. The volume of fresh potato exports in Indonesia was 4,357 tons in 2020, 3,121 tons in 2021, and 2,666 tons in 2022. This indicates that there is a domestic challenge to increase potato crop productivity in order to reduce the volume of potato imports from abroad so that the needs of the people can be met.

East Java Province is the province with the highest contribution to potato production in Indonesia. In 2022, East Java had a production of 385.12 thousand tons, which

is about 25.61% of the total production of potatoes on a national scale (Irijayanti et al., 2023). The potato cultivation center areas in East Java are located in several districts, two of which are Pasuruan District and Malang District. Tosari Sub-district is a production center area in Pasuruan District with a production reaching 1,542,806 quintals in September 2020 (Pasuruan District Central Statistics Agency, 2022). Poncokusumo Sub-district is a production center area in Malang District with a production reaching 252,080 quintals in 2020 (Malang District Central Statistics Agency, 2022).

One of the challenges faced in potato cultivation is global warming. This phenomenon is caused by an increase in greenhouse gas emissions in the atmosphere originating from various human activities. These gases can trap solar heat in the atmosphere, causing the air temperature to rise and affecting climate variations. This has the potential to result in shifting weather patterns and increasing atmospheric temperatures that can affect changes in planting seasons and agricultural production.

Potatoes are sensitive to high temperatures, and temperature stress can significantly reduce potato yields (Singh et al., 2020). Temperature stress can inhibit seedling growth, leaf formation, and quality tuber formation. Changes in rainfall patterns within a year can affect the optimal planting season for potato growth. Thus, appropriate adaptation strategies based on the analysis of possible climate variations in the coming years are needed to maintain and strengthen potato crop productivity.

The process of predicting and identifying climates in the future currently uses climate projection models that have been developed in various countries. Climate projection models are models developed to simulate the atmosphere, ocean, ice, and land to obtain outputs in the form of information on the extent of human activity's influence on the climate (Akbar et al., 2022). This modeling has been utilized and developed to estimate climate events in the future and analyze their causes by simulating historical data. The use of climate projections can be applied in various fields, including agriculture.

The application of climate projections is done by simulating scenarios of greenhouse gas emissions in the atmosphere with projections of radiation on different scales. These scenarios are called Representative Concentration Pathways (RCP). There are four different RCP scenarios, namely RCP 2.6, RCP 4.5, RCP 6.0, and RCP 8.5 (Rao et al., 2020). These four scenarios are projected based on trends and the possibility of tightness or looseness of climate change mitigation measures implemented in a region. RCP 2.6 is a scenario of strict climate change mitigation, RCP 4.5 and 6.0 are moderate scenarios, and RCP 8.5 is a scenario with very high greenhouse gas concentrations (Bienvenido-Huertas et al., 2021).

This research was conducted to project climate variations until 2100 in potato production center areas, namely Tosari and Poncokusumo Sub-districts. Climate projections were carried out using Global Climate Models (GCM) based on three different Representative Consentration Pathways scenarios on different scales, namely RCP 2.6 (low scale), RCP 4.5 (medium scale), and RCP 8.5 (high scale). This prediction was made to analyze the impact of climate variations on three emission scenarios and concentrations of greenhouse gases on potato crop productivity so that it can be considered for the development of potato cultivation strategies in the future.

II. MATERIALS AND METHODS

This study investigates the impact of climate variations on potato crop productivity in two research areas, which are the central potato production areas in East Java, namely Tosari Sub-district, Pasuruan Regency, and Poncokusumo Sub-district, Malang Regency. The coordinates of Tosari Sub-district are 7.889844 South Latitude and 112.924890 East Longitude. The coordinates of Poncokusumo Sub-district are 7.9860258 South Latitude and 112.9110814 East Longitude. Both areas have highland topography. The research was conducted from May to October 2023.

The tools used in this study included Microsoft Excel 2013 software, Coordinated Regional Climate Downscaling Experiment (CORDEX), ArcMAP 10.8, and NASA Prediction Of Worldwide Energy Resources (NASA POWER). The materials used in this study consisted of historical climate data, including daily rainfall, daily minimum temperature, and daily maximum temperature for the last 30 years, as well as potato production data from Tosari Sub-district and Poncokusumo Sub-district from 2018 to 2021.

This research projected climate variations at observation points until 2100 based on a comparison of three different climate projection models (CNRM-CM5, HadGEM2-ES, and MPI-ESM-LR). The study comprised data collection, climate projection, bias correction validation, classification of climate variation trends, and projection of potato productivity using multiple linear regression analysis.

HASIL DAN PEMBAHASAN

Historical Climate Observation Data

Historical climate observation data in both study areas show differences in rainfall and temperature. In the Tosari District, the highest average rainfall peak occurs in

January (691.818 mm/month) and the lowest rainfall is recorded in September (37.232 mm/month). Tosari District experiences wet months from November to May, moist months in June and October, and dry months from July to September. In the Poncokusumo District, the highest rainfall peak is in February (343.583 mm/month) and the lowest is in August (21.361 mm/month). Poncokusumo District experiences wet months from November to March, moist months in April and October, and dry months from May to September. Based on the conversion results of maximum and minimum temperatures, both observation areas have the highest average maximum temperature peak per month in October and the lowest minimum temperature in August. The average maximum temperature throughout the year in Tosari District is 19.219°C and in Poncokusumo District is 18.93°C. Meanwhile, the average minimum temperature throughout the year in Tosari District is 10.91°C and in Poncokusumo District is 10.36°C.

In the Tosari District, the average annual rainfall is 4115.63 mm/year. Meanwhile, in the Poncokusumo District, the average annual rainfall is 2024.15 mm/year. Based on the average rainfall in both locations, the Tosari District has higher rainfall compared to the Poncokusumo District. The primary factor influencing rainfall in an area is the sea surface temperature anomalies in the equatorial Pacific Ocean caused by the ENSO (El Nino Southern Oscillation) phenomenon (Somadayo *et al*., 2022). This phenomenon is a global interaction between the ocean and atmosphere consisting of two main phases, El Niño and La Niña. During the El Niño phase, there is a rise in sea surface temperatures accompanied by decreased rainfall and increased air temperatures in Indonesia. During the La Niña phase, there is a decrease in sea surface temperatures leading to increased rainfall and decreased air temperatures in Indonesia. This affects various climate factors such as wind patterns, atmospheric pressure, humidity, temperature, and rainfall distribution.

Both study areas have characteristics of monsoon rainfall. This is characterized by distinct differences between the rainy season and dry season periods, and is characterized by only one peak in rainfall within a year. In the Tosari District, the rainy season occurs from November to May and the dry season from June to October. In the Poncokusumo District, the rainy season occurs from November to April and the dry season from May to October. The characteristics of monsoon rainfall are influenced by monsoon winds driven by high-pressure cells and low-pressure cells in Asia and Australia that alternate (Tukidi, 2010).

Based on data observation analysis, the differences in rainfall are also influenced by topographic factors. Both study areas are mountainous regions, leading to the presence of orographic effects that affect rainfall patterns. Mountain ranges or hills can act as physical barriers affecting wind movement. Topographic barriers consist of two different sides: the windward side and the leeward side. On the windward side, moist air coming from the sea or lowlands is pushed upwards due to the presence of mountains. The rising air cools and undergoes condensation processes. On the leeward side, air that has lost moisture descends to lower elevations. Therefore, rainfall on the windward side is high, while on the leeward side it is low. The leeward side is the rain shadow side because it has drier conditions.

The Tosari District experiences the highest rainfall in January with 691.818 mm/month, while the Poncokusumo District experiences the highest rainfall in February with 343.583 mm/month. This indicates that the maximum rainfall in the Tosari District is more than twice as high as in the Poncokusumo District. This is because the Tosari District is a mountainous area closer to the monsoon winds on the northern coast of Java Island, making it the windward side. Meanwhile, the Poncokusumo District is on the leeward side, resulting in lower rainfall. Although the Poncokusumo District is closer to the southern coast, based on rainfall observation data, it can be concluded that stronger winds are present on the coast closer to the Tosari District.

Climate Model Validation Test

Based on the validation test results of the three climate models, climate determination is conducted based on the lowest RMSE values for each observation variable. The selection of the best climate GCM model for each variable can be seen in Tables 1 and 2. RMSE values approaching zero (0) indicate high data closeness to historical data.

Table 1. Selection of GCM for Climate Projections in Tosari District

Variabel	Scenario	Selected GCM	RMSE
Rainfall	RCP 2.6	MPI-ESM-LR	23,54
	RCP 4.5	HadGEM2-ES	4,12
	RCP 8.5	HadGEM2-ES	11,25
Maximum Temperature	RCP 2.6	MPI-ESM-LR	6,16
	RCP 4.5	MPI-ESM-LR	1,86
	RCP 8.5	MPI-ESM-LR	0,54
Minimum Temperature	RCP 2.6	MPI-ESM-LR	32,28
	RCP 4.5	MPI-ESM-LR	33,96
	RCP 8.5	MPI-ESM-LR	34,83

Description:

RMSE : *Root Mean Square Error*

Table 2. Selection of GCM for Climate Projections in Poncokusumo District

Variabel	Scenario	Selected GCM	RMSE
Rainfall	RCP 2.6	HadGEM2-ES	27,85
	RCP 4.5	CNRM-CM5	23,27
	RCP 8.5	CNRM-CM5	36,41
Maximum Temperature	RCP 2.6	HadGEM2-ES	47,54
	RCP 4.5	MPI-ESM-LR	46,41
	RCP 8.5	MPI-ESM-LR	44,11
Minimum Temperature	RCP 2.6	MPI-ESM-LR	8,94
	RCP 4.5	MPI-ESM-LR	10,50
	RCP 8.5	HadGEM2-ES	10,49

Description:

RMSE : *Root Mean Square Error*

Rainfall and Temperature Projections

Rainfall and temperature projections are carried out using Global Climate Models to compare climate variations in three different greenhouse gas concentration scenarios, namely RCP 2.6 (low level), RCP 4.5 (medium level), and RCP 8.5 (high level). Projections are made by multiplying data from climate models by monthly correction factors for each study area. Figures 1-6 depict rainfall projections in both study areas for the periods 2021-2040, 2041-2060, 2061-2080, and 2081-2100.

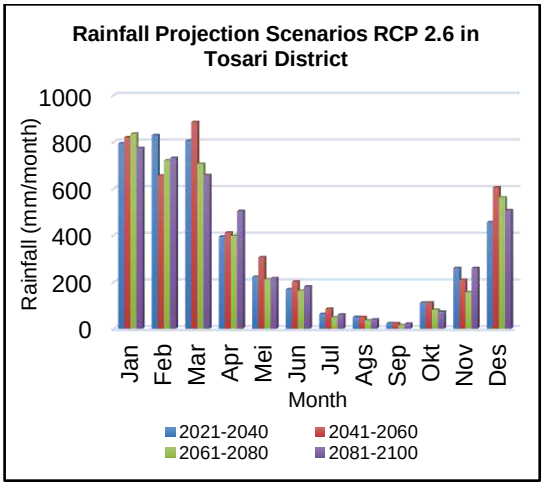

Fig.1. Rainfall Projection Scenario RCP 2.6 in Tosari District

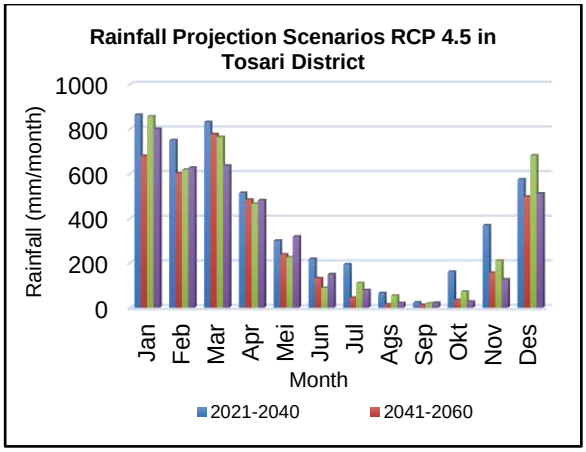

Fig.2. Rainfall Projection Scenario RCP 4.5 in Tosari District

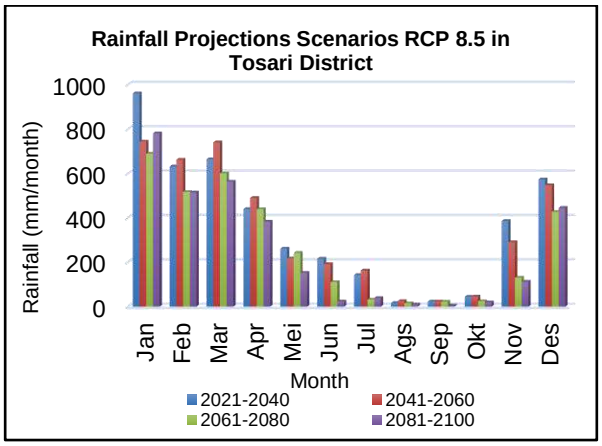

Fig.3. Rainfall Projection Scenario RCP 8.5 in Tosari District

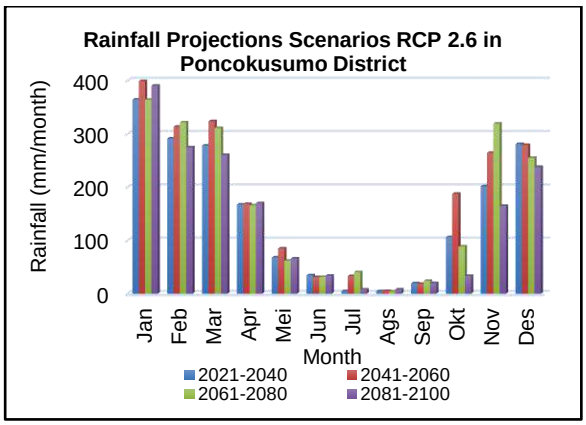

Fig.4. Rainfall Projection Scenario RCP 2.6 in Poncokusumo District

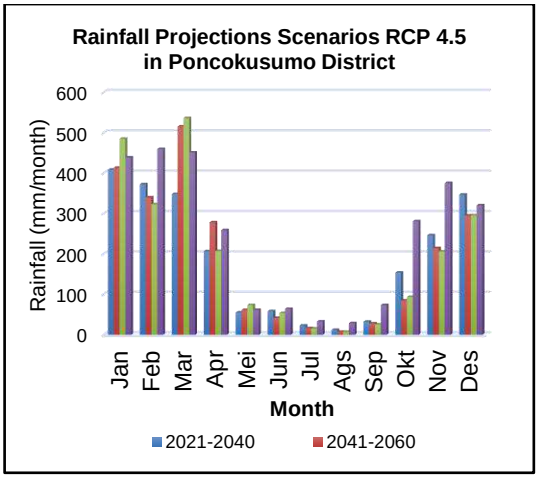

Fig.5. Rainfall Projections Scenarios RCP 4.5 in Poncokusumo District

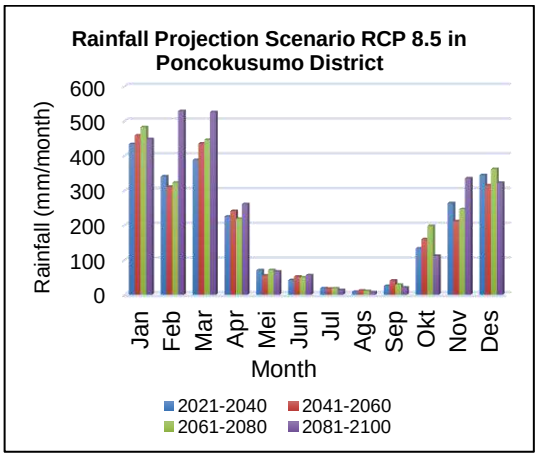

Fig.6. Rainfall Projection Scenario RCP 8.5 in Poncokusumo District

Temperature projections are also conducted by multiplying the maximum and minimum temperature data for each climate model by monthly correction factors for each study area. Figures 7 and 8 show the projection of maximum temperature in the study areas based on three climate projection scenarios, while Figures 9 and 10 show the projection of minimum temperature based on three climate projection scenarios.

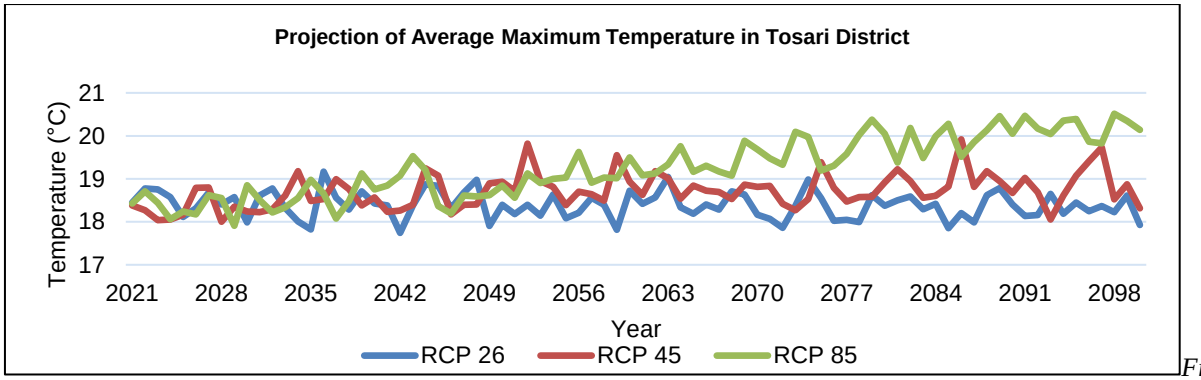

Fig.7. Projection of Monthly Average Maximum Temperature in Tosari District

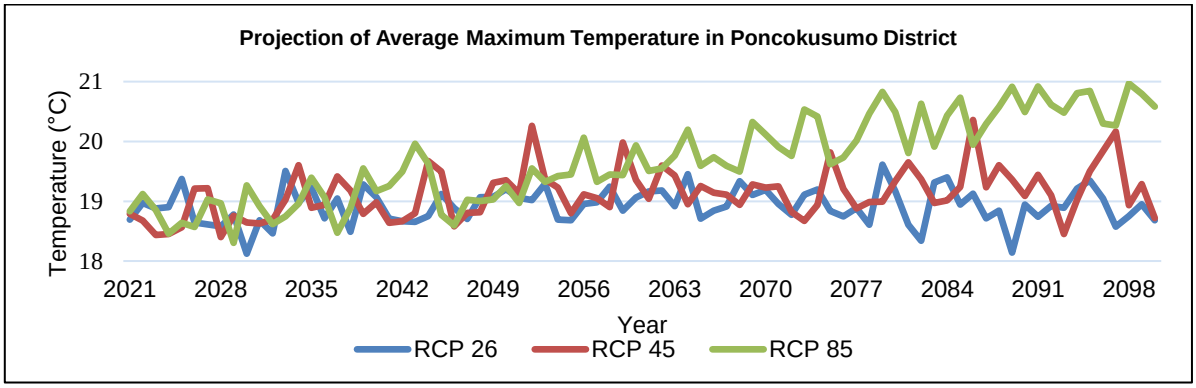

Fig.8. Projection of Monthly Average Maximum Temperature in Poncokusumo District

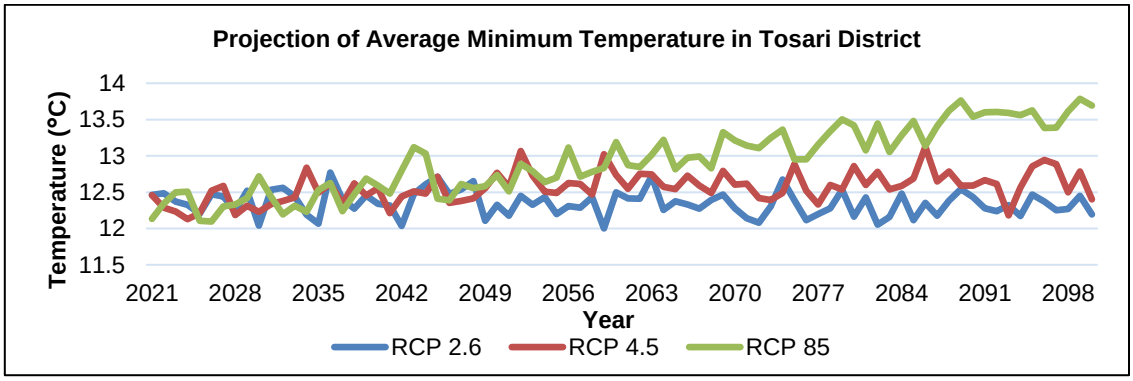

Fig.9. Projection of Monthly Average Minimum Temperature in Tosari District

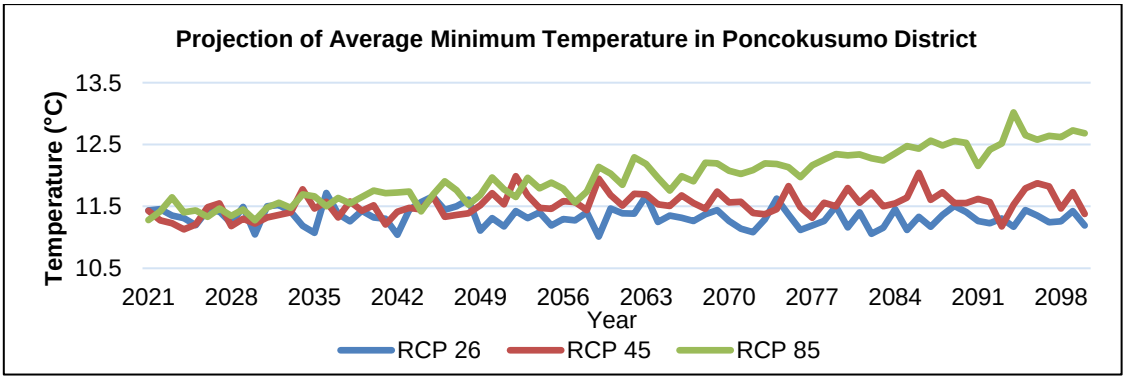

Fig.10. Projection of Monthly Average Minimum Temperature in Poncokusumo District

Based on the projections, greenhouse gas emissions and concentrations in the atmosphere affect climate variability based on rainfall and temperature variables in the study areas. Changes become more significant in scenarios with higher emissions and greenhouse gas concentrations, as indicated by the comparison of projections based on RCP scenarios. In the RCP 8.5 scenario, changes in the rainy and summer seasons in both areas occur earlier compared to the RCP 2.6 and RCP 4.5 scenarios. This is shown by changes in the transition between wet and dry months. With higher emissions and greenhouse gas concentrations scenarios, climate variability projections are also accompanied by more significant increases in both maximum and minimum temperatures. In the RCP 8.5 scenario, temperature increases in both study areas are higher compared to the RCP 2.6 and RCP 4.5 scenarios. The annual average temperature increase in the RCP 8.5 scenario until 2100 can reach 1.26-2.23°C. Meanwhile, in the RCP 4.5 scenario, it can only reach 0.41-1.21°C, and in the RCP 2.6 scenario, there are no significant temperature variations. The months with the highest maximum and minimum temperatures in a year experience temperature increases of 1.16-2.67°C in the RCP 8.5 scenario, while in the RCP 4.5 scenario, they experience increases of 0.29-1.44°C.

The three climate variability scenarios used to project climate in this study depict differences in emission assumptions and the stringency of mitigation measures against greenhouse gas emissions. Greenhouse gases included in these projection models include carbon dioxide (CO2), methane (CH4), nitrous oxide (N2O), sulfur dioxide (SOx), carbon monoxide (CO), non-methane volatile organic compounds (NMVOCs), nitrogen oxides (NOx), black carbon (BC), organic carbon (OC), and ammonia (NH3) (Meinshausen et al., 2011). The RCP 2.6 scenario is a very stringent mitigation scenario, assuming constant emissions and CO2 concentrations returning to 360 ppm by 2300. Therefore, climate variability in the RCP 2.6 scenario represents a scenario projecting stable climate variability over the long term. The RCP 4.5 scenario is a moderate mitigation scenario, reaching its peak in 2040 and then declining, stabilizing CO2 at around 540 ppm by 2100. In the RCP 8.5 scenario, there are no significant efforts to mitigate greenhouse gases, so emissions and greenhouse gas concentrations are assumed to increase throughout the 21st century. This scenario assumes a CO2 concentration of 1370 ppm by 2100. Thus, each RCP scenario has a different rate of emissions increase over the period up to 2100.

The level of greenhouse gas emissions affects the radiative forcing value, which represents the radiative energy entering and leaving the Earth's atmosphere. A positive radiative forcing value indicates that more energy enters the atmosphere than leaves it due to high greenhouse gas emissions. In the RCP 8.5 scenario, the atmosphere has an estimated radiative forcing value of 8.5 W/m² by 2100, and this radiative forcing value is the highest compared to the RCP 2.6 and RCP 4.5 scenarios (van Vuuren *et al.*, 2011). This affects the impacts on climate variability in the study areas. Radiation leads to an increase in average temperatures both in the ocean and on land, and affects rainfall patterns in the study areas. In the temperature projection results, temperature increases in the RCP 8.5 scenario are higher compared to the other scenarios.

Potato Crop Productivity Projection Results

Multiple linear regression analysis was conducted to determine the relationship between potato crop productivity and two climate variables in both study areas, namely rainfall and temperature. Potato crop productivity in the Tosari District has a regression equation Y = 5.58947 - 0.00395 x1 + 3.13113 x2, and potato crop productivity in the Poncokusumo District has a regression equation Y = -105.235 - 0.00253x1 + 8.66131x2 (Y is the potato crop productivity in the Poncokusumo District, x1 is the average annual rainfall value, and x2 is the average annual temperature in the study areas). These equations are used to determine the projected values of potato crop productivity until 2100 (Figures 11 and 12; Tables 3 and 4).

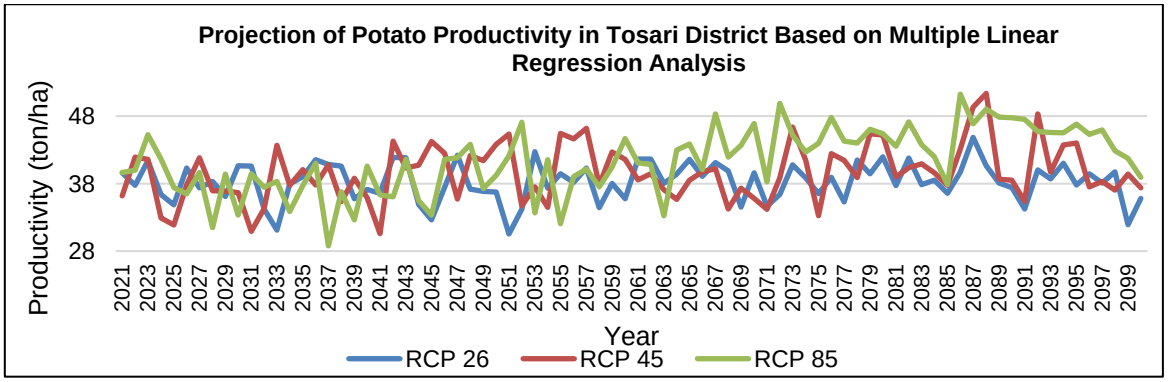

Fig.11. Projection of Potato Crop Productivity in Tosari District Based on Multiple Linear Regression Analysis

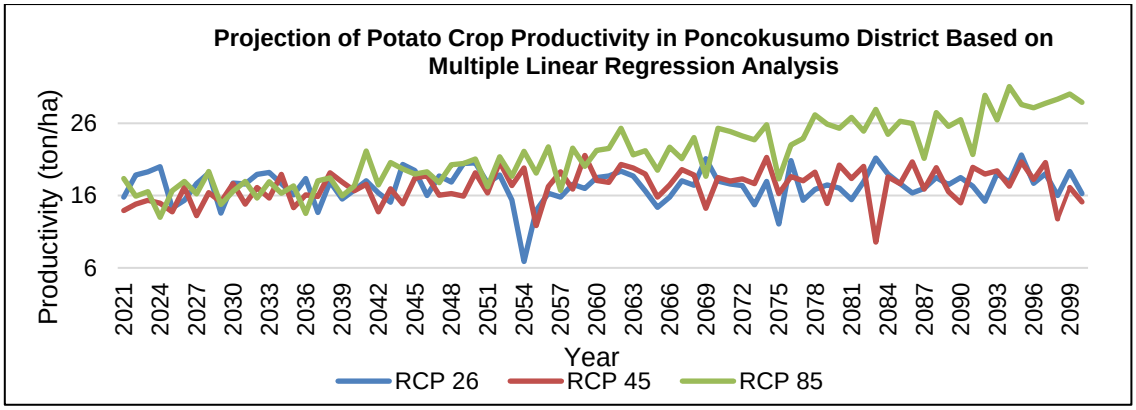

Fig.12. Projection of Potato Crop Productivity in Poncokusumo District Based on Multiple Linear Regression Analysis

Table 3. Projection of Average Potato Crop Productivity in Tosari District

Year	RCP 2.6		RCP 4.5		RCP 8.5	
	Projection (ton/ha)	Increase/ Decrease Based on Baseline	Projection (ton/ha)	Increase/ Decrease Based on Baseline	Projection (ton/ha)	Increase/ Decrease Based on Baseline
2021-2040	38,072	10,00%	37,491	8,32%	37,550	8,49%
2041-2060	37,455	8.22%	40,803	17,90%	39,239	13,37%
2061-2080	39,041	12,80%	39,198	13,26%	43,517	25,74%
2081-2100	38,490	11,21%	40,968	18,37%	45,144	30,44%

Table 4. Projection of Average Potato Crop Productivity in Poncokusumo District

Year	RCP 2.6		RCP 4.5		RCP 8.5	
	Projection (ton/ha)	Increase/ Decrease Based on Baseline	Projection (ton/ha)	Kenaikan/ Penurunan Berdasarkan *Baseline*	Projection (ton/ha)	Increase/ Decrease Based on Baseline
2021-2040	17,165	-3,03,%	15,945	-9,92%	16,698	-5,67%
2041-2060	17,039	-3,74%	17,343	-2,02%	20,027	13,14%
2061-2080	17,276	-2,40%	18,198	2,81%	23,266	31,44%
2081-2100	17,916	1,21%	17,664	-0,21%	26,997	52,52%

Projection of average annual potato crop productivity based on multiple linear regression analysis shows the different climate variability impacts in the two study areas. Tosari District projects an increase in average annual potato crop productivity in scenarios RCP 2.6, RCP 4.5, and RCP 8.5. Scenario RCP 8.5 experiences a significant increase pattern with an average increase of 6.91% every 20 years until 2100. Meanwhile, the average annual potato crop productivity in Poncokusumo District experiences a long-term decrease in average potato crop productivity in scenarios RCP 2.6 and RCP 4.5. Productivity projections based on scenario RCP 8.5 show a significant increase pattern with an average increase of 17.38% every 20 years until 2100. This indicates that: 1) there is an influence of increasing greenhouse gas concentrations and emissions on potato crop productivity in a region, 2) geographic location plays a role in supporting future potato crop productivity based on climate factors, particularly regarding temperature and rainfall projections.

Based on the research findings, the increase in average annual temperature due to increasing greenhouse gas emissions is directly proportional to the potato crop productivity projections in both regions. This is indicated by the average annual potato crop productivity showing an increasing trend in the RCP 8.5 scenario, while fluctuations occur in the RCP 2.6 and RCP 4.5 scenarios. This phenomenon is consistent with the research conducted by Jennings et al. (2020), which suggests a potential increase in potato productivity globally due to global warming. This is because the increase in atmospheric CO2 can enhance carbon dioxide fertilization in potato plants, potentially increasing productivity by 22-33%. Atmospheric carbon dioxide is an essential component for plants in carrying out the process of photosynthesis. The accumulated carbohydrates produced from photosynthesis are used for plant growth and tuber formation, thus potentially increasing yields.

Geographic location is also one of the factors influencing potato crop productivity projections. Based on the previous observational data analysis, Tosari District is the windward side facing the sea experiencing monsoon winds, while Poncokusumo District is the leeward side opposite the direction of the wind. With the RCP 8.5 scenario, the average annual potato crop productivity on the leeward side is projected to experience a higher percentage increase in productivity compared to the windward side until 2100. The leeward side is a relatively drier area compared to the windward side, and the high greenhouse gas emissions scenario has the potential to increase rainfall in that area and can increase soil moisture (Kagawa-Viviani et al., 2018). This is consistent with climate projection results showing that Poncokusumo District has increased rainfall until 2100 in the RCP 8.5 scenario. Meanwhile, climate projections show that Tosari District, which has wetter conditions, experiences decreased rainfall until 2100 in the RCP 8.5 scenario. However, overall potato crop productivity in Tosari District remains higher in all three climate projection scenarios. This is because Tosari District has higher observed and projected rainfall compared to Poncokusumo District. Tosari District has rainfall of 4115.65 mm/year based on observational data, and projections show a decrease in rainfall to 3049.08 mm/year in the RCP 8.5 scenario. Meanwhile, Poncokusumo District has rainfall of 2024.15 mm/year based on observational data, and projections show an increase in rainfall to 2847.32 mm/year.

When viewed based on atmospheric temperature, the projected atmospheric temperatures in both areas in the RCP 8.5 scenario are suitable for the cardinal temperature of potato plants. According to Singh et al. (2013), the optimum cardinal temperature for potato plants ranges from 16-25°C. With the suitability of the optimum cardinal temperature, the processes of photosynthesis and transpiration in potato plants can proceed smoothly, resulting in optimal plant development. If the temperature is too high, it can delay tuber initiation and affect potato crop productivity and tuber quality. These conditions have the potential to damage potato plants.

The impact of climate variability on potato growth and productivity is complex, so increasing atmospheric greenhouse gas emissions can affect productivity both positively and negatively. The impact depends on factors related to supporting components of potato growth, such as atmospheric temperature and rainfall. Temperature plays a crucial role in supporting each phase of potato plant growth. High atmospheric temperatures accelerate evapotranspiration, causing water stress for plants due to water loss. This condition can affect potato plant growth in several phases, such as tuber formation and maturation. If the tuber formation and maturation processes are not optimal, it can lead to yield reduction. Additionally, global warming affects rainfall distribution in a region and triggers increased extreme weather. Extreme dry weather can cause soil moisture to be insufficient for potato plants to perform photosynthesis and form good tubers. Meanwhile, excessive rainfall can also cause soil erosion, leading to a lack of soil fertility for potato growth and development. To adapt to extreme weather changes, adaptive measures can be taken by planting potatoes earlier to avoid extreme dry weather disasters (Zhao and Li, 2015). This step has the potential to increase yields.

The response of potato crop productivity to increased greenhouse gas emissions can vary. Farming practices that can adapt to climate change and the implementation of policies tailored to regional conditions are essential. Based on the research conducted, greenhouse gas emissions affect rainfall distribution in a region, leading to different projections in different areas. Therefore, appropriate water management measures are needed to maintain optimal potato crop productivity. Water management for potato cultivation can be carried out by regulating proper irrigation, efficient water usage, and good rainwater management (Pratama et al., 2021). Irrigation and drainage systems, in adhering to water management principles, can utilize suitable technologies. Efficient irrigation technologies such as drip irrigation or sprinklers with proper irrigation scheduling can be employed. Drainage management can also be achieved by regulating drainage channels to function effectively in preventing potato plants from being waterlogged.

III. CONCLUSION

In the RCP 2.6 scenario, there is no pattern of average atmospheric temperature increase until 2100. In scenarios RCP 4.5 and RCP 8.5, there is a temperature increase pattern of 0.41-1.21°C and 1.26-2.23°C, respectively. The temperature increase in Tosari District is potentially higher than in Poncokusumo District. Based on productivity projections, there is an increase in potato crop productivity in both Tosari and Poncokusumo Districts based on productivity projections until 2100. Meanwhile, there is a decrease in potato crop productivity in Poncokusumo District in scenarios RCP 2.6 and RCP 4.5. According to the RCP 8.5 scenario, there is a higher productivity increase pattern in Poncokusumo District compared to Tosari District.

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To evaluate trends in weather variables in Haryana using Mann Kendall test and Sen's slope estimator

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Abstract**—***Over the 35-year period from 1985 to 2020, an extensive analysis of rainfall and temperature trends across several cities in Haryana, including Hisar, Rewari, Ambala, Karnal, and Rohtak, was conducted. Hisar exhibited a significant yearly increase in May rainfall, while the monsoon season showed a non-significant decreasing trend. Rewari experienced notable rises in March rainfall but insignificant changes during monsoon, while Ambala saw an overall decrease in rainfall, particularly in September. Karnal indicated significant January rainfall increases but insignificant changes during monsoon. Rohtak displayed noteworthy reductions in February and August rainfall but insignificant changes during monsoon. Temperature trends varied across cities, with consistent increases observed in minimum temperatures and sporadic changes in maximum temperatures. Understanding these patterns is crucial for evaluating regional climate variations and devising appropriate adaptation strategies. This comprehensive research provides valuable insights into the complex interplay between rainfall and temperature dynamics, offering essential information for policymakers and stakeholders tasked with addressing climate change challenges in the Haryana region.*

Keywords**—*Climate change, Haryana, Monsoon, Rainfall, Temperature***

I. INTRODUCTION

Land constitutes a valuable asset encompassing natural elements such as terrain, weather, soil, water, and plants. Humanity relies on this resource for sustenance, textiles, and energy, leading to a swift expansion of farming areas. The growth of agriculture paved the way for the surge in industrialization, akin to how the agricultural revolution preceded the industrial revolution spanning from 18th-century England to 19th-century Japan (World Development Report 2008). The adoption of contemporary farming techniques in the 1970s, notably recognized as the Green Revolution, in Haryana state brought about remarkable advancement over the past 40-50 years. This progress stands as an unparalleled instance in the annals of global agricultural development. (Rangi and Sidhu 1998).

Hussain *et al.,* (2021) examined monthly minimum temperature (T_{min}), maximum temperature (T_{max}), and precipitation from nine Upper Indus Basin (UIB) meteorological stations to study the spatiotemporal variations of temperature and precipitation on month to month, seasonal, and yearly scales. Mondal *et al.,* (2015) studied alterations in the trends of rainfall and temperature for 141 (1871-2011) and 107 years (1901-2007) respectively in India. Pal *et al.,* (2011) documented the protracted trends for the four yearly, seasonal and precipitation types in India's main climatological areas and sub-regions.

Despite the remarkable advancements in agricultural technology over the past few years, the weather and climate still play a significant role in agricultural production. The net primary productivity of plants will raise as CO_2 concentrations rise, but climate change and the concomitant changes in disturbance regimes could result in either an increase or a decrease in net ecosystem production. The hydrologic cycle and groundwater recharge, which regulate waterlogging and salinity

distribution of shallow aquifer systems and consequently impact the amount of land that can be used for agriculture, can be considerably altered by changes in climatic variables. A number of significant weather abnormalities that occur in the State have a negative impact on crop production, including insufficient rains, excessive rains that fall at the wrong time, heat waves, cold waves, high and hot winds during the summer (known locally as "loo"), dust storms, fog, frost, and hail. The south-westerly monsoon and north-westerly cold winds have a significant impact on Haryana's climate. From July to September, only the tails of the summer monsoon depressions are experienced.

II. MATERIAL AND METHODS

For evaluation of climate change, 5 districts are selected in such a way that whole Haryana is represented. These 5 districts are- Ambala, Rohtak, Hisar, Karnal and Rewari. The geographical area is 1569 km^2 of Ambala, 1745 km^2 of Rohtak, 3083 km^2 of Hisar, 2520 km^2 of Karnal and 1594 km^2 of Rewari.

To analyze the trend in weather variables, three parameters have been taken i.e., Maximum temperature, Minimum temperature and Rainfall for five districts (Ambala, Rohtak, Hisar, Karnal and Rewari). To evaluate climate change at all districts, climatic data of 35 years (1985-2020) has been taken of their respective district. Data was taken from Department of Agricultural Meteorology, CCSHAU, Hisar and IMD.

Trends in long term weather data will be carried out by the widely accepted non-parametric test for weather variables working with time series trends i.e. Mann Kendall test and Sen's slope estimator using MAKESENS programme. Where Mann- Kendall Test shows the trend analysis and the value of Sen's slope estimator gives the magnitude of the trend.

III. RESULTS AND DISCUSSION

Five districts data has been tabulated and precessed through the MAKESENS programme and the results have been discussed below:

3.1 Weather variables at Hisar

During the period of 1985-2020 at Hisar, May month observed significant increasing trend in rainfall @ 0.23 mm/year. Whereas during monsoon, there is a non-significant decreasing trend with rainfall decreasing @ 0.04mm/year. Yearly trend at Hisar observed increasing trend in rainfall @ 1.31 mm/year.

The results revealed that during the last 35 years (1985-2020), the annual mean maximum temperature of Hisar had shown a non-significant increasing trend by 0.01°C/year, while in January, the mean maximum temperature had shown a significant decreasing trend and decreased by 0.04°C (Table 1). Meanwhile, in September, the mean maximum temperature had shown a significant increasing trend and increased by 0.03°C.

The average minimum temperature exhibited a noteworthy upward trend throughout each month. On an annual basis, there was a significant increase in the minimum temperature, reaching 0.02°C per year. Similarly, February, April, and August experienced notable rising trends, with temperature increments of 0.03°C, 0.04°C, and 0.02°C, respectively, at Hisar during the period of 1985-2020.

3.2 Weather variables at Rewari

The findings indicate that over the past 35 years (1985-2020) in Rewari, there has been a notable rise in rainfall during the month of March, with an increase of 0.17 mm per year. Conversely, during the monsoon season, there is a slight, albeit statistically insignificant, upward trend in rainfall, which amounts to an increase of 2.09 mm per year. Overall, the annual trend in rainfall at Rewari demonstrates a significant increase of 2.65 mm per year.

During the period of 35 years (1985-2020), the average maximum temperature in Rewari showed no change at all. However, in January, there was a notable and significant decrease in the mean maximum temperature, with a decrease of 0.05°C (Table 2). Similarly, in November, there was also a significant decrease observed in the mean maximum temperature, with a decrease of 0.03°C.

The mean minimum temperature in Rewari displayed a considerable upward trend across all months. Annually, there was a significant rise in the minimum temperature, amounting to 0.02°C per year. Similarly, February, April, August, and September witnessed noticeable increasing trends, with temperature increments of 0.03°C, 0.04°C, 0.02°C, and 0.02°C, respectively.

3.3 Weather variables at Ambala

The results suggest that in Ambala over the past 35 years (1985-2020), there was a considerable increase in rainfall during April, averaging a rise of 0.36 mm per year. However, in September, there was a significant decrease noted, amounting to 2.91 mm per year. Conversely, during the monsoon season, there was a slight, yet statistically significant, decline in rainfall, equating to a decrease of 10.94 mm per year. Overall, the annual trend in rainfall for Ambala indicated a significant decrease of 10.52 mm per year.

During the last 35 years (1985-2020), the mean maximum temperature in Ambala showed a non-significant annual increase of 0.01°C. However, July stood out with a significant and noticeable rise in the mean maximum temperature, amounting to an increase of 0.03°C (Table 3). Similarly, in August, September, and October, there was also a significant increase observed in the mean maximum temperature, each with an increase of 0.03°C.

The average minimum temperature in Ambala exhibited a considerable upward trend across all months. Annually, there was a significant rise in the minimum temperature, amounting to 0.02°C per year. Similarly, February, April, July, August, September, October, and November experienced noticeable increasing trends, with temperature increments of 0.03°C, 0.03°C, 0.02°C, 0.02°C, 0.02°C, 0.03°C, and 0.02°C, respectively.

3.4 Weather variables at Karnal

The findings indicate that in Karnal during the last 35 years (1985-2020), there has been a significant rise in rainfall during January, averaging an increase of 0.31 mm per year. However, during the monsoon season, there is a slight but statistically insignificant decrease in rainfall, amounting to a decline of 0.97 mm per year. Overall, the annual trend in rainfall for Karnal suggests an insignificant decrease of 0.61 mm per year.

Over the past 35 years (1985-2020), the average maximum temperature in Karnal exhibited a non-significant annual rise of 0.01°C. However, October notably demonstrates a significant and conspicuous increase in the mean maximum temperature, with a rise of 0.03°C (Table 4).

The average minimum temperature in Karnal exhibited a notable upward trend throughout each month. On an annual basis, there was a significant increase in the minimum temperature, reaching 0.02°C per year. Similarly, February, April, July, August, and September experienced observable rising trends, with temperature increments of 0.03°C, 0.04°C, 0.02°C, 0.03°C, and 0.02°C, respectively.

3.5 Weather variables at Rohtak

The results suggest that over the past 35 years (1985-2020) in Rohtak, there has been a noteworthy reduction in rainfall during February and August, averaging decreases of 0.15 mm and 2.66 mm per year, respectively. However, during the monsoon season, there is a slight but statistically insignificant decrease in rainfall, with a decline of 4.71 mm per year. Overall, the annual trend in rainfall for Rohtak indicates an insignificant decrease of 5.58 mm per year.

Mean maximum temperature in Rohtak showed no change in the trend annually during the last 35 years (1985-2020). However, January and November stood out with significant and noticeable decreases in the mean maximum temperature, with declines of 0.05°C and 0.03°C, respectively (Table 5).

The mean minimum temperature in Rohtak exhibited a noticeable upward trend across all months. Annually, there was a significant rise in the minimum temperature, reaching 0.02°C per year. Similarly, February, April, August, and September experienced observable significant increases, with temperature increments of 0.03°C, 0.04°C, 0.02°C, and 0.02°C, respectively.

IV. FIGURES AND TABLES

Table 1: Monthly Significant trends in weather variables at Hisar

Weather Parameters	Months	Mann-Kendall Trend (Test Z)	Sen's slope estimation (Q)
Rainfall			
	MAY	1.98*	0.23
	S-W MONSOON	-0.09	-0.04
	ANNUAL	0.67	1.31
Maximum Temperature			

	JANUARY	-1.70 +	-0.04
	SEPTEMBER	1.68 +	0.03
	ANNUAL	0.69	0.01
Minimum Temperature			
	FEBRUARY	1.81 +	0.03
	APRIL	1.76 +	0.04
	AUGUST	2.77 **	0.02
	ANNUAL	2.38 *	0.02

Table 2: Monthly Significant trends in weather variables at Rewari

Weather Parameters	Months	Mann-Kendall Trend (Test Z)	Sen's slope estimation (Q)
Rainfall			
	MARCH	2.05 *	0.17
	S-W MONSOON	0.59	2.09
	ANNUAL	0.69	2.65
Maximum Temperature			
	JANUARY	-1.92 +	-0.05
	NOVEMBER	-1.87 +	-0.03
	ANNUAL	-0.40	0.00
Minimum Temperature			
	FEBRUARY	1.81 +	0.03
	APRIL	1.76 +	0.04
	AUGUST	1.88 +	0.02
	SEPTEMBER	2.04 *	0.02
	ANNUAL	2.08 *	0.02

Table 3: Monthly Significant trends in weather variables at Ambala

Weather Parameters	Months	Mann-Kendall Trend (Test Z)	Sen's slope estimation (Q)
Rainfall			
	APRIL	2.52 *	0.36
	SEPTEMBER	-1.87 +	-2.91
	S-W MONSOON	-2.49 **	-10.94
	ANNUAL	-2.74 *	-10.52
Maximum Temperature			
	JULY	1.76 +	0.03

	AUGUST	2.11 *	0.03
	SEPTEMBER	2.03 *	0.03
	OCTOBER	1.70 +	0.03
	ANNUAL	0.94	0.01
Minimum Temperature			
	FBRUARY	1.68 +	0.03
	APRIL	1.68 +	0.03
	JULY	1.89 +	0.02
	AUGUST	3.17 **	0.02
	SEPTEMBER	2.03 *	0.02
	OCTOBER	2.44 *	0.03
	NOVEMBER	1.73 +	0.02
	ANNUAL	2.68 *	0.02

Table 4: Monthly Significant trends in weather variables at Karnal

Weather Parameters	Months	Mann-Kendall Trend (Test Z)	Sen's slope estimation (Q)
Rainfall			
	JANUARY	1.90 +	0.31
	S-W MONSOON	-0.29	-0.97
	ANNUAL	-0.26	-0.61
Maximum Temperature			
	OCTOBER	2.06 *	0.03
	ANNUAL	1.13	0.01
Minimum Temperature			
	FEBRUARY	1.81 +	0.03
	APRI	1.89 +	0.04
	JULY	1.70 +	0.02
	AUGUST	3.42 ***	0.03
	SEPTEMBER	2.36 *	0.02
	ANNUAL	2.74 **	0.02

Table 5: Monthly Significant trends in weather variables at Rohtak

Weather Parameters	Months	Mann-Kendall Trend (Test Z)	Sen's slope estimation (Q)
Rainfall			
	FEBRUARY	-1.74 +	-0.15
	AUGUST	-2.06 *	-2.66

	S-W MONSOON	-1.51	-4.71
	ANNUAL	-1.46	-5.58
Maximum Temperature			
	JANUARY	-1.92 +	-0.05
	NOVEMBER	-1.87 +	-0.03
	ANNUAL	-0.40	0.00
Minimum Temperature			
	FEBRUARY	1.81 +	0.03
	APRIL	1.76 +	0.04
	AUGUST	2.47 *	0.02
	SEPTEMBER	2.19 *	0.02
	ANNUAL	2.08 *	0.02

V. CONCLUSION

Over the 35-year period from 1985 to 2020, analysis of weather patterns in various cities of Haryana reveals significant trends. Hisar experienced increasing rainfall in May and yearly, but a non-significant decrease during monsoon. Rewari saw notable rises in March and overall rainfall, with stable maximum temperatures but notable increases in minimum temperatures. Ambala witnessed mixed rainfall trends, notably decreasing in September. Karnal showed rising rainfall in January but a slight decrease during monsoon, with stable maximum temperatures and significant increases in minimum temperatures. Rohtak displayed reduced rainfall in February and August, stable maximum temperatures, and significant rises in minimum temperatures throughout the year. These trends reflect complex climate dynamics requiring continued monitoring and adaptation efforts.

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Impact of added Phosphorus and Phosphorus Solubilizing Bacteria in Yield and Yield Attributes of Mungbean *(Vigna Radiata L.)*

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Abstract— *Mungbean is an excellent rotational legume crop for farmers with small land holdings. Phosphorus Solubilizing Bacteria (PSB) solubilizes phosphorus to make it available to the plant so, their integration can be a supporting factor to increase its yield and enhance soil fertility. A field experiment was conducted at the Agronomy Research Farm of the Institute of Agriculture and Animal Sciences (IAAS), Paklihawa Campus, Rupandehi during the summer season of 2023. The experiment comprised seven treatments, namely T1: Control, T2: 10 kg P2O5ha-1 , T3: 15 kg P2O5ha-1 , T4: 20 kg P2O5ha-1 , T5: 10 kg P2O5ha-1 + PSB, T6: 15 kg P2O5ha-1 + PSB, T7: 20 kg P2O5ha-1 + PSB tested in Randomized Block Design and replicated three times. Pratigya variety of Mungbean was used. The soil of the experimental field was medium in organic matter, nitrogen, phosphorus, and potassium before the experiment. The analysis was done in R studio software. Results revealed that all the growth and yield attributes increased significantly ($p < .05$) under the integrated treatment (20 kg P2O5ha-1 + PSB). The growth characters viz., plant height (61.65 ± 1.25 cm), nodule number (23.90 ± 2.22), dry matter accumulation(30.74± 1.37 g), and yield attributes like the number of pod plant-1 (26.6± 1.00), pod length (8.24 cm ± 0.05), the number of grains pod-1 (8.84 ± 0.08), biological yield (30.2 ± 0.79 mt/ha), seed yield (2.44 ± 0.05 mt/ha) and harvest index (7.51 ± 0.18 %) increased significantly in T7 (20 kg P2O5 ha-1 PSB).*

Keywords— ***Mungbean, Phosphorus, Phosphorus Solubilizing Bacteria (PSB), Nodules, Yield***

I. INTRODUCTION

Mung bean (Vigna radiata), popularly known as green gram belongs to the family Fabaceae and has been consumed as a common traditional food for more than 3500 years across the world. Kole, C. (2007). Mung bean is an excellent rotation crop for smallholder farmers because of its short crop duration, tolerance to heat, minimal input requirement, and strong global demand. It provides its users with a triple benefit: more money, more nutrient-rich food, and more fertile soil (Nair et al., 2020). The crop responds favorably to the application of fertilizer phosphorus. It is the second most crucial macronutrient needed by plants after nitrogen. The characteristics of phosphorus nutrition include root development, stalk, and stem strength, flower and seed formation, crop maturity and production, N-fixation in legumes, crop quality, and resistance to plant diseases. Although microbial inoculants have been used to increase soil fertility over the past century, P solubilization research has been reported on much less frequently than nitrogen fixation (Jilani et al., n.d.) By providing assimilates to the roots, phosphorus not only plays a crucial function in root growth and proliferation but also enhances nodulation and N fixation (Kumar & Yadav, 2018). By increasing the soil microbial biomass and reducing the ineffective nodules, phosphate-solubilizing bacteria (PSB) like *Pseudomonas striata* and *Bacillus polymyxa* showed that mung bean growth and productivity had been positive, offering a biotechnological solution for sustainable agriculture(Hassan et al., 2017). PSB inoculation increases

the concentration of phosphorus in shoots and roots and its uptake in Mungbean varieties.

II. MATERIALS AND METHODS

A research entitled "Impact of added phosphorus and Phosphorus Solubilizing Bacteria (PSB) in yield and yield attributing characters of Mungbean (*Vigna radiata L.*)" was conducted at the agronomy farm of IAAS (Institute of Agriculture and Animal Science), Paklihawa campus during the summer season in 2023. The latitude and longitude of the farm's location are 27° 28' 48.252" North and 83° 26' 50.172"East respectively and 108 meters above sea level. The soil of the site before the research was 8.58. The climate of Paklihawa where the experiment was conducted is sub-tropical humid with very little rainfall. The size of the experimental unit was 4 m² (2 m*2m). The experiment was laid out in a Simple Randomized Complete Block Design (RCBD) with 7 treatments and 3 replications. Mungbean variety, Pratigya with different doses of phosphorus fertilizer, and PSB inoculation were the treatments. The collected data were tabulated in an MS Excel worksheet and analyzed in R studio software.

III. RESULTS AND DISCUSSION

3.1 Growth attributes

3.1.1 Plant height

Plant heights were recorded at 20DAS, 40DAS, 60DAS, and during harvest. At all growth stages plant height was maximum in T_7 (i.e. 13.47cm at 20DAS, 32.26cm at 40DAS, 55.06cm at 60DAS, and 61.65cm during harvest).

PSB inoculation with phosphorus fertilizer improved the availability of soluble phosphorus, which aided in plant growth and resulted in higher plant height (Singh et al., 2021).

3.1.2 Nodulation

Nodule numbers were recorded at 20DAS, 40DAS and 60DAS. The nodule number was significantly higher at T7 during all growth stages (i.e. 7.43 at 20DAS, 15.00 at 40DAS, and 29.90 at 60DAS). A higher No. of nodules/plant at a higher P level may have given better growth, which helps for nodule formation as PSB solubilizes insoluble P making it available to the plants. This is similar to the results of (Sumanth & Singh, 2021).

3.1.3 Dry matter

The finding of our research about dry matter content was not significant at 20DAS but significantly increased at 40DAS, 60DAS, and during harvest. The highest dry matter was observed in 20 kg P2O5/ha + PSB at all growth stages i.e. 0.247gm at 20DAS, 4.39gm at 40DAS, 26.14gm at 60DAS, and 30.74gm during harvest.

The increased availability of N, and P and their synergistic effect might have caused better dry matter accumulation, and as a result increase in the value of the above growth parameters was observed. Similar observation was also found in Mitra et al., (1999); and Perveen et al., (2002).

3.2 Yield and Yield Attributes

3.2.1 Pods per plant

Pods per plant were recorded highest in T7 i.e. 26.60 pods. The favorable role of phosphorus is mainly due to its primary effect in photosynthesis by way of rapid energy transfer which affects various biochemical processes from the beginning of seedling growth to the formation of grain and maturity (Sumanth & Singh, 2021).

3.2.2 Pod length

Pod length increased with phosphorus and PSB application in T7 which was recorded to be 8.24 cm. This might be due to the increase in vegetative development and reproductive attributes under proper availability of phosphorus and better physical condition of soil. The same results were revealed by Choudary et al., (2014).

3.2.3 Grains per pod

The highest grains per pod was recorded in T7 with 8.84 grains per pod. This is due to an increase in photosynthetic activity of leaves and translocation of photosynthates from source to sink and nutrient uptake by the application of bio-fertilizer and phosphorus dose. The minimum values of all the attributes were observed under the control plot because plants were unable to receive more nutrients. The results agree with those of Prakash et al. (2002).

3.2.4 Thousand seed weight

An increase in thousand seed weight was insignificant in T7 with the value 38.89 g. The increased availability of N, and P and their synergistic effect might have caused better dry matter accumulation, and as a result increase in the value of the above growth parameters was observed. Similar observation was also made by Mitra et al., (1999); Perveen et al., (2002).

3.2.5 Grain yield

The highest grain yield was obtained in T7 i.e. 2.44 ton/ha. Phosphorus is also a component of RNA, the compound that reads the DNA genetic code to build proteins and other compounds essential for plant structure, seed yield, and genetic transfer. These results are in line with Chowdary et al., (2003); and Yadav (2004).

3.2.6 Biological yield

Higher stover yield in the highest phosphorus treatment and PSB (30.2t/ha), though insignificant difference, might be due to increased availability of P which favored nodule formation, higher nitrogen fixation, dry matter accumulation, rapid growth, higher absorption, and utilization of other nutrients. The same results were revealed by Choudary et al., (2014).

3.2.7 Harvest Index (HI)

An increase in harvest index with phosphorus application (7.51 in T7) is the indication of better translocation of photosynthates from source to sink. These results confirm with the findings of (Singh et al., 2018).

IV. SUMMARY AND CONCLUSION

Results suggest that Mung bean can be an excellent rotational crop as it improves soil fertility as verified by various soil test results. The PSB helps to make insoluble phosphorus available to the plant by converting it into a soluble form. The plant, then, uses phosphorus for root growth, nodule development, and pod development. Ultimately, this results in better soil health and crop performance. As seen in the research, Phosphorus, Nitrogen, Potassium, and Organic Matter increased significantly in the integrated treatment of the highest level of phosphorus and PSB. Integrating phosphorus with PSB significantly increases growth, nodule number, yield, and yield-attributing characters in Mungbean.

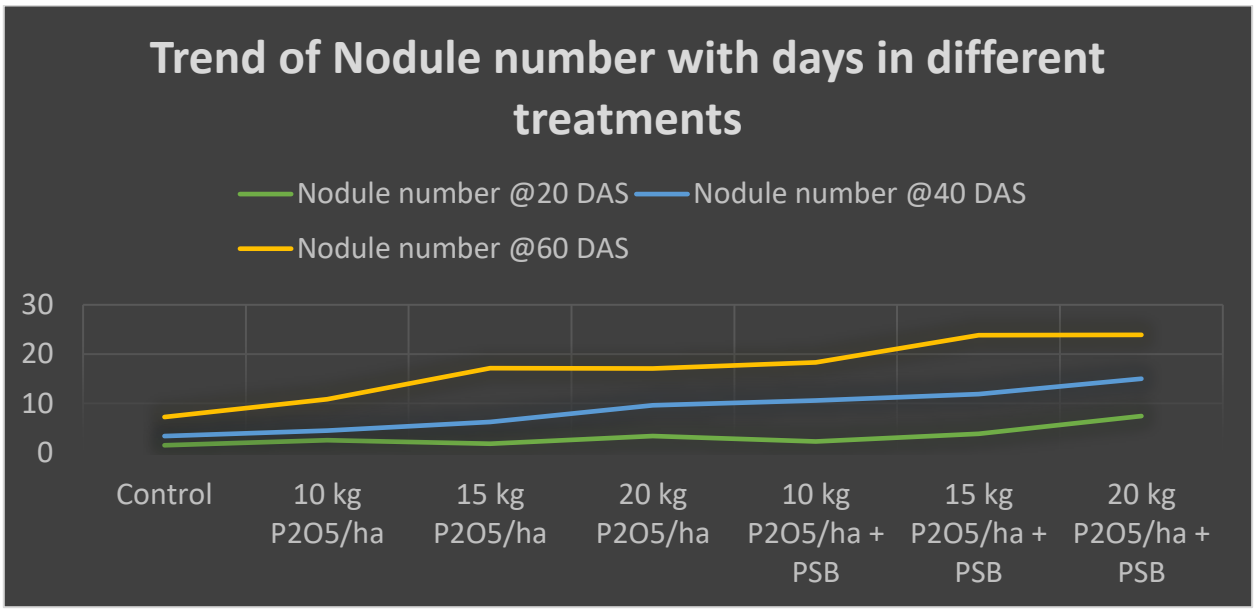

Fig.1: Effect of Phosphorus levels and PSB on nodule number at 20, 40 and 60 days of sowing

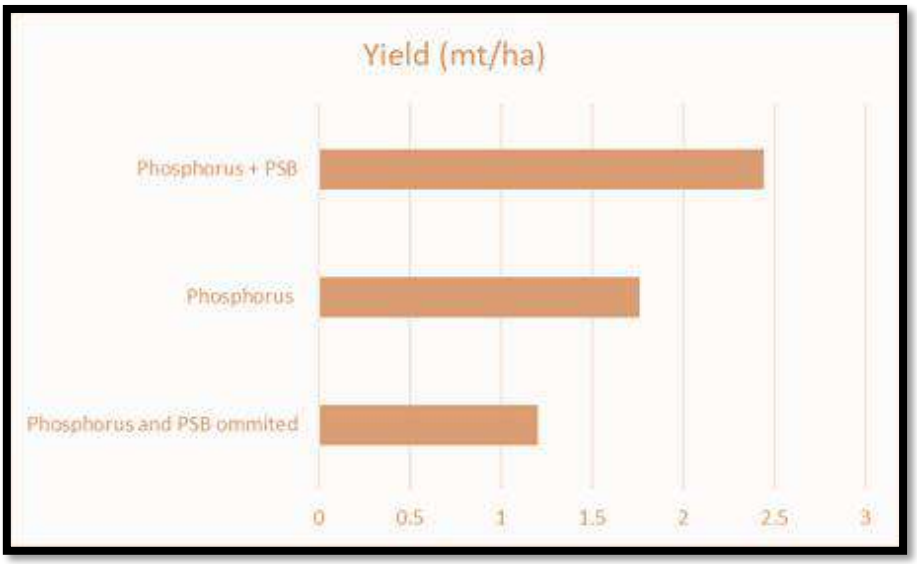

Fig.2: Yield variation in different conditions of fertilizer and bacteria addition in Mungbean at Paklihawa, 2023

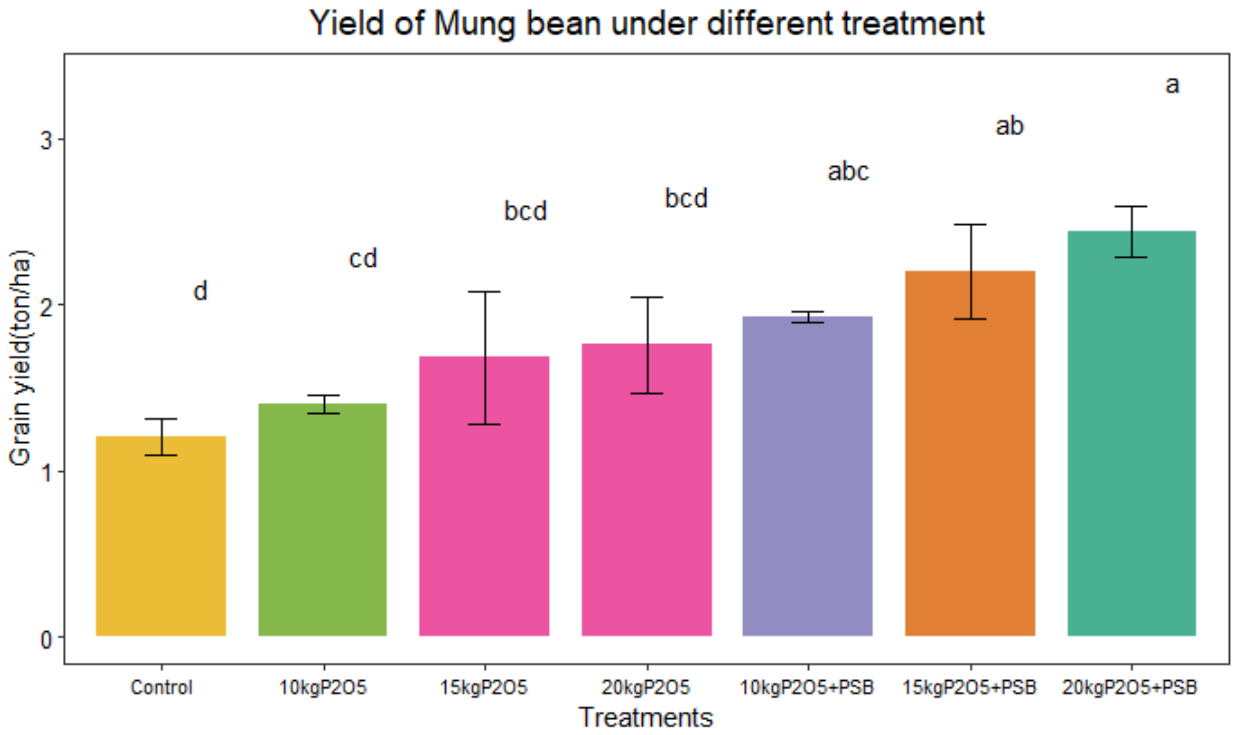

Fig.3: Effect of Phosphorus levels and PSB on yield of Mungbean at Paklihawa, 2023

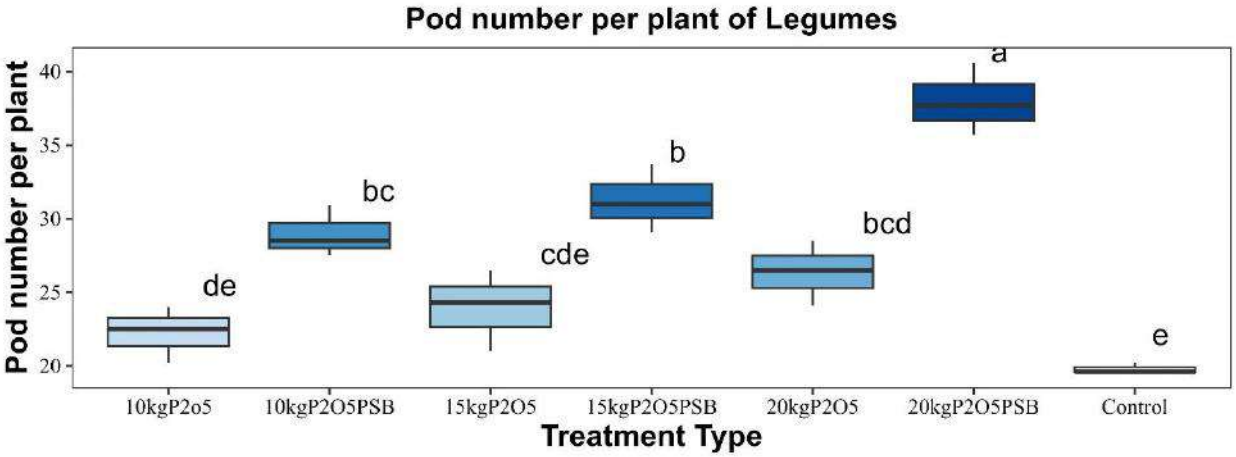

Fig.4: Effect on pod number per plant in different treatments of phosphorus and PSB in Mungbean at Paklihawa, 2023

Table -1 Effect of added phosphorus and PSB on plant height of Mungbean at Paklihawa, 2023

Treatments	Plant height@ 20DAS	Plant height@40DAS	Plant height@60DAS	Plant height during harvest
T1	11.63 ± 0.22^{c}	26.02 ± 0.46^{c}	47.21 ± 0.58^{e}	52.19 ± 0.92^{d}
T2	11.86 ± 0.44^{bc}	27.37 ± 0.56^{bc}	49.11 ± 0.60^{d}	54.66 ± 0.21^{cd}
T3	12.53 ± 0.54^{abc}	27.73 ± 0.71^{bc}	51.01 ± 0.67^{c}	55.54 ± 0.40^{cd}
T4	12.81 ± 0.07^{abc}	28.57 ± 0.48^{bc}	51.57 ± 0.64^{c}	56.79 ± 0.22^{bc}
T5	13.23 ± 0.14^{ab}	29.87 ± 0.81^{ab}	52.61 ± 0.60^{bc}	57.71 ± 1.04^{bc}
T6	12.82 ± 0.40^{abc}	30.39 ± 0.63^{ab}	53.45 ± 0.46^{ab}	60.51 ± 0.96^{ab}

T7	13.47±0.46[a]	32.26±0.63[a]	55.06±0.49[a]	61.65±0.68[a]
LSD	1.41	3.04	1.69	3.57
F-Prob	ns	*	***	**
SEm (±)	0.25	0.79	1.0007	1.245
CV (%)	6.28	5.93	1.85	3.52
Grand mean	12.62	28.88	51.43	57.009

Table -2 Effect of added phosphorus and PSB on nodule number of Mungbean at Paklihawa,2023

Treatments	Nodule number @20DAS	Nodule number @40DAS	Nodule number @60DAS
T1	1.5±0.64[b]	3.37±0.12[d]	7.23±0.24[c]
T2	2.5±0.45[b]	4.50±0.48[d]	10.83±1.43[bc]
T3	1.83±0.78[b]	6.23±0.32[cd]	17.17±2.87[ab]
T4	3.37±0.65[b]	9.63±1.91[bc]	17.10±2.74[ab]
T5	2.27±0.58[b]	10.60±0.65[b]	18.30±2.57[a]
T6	3.83±0.15[b]	11.90±0.20[ab]	23.80±3.51[a]
T7	7.43±0.37[a]	15.00±0.56[a]	23.90±3.38[a]
LSD	2.29	3.82	6.49
F-Prob	**	***	***
SEm (±)	0.76	1.59	2.33
CV (%)	39.69	24.52	21.59
Grand mean	3.24	8.75	16.904

Table -3 Effect of added phosphorus and PSB on dry matter of Mungbean at Paklihawa, 2023

Treatments	Dry matter@ 20 DAS	Dry matter@40 DAS	Dry matter@60 DAS	Dry matter during harvest
T1	0.16±0.01[b]	1.99±0.17[d]	12.65±1.15[c]	19.35±0.36[e]
T2	0.21±0.02[ab]	3.18±0.27[bc]	15.74±1.74[bc]	22.68±1.36[d]
T3	0.19±0.02[ab]	2.63±0.09[cd]	16.44±0.63[bc]	24.37±2.33[cd]
T4	0.21±0.01[ab]	3.26±0.17[bc]	19.11±1.70[b]	26.06±2.50[bc]
T5	0.22±0.24[ab]	3.73±0.32[ab]	17.33±1.1[bc]	25.96±0.28[bc]
T6	0.23±0.01[a]	3.54±0.23[abc]	19.32±3.60[b]	27.59±2.13[b]
T7	0.25±0.01[a]	4.39±0.52[a]	26.14±0.89[a]	30.74±2.51[a]
LSD	0.06	0.89	4.96	1.92
F-Prob	ns	**	**	***
SEm (±)	0.01	0.29	1.58	1.37
CV (%)	15.68	15.40	15.41	4.28

Grand mean	0.21	3.25	18.10	25.25

Table-4 Effect of added phosphorus and PSB on yield and yield attributing characters of Mungbean at Paklihawa, 2023

Treatments	Pod per plant	Pod length (cm)	Grains per pod	Test Weight (g)	Grain yield (mt/ha)	Biological yield (mt/ha)	Harvest Index (%)
T_1	19.37 ± 0.12^{g}	6.96 ± 0.06^{e}	6.95 ± 0.09^{d}	35.64 ± 1.11^{a}	1.20 ± 0.041^{e}	17.8 ± 0.3^{e}	6.30 ± 0.14^{a}
T_2	20.93 ± 0.51^{f}	6.44 ± 0.11^{d}	7.21 ± 0.04^{d}	36.30 ± 0.86^{a}	1.40 ± 0.019^{de}	21.1 ± 1.11^{de}	6.27 ± 0.23^{a}
T_3	23.63 ± 0.43^{e}	7.55 ± 0.11^{cd}	7.58 ± 0.01^{c}	36.85 ± 2.79^{a}	1.68 ± 0.14^{cd}	22.8 ± 0.72^{cd}	6.94 ± 0.75^{a}
T_4	24.47 ± 0.37^{d}	7.69 ± 0.11^{bcd}	7.87 ± 0.08^{c}	37.19 ± 1.52^{a}	1.76 ± 0.10^{cd}	24.8 ± 1.18^{cd}	6.74 ± 0.65^{a}
T_5	25.20 ± 0.18^{c}	7.82 ± 0.07^{bc}	8.31 ± 0.09^{b}	37.35 ± 0.16^{a}	1.93 ± 0.012^{bc}	26.8 ± 0.88^{bc}	6.75 ± 0.20^{a}
T_6	25.83 ± 0.30^{b}	7.95 ± 0.05^{b}	8.77 ± 0.08^{a}	38.24 ± 1.86^{a}	2.20 ± 0.10^{ab}	27.8 ± 1.06^{ab}	7.39 ± 0.51^{a}
T_7	26.60 ± 0.17^{a}	8.24 ± 0.05^{a}	8.84 ± 0.08^{a}	38.89 ± 0.86^{a}	2.44 ± 0.05^{a}	30.2 ± 0.79^{a}	7.51 ± 0.23^{a}
LSD	0.47	0.27	0.35	7.76	0.40	4.73	1.83
F-Prob.	***	***	***	ns	***	***	Ns
SEm	1.00	0.15	0.28	0.42	0.16	2.00	0.18
CV (%)	1.11	1.99	2.48	11.72	12.57	10.53	15.10
Grand mean	23.72	7.67	7.93	37.20	1.80	25.27	6.84

Table-5 Effect of added phosphorus and PSB on soil parameters before cultivation and after harvest of Mungbean at Paklihawa, 2023

Soil parameters	Total Nitrogen (%)	Phosphorus (kg/ha)	Organic matter (%)
Before Mungbean cultivation	0.04	35.88	1.77
T1	0.06^{b}	34.27^{e}	1.83^{e}
T2	0.074^{ab}	37.61^{d}	1.90^{d}
T3	0.076^{ab}	44.17^{c}	2.17^{c}
T4	0.09^{ab}	47.17^{b}	2.20^{c}
T5	0.08^{ab}	47.45^{b}	2.37^{bc}
T6	0.09^{ab}	48.17^{b}	2.79^{ab}
T7	0.1^{a}	54.7^{a}	3.32^{a}
LSD	0.026	2.85	0.95

F-Prob.	ns	***	***
SEm	0.0049	2.6	0.54
CV (%)	14.2	3.58	11.41
Grand mean	0.08	44.79	2.28

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Optimization and Modeling for the use of Machine and Maintenance in Agricultural Production System in Allahabad District (Uttar Pradesh), India

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Abstract— *This study constitutes the status of Machinery Power in Allahabad district based upon the survey conducted and data as references and hence proposing an estimated data of the current status of power utilization in Allahabad district and more specifically its three block; viz, Chaka-Block, Jasra-block and Shankargarh-block. Based on these three different block's differences, the study was conducted for different farming system of Allahabad district to find the status of energy inputs and availability of farm machinery for particular system in blocks. The survey was carried out in the month of March to April, 2016 find the ". One hundred and sixty seven farmers were selected considering the owning improved implements of bullock, tractor and power operated categories were surveyed this study. The data were collected in the prescribed profarma (Appendices .A,B,C and E,F,G) w.r.t. farmers and implements separately, The data regarding farm machines and implements have also been collected from Block development office Allahabad, The survey data indicate that the number of implement per hector, cultivated area in different selected blocks was361.65%, 274.96% and 333.41% in Chaka, Jasra and Shankargarh, respectively with the district average of 36.3. The percentage of Chaff-cutter in different blocks varied from 16.66%,19.64% & 19.60% in Chaka, Jasra and Shankargarh blocks, respectively. Plant protection equipment was very low as the number of sprayer and duster75%,8.35% & 72.54% cultivated area in surveyed blocks. However, most of the farmers were using plant protection equipment and other machines on custom hiring basis. Power operated centrifugal pump sets were most common for under around water pumping and its availability was highest in Chaka block 10.71 % cultivated area followed by 13.33% and 11.76%in Jasra and Shankargarh blocks, respectively.*

Keywords— ***Agriculture production system, optimization of machine, modeling of machine, maintenance of machine, Lucknow, Agricultural Engineering.***

I. INTRODUCTION

Agriculture being the backbone of Indian economy needs a thrust, as recent surveys show that agricultural growth rate seems to be stagnant. Agriculture has an immense effect on our GDP growth rate. As about 13.6 of our GDP come from agriculture, Nearly 58% of our country population depends up on agriculture as sources of their livelihood. But after numerous govt. Initiatives the results are not up to the mark.

Farm mechanization helps in effective utilization of input to increase the productivity of land and labour. Beside it helps in reducing the drudgery in farm operation.

The early agricultural mechanization in India was greatly influenced by the technological development in England. Irrigation pump, tillage equipment, chaff cutters, tractor and thresher were gradually introduced for farm mechanization .The high yielding varieties with assured irrigation and higher rate of application of fertilizers gave higher returns that enabled farmers to adopt mechanization inputs, especially after Green revolution in 1960s.

Agricultural mechanization is the process whereby equipments, machineries and Implements are utilized to boost agricultural and food production. It is the application of machineries, equipments and implements in the day to day farm activities to increase marginal output in food production and poverty eradication.

Uttar Pradesh has a population is estimated to be 215 million people, of which nearly 70% is dependent on agriculture directly or indirectly. Therefore, about 13 corer people are linked to farming activities, over 21 million lands holding and six to seven people linked to each land holding. Agriculture is the most crucial sector for socio economic development of state. It contributes the highest share of 33% to the total income of the state. A higher growth in the state's total economy cannot be achieved or sustained on a long term basis, without good growth in Agriculture. In U.P., Agriculture plays a dominant role in the economy and growth rate of Agriculture and allied sector is 5.3 percent that of primary sector of 5.4 percent, and the national figure is 4.0 percent. Secondary and Tertiary sector are also growing very fast in the country as a whole but in U.P though growth of secondary sector is close to national figure, the tertiary sector is lagging behind. Yet Agriculture and allied sector growth being the lowest has a potential to improve and thus provide boost to the overall economy of the state. So by using farm mechanization and proper training of advanced machinery of agriculture, the productivity of the agriculture field and economy of the state could be improved. Farm machinery can reduce the time of different process of the agriculture and help the farmers to improve the quality of their crops.

As major increase of agricultural production and rural development, mechanization aims are increase the power input to farming activities, hence putting more land into production. There are direct and as well as indirect effects of agricultural machinery and implement on productivity through better use of other input more efficient and timely completion on agricultural operation and increase in cropping intensity ***(Venugopal, 2004).*** Agricultural mechanization and conservation agriculture refers to interjection of improved tools, implements and machines between farm workers and materials handled by them. Independent Indian ushered in a process of agricultural mechanization and revival of rural agro-processing which got acceleration during post-green revolution period. Irrigation pump sets, power thresher , tractor, power tillers and matching implements, including for 65 million draft animal have became popular.

Allahabad District is in the Uttar Pradesh, Allahabad district is located at the confluence of Ganga and Yamuna. The Allahabad district is located at 25°26' N/ 81°50' E at elevation of 98m (322FT). 25.45 latitude and 81.83 longitude and it is situated at elevation 104 meter above sea level .Allahabad District is one of the productive regions of Uttar Pradesh and a majority of farmers are employed in agricultural activates. optimization to assess status of mechanization have been done in past, but due to the advent of new machine and technological advancement the use of farm machines has considerably increased. So a current optimization was felt needed. This study will help to make strategies for propagation of mechanization in the region.

It has been reported by the research workers ***(Srivastava et al. 2000)*** that timely and proper seed bed preparation results in increase of production by 20 to 30 percent. Similarly, proper placement of seeds, results in 10 to 20 percent higher yields. Weed control and plant protection are important tools for increased production. Timely harvest by appropriate implements and machines results in more and better quality agricultural produce. The increased intensity of cropping in the modem agriculture also leads to the increased use of improved implements and mechanical equipments for land leveling. ploughing, seed-bed preparation, sowing, plant protection, inter-culture, harvesting and threshing. It also leads to the requirement for better facilities for transport of crops storage. Processing and marketing.

The scope of farm mechanization has generated demands for design and manufacture of new agricultural implements and machinery in the country. It has been observed that tools /implements/machines developed as a result of research and development efforts of ICAR and agricultural universities have not been adapted by the farmers to the desired level. Commercialization of these machines also takes long time. This is due to insufficient efforts put in to analyze the users need and identification of product specifications and their development with due consideration of its market. Farmers need analysis has to take into account the cropping systems and farming practices prevalent and the socio-agro-economic condition in the region.

The mechanization process being emphasized in the country is still beyond the scope of the small scale farmers who produce the bulk of the food. The present study was planned to study the status of mechanization of major cropping system and agricultural practices followed in the Yamuna par region of Allahabad to identify mechanization gaps and needs of farm tools machines along with their broad specification for development.

Status of Agricultural Mechanization in India:

Verma (1988) studied the appropriate farm mechanization for sustainable agriculture in Eastern U.P. The study showed that in eastern region only one type of mechanization can not be adopted due to great diversity in land holding size, farm size, power source, socio economic condition of farmers. The use of improved and matching equipment package showed the increase in grain yield by 15-20 % of different crops and considerable saving in production cost with an increased command area in comparison to traditional fanning .Study also showed that level of mechanization had improved considerably in India, but efforts were not only to be continued, but also to be accelerated.

Position and Utilization of Farm Tractors in India:

Ram (1996) conducted study to observe trends of farm mechanization in South Bihar alluvial plain zone. It was observed that the maximum number of tractors (44%) was operated for 800-1100 h per year. The common implements used were field cultivator, mould board plough, leveler, crop thresher and trolley. The tractors were used for about 32 % of time for custom work.

Singh and Dohery (1999) studied the position of tractor industry in India. Their analysis showed the number of tractor produced in India had increased from 0.12 million in 1971 to 1.24 million in 19-91 with overall annual growth rate of 9.25 percent.

Singh, Gyenendra (1999) studied the use of draught animal power in India and concluded that use of draught animal power is reduced in farm power. Percentage of draught animal power in total power available KW/ha has decreased from 52.12 percent so 12.72 percent between 1971 to 1991

De et al. (2000) analyzed the data of the AICRP on Energy Requirements. in Agricultural Sector to assess the farm power availability in selected states which showed that draught animal population, mainly derived from bovines, was 80.17 million in 1972 and reduced to 67.02 million by 1982. Thereafter, the population recorded increase to 78.54 million in 1992. Analysis showed that national scenario about tractor population .Tractor population in India has from 119.39 thousands in 1971 to 2600 thousands in the year 2000 at an annual growth rate of 11.21 percent between 1971 to 1981, and decreased to 9.25 percent during next ten year. 2.3 Farm Power Availability in India:

Impact of Mechanization on Cropping Intensity and Productivity:

Srivastava (1999) suggested ways in which the Agricultural Engineering Technology could help in accelerating the growth of agricultural production in next ten years to achieve high target. He observed that the some farmers who had achieved yields of 8-12 tonnes/ha they are belonged to mechanized category of farmers. On the basis of analysis of trend in food grain production in important states during 1996-97, it was observed that the main reason of low productivity was low availability of farm power per ha, non availability, of improved and high capacity precision equipment for timely farm operations, less irrigation, low fertilizer consumption, less use of high yielding varieties and non availability of appropriate post harvest technology for reducing post harvest losses. The improved farm implements and machinery had to play important role in timely farm operations at reduced cost, maximizing efficiency of agricultural inputs by-proper placement/application and minimizing losses in production, processing, handling, transport and storage of grains, fruits, vegetables and other agricultural produce.

Impact of Mechanization on Employment

Balishter, Gupta and Singh (1991) conducted a study in Mathura district of Uttar Pradesh on the basis of three levels of mechanization, i.e. (i) non-mechanized farms having neither tube-well nor tractor (ii) Partially mechanized farms having only tube-well and (iii) mechanized farms having both tube-well and tractor. The yield was reported to be higher about 10 to 27 percent in mechanized farms and by about 2 to 26 percent in partially mechanized farms in comparison with non- mechanized farms for all the major crops grown on the sample farms.

II. MATERIALS AND METHODS

The survey has been conducted to all eight Tehsil of Trans-Yamuna region of Allahabad Districts; by choosing 167 farmers who have different land capacity. The methodology adopted has been described under the following heads provided with the entire study of optimization and modeling for the use of appropriate machine and maintenance in agricultural production system of Allahabad.

Description of Study Area

Allahabad Division of Uttar Pradesh. Allahabad is a district in the Uttar Pradesh State of India. Total area of

Allahabad is 5,482 km² including 5,279.07 km² rural area and 202.93 km² urban area. Allahabad has a population of 59, 54,391 peoples.The Allahabad district is further divided in to 20 Blocks/Tehsils for administrative purposes.

Classification of Energy on the basis of Farm operations (MJ/ha)

Land preparation: Typically involves ploughing, harrowing and leveling the field to makes it suitable for the crop established. Draft animals, such as buffalo or tractor can all be used as power sources in land preparation. The initial soil tillage can also be performed with a cultivator instead of a plough.

Sowing : It is the process of casting handfuls of seed over prepared ground broadcasting (for which the technology term is derived from).Usually, a drag or harrow is employed to incorporate the seed into the soil. Though labor intensive for any but small areas .A hand seeder can be used for sowing, though it is less of help it is fir the smaller seeds of grasses and legumes. Seeds now sown using a seed drill, pneumatic planter which offers greater precision; seed is sown evenly at the desired rate. The drill also places the seed at a measured distance below the soil, so that less seed is required. The standard design used a fluted feed metering system, which is volumetric in nature; individual seeds are not counted. Row is typically about 10-30cm apart, depending on the crop species and species and growing conditions. Several row drawn by the tractors, but can also be pulled by horses

Irrigation : It is an artificial application of water to the soil. It is used to assist in the growing of agricultural crops, maintenance of landscapes, and re-vegetation of disturbed soil in dry areas and during periods of inadequate rainfall. Additionally, irrigation also has a few other use in crops production, which includes protecting plants against frost, suppressing weed growing in grain fields and helping in preventing soil consolidation. In contrast, agriculture that relies only on direct rainfall is referred to as rain-fed or dry land farming. Irrigation is also used for dust suppression, disposal of sewage, and in mining. Irrigation is after studied together with drainage, which is the natural and artificial removal of surface and sod-surface water from a given area. Various type of irrigation technology differs in how the water obtained from the source is distributed with in the field. Tube wells pumping sets operated by diesel engine were used for irrigation.

Harvesting: Sickles were used for manual harvesting. Farmers generally use Combine harvester, Reaper binder, mower self-propelled or tractor operated for harvesting their crops.

Threshing: Method of threshing includes beating the crop material with bamboo grates or wooden plank manually while in partly mechanized system threshing was done by the tractor operated thresher. Mechanical thresher wheat and paddy and pedal operated paddy thresher are used the large and medium farmers.

Population and sample size : From a 3 different block population of Allahabad Fast Track Land Reform Program (FTLRP) of 2000 successfully resettled a total population 9,76,733 households under A1 model and 1,684 beneficiaries under A2 farming. The study used data from 80 Fast Track Resettlement farmers, and a control group of 10 traditional commercial farmers, purposively sampled. A stratified random sampling technique was used to select 167 Resettled farmers in the study area.

Data collection techniques

Interviews

An interview is a series of questions a researcher addressed personally to the farmers. Both structured (clearly defined questions) and unstructured, questioning led to the farmer of the interviewee were used. Interviews were advantageous in discovering with individual farmers thought and felt about impacts of mechanization on the current trends of agricultural performance and kind of farmers had such opinions.

Questionnaires

A questionnaire with a serial of questions was applied to responds for questions , to get their response. The way the data were to be analyzed influenced according to the layout of the questionnaire. The questionnaire made use of closed questions which provided boxes for the respondents to tick. The study avoided open questions as these would require respondents to write answers, though giving more freedom of information;

Analytical tools

Mechanization index: qualitative and quantitative criteria by which the impact of mechanization was used to identify the significance machinery use efficiencies with different farm categories.

III. RESULTS AND DISCUSSION

Based on the objectives the Results derived were quite of use for us. Agricultural mechanization in different Farming practices in blocks, the studies were conducted and the results found are hence wise compiled and is presented here as followings.

The status of selected farm implement used for different farming system :

The Farm Implements used generally in Allahabad regions are Cultivator, Mould Board Plough, Disc Plough, Harrow, Leveler, Sprayer, Seed-Drill, Planter, Transplanter, Reaper and Thresher. In all the blocks, the farming system defined these implement are in use of Cultivator, Mould Board Plough, Disc Plough and other farm implements. Hence, it can easily be said that Farming practices for three block can be use with any farm implements. (Appendix A,B.D; Table A-1, A-2, A-3).

Status of Improved Implements Use

For the purpose of this study, information regarding availability of improved tools and equipment used by farmers in die selected villages have been collected and presented below.

Status of manually operated equipment use

The block wise data regarding the use of manually operated tools and equipment based on sample survey on the basis of marginal ,small and medium of cultivated area have been given in the table for Chaka ,Jasrs and Shankargarh. For weeding purpose manually operated improved weeders were not found in all selected surveys villages whereas there is good scope of their introduction because most of the crops are shown in line. For Chaka, Jasra and Shankargarh block in Allahabad district serrated sickle, was used by 28.33%,35.71% & 25.49% farmers. Herbicide were used by 8.33%, 7.14% &7.84% farmers.

The number of chaff cutters in different blocks 16.66%,19.64% & 19.60% aried from to marginal, small & medium.The table indicates that the use of self purchased plant protection equipment was very low as the number of sprayer and duster varied from 75%,8.35% & 72.54% only in different surveyed blocks .

Status of tractor and tractor drawn equipment

The position in respect of the numbers of tractors and tractor drawn equipment like disc harrow, cultivator, cage wheel. leveler, bund former, seed cum ferti drill, potato digger, ridger, harvester, thresher, trolley, puddler etc. In three selected villages of each block of district Allahabad has been studied and data are presented in. The block-wise data of these equipments based on this sample survey are also given in(Appendix A,B.D).

. The sample survey data given in table indicates that the number of tractor per ha cultivated area in different selected blocks was 361.65%, 274.96% and 333.41% in Chaka, Jasra and Shankargarh respectively . The tractor drawn primary tillage implement like ploughs, sub soiler etc. were almost non existent through out the all selected villages. It was observed that the tractor drawn cultivator is the only equipment which is available with almost every tractor owners. The harrow which is one of the most important secondary tillage implements was also owned by 32.14%,30% & 31.36 of tractor owner farmers. The other seed bed preparation implements like leveler, puddler, ridger, etc. were very rarely found with the farmers. The trolley is another equipment which was very much popular and almost all tractor oweners were trying to have during survey 83.33%,73.21%,& 76.46 % tractor farmers were found to be own it. The number of seed cum ferti drill varied from 23.33%,17.85 & 41.16% in surveyed blocks. The use of seed cum ferti drill was found in Chaka, Jasrs and Shankargarh blocks by some tractor owners, but other farmers and also some of tractor owners hire seed-drill to fulfill their requirement. The use of other tractor drawn equipments like potato digger, sugar cane planter, reaper etc. were rarely found in selected blocks of Allahabad district.

Status of Engine operated equipment use

Information in respect of power operated equipment like centrifugal pumps, water pumps collected from selected villages are given in Appendix G Table 2. The consolidated block-wise based on sample survey have been presented.

The centrifugal pumps were the most common engine operated irrigation equipment used for underground water pumping as well as lifting of water from shallow water sources like wells, ponds, rivers, canals, etc, throughout the surveyed area of Allahabad district. The availability of these pumps in Chaka (10.71%) and Jasra(13.33).and Shankargarh(11.76).

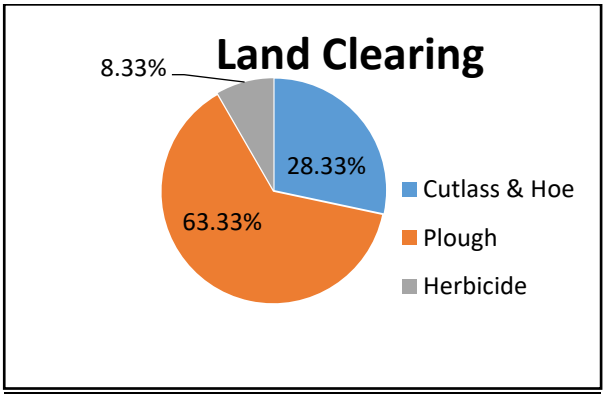

Fig.1: Land Clearing for chaka block

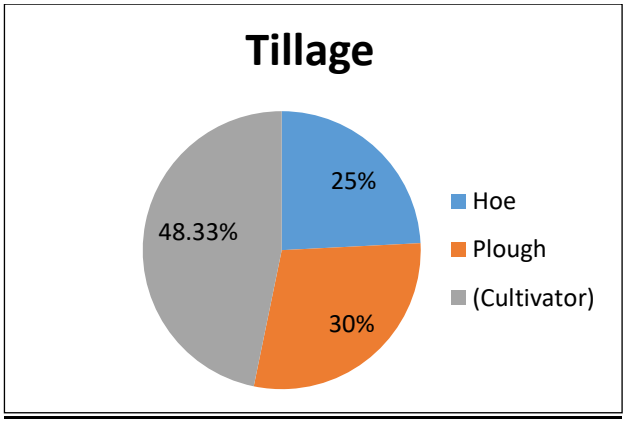

Fig.2: Tillage for chaka block

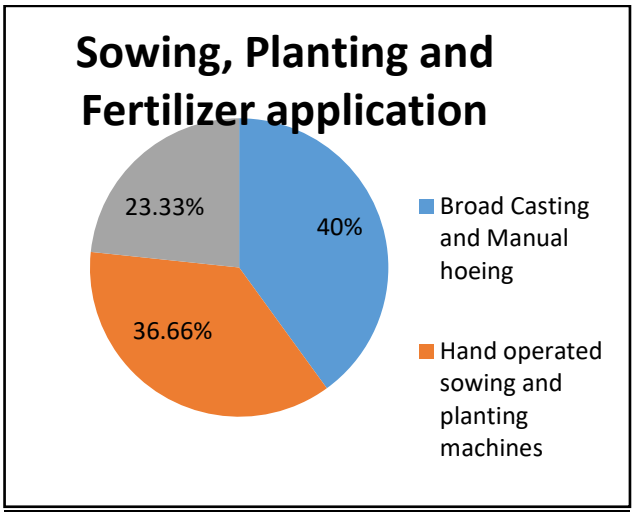

Fig.3: Sowing, Planting and Fertilizer application for Chaka block

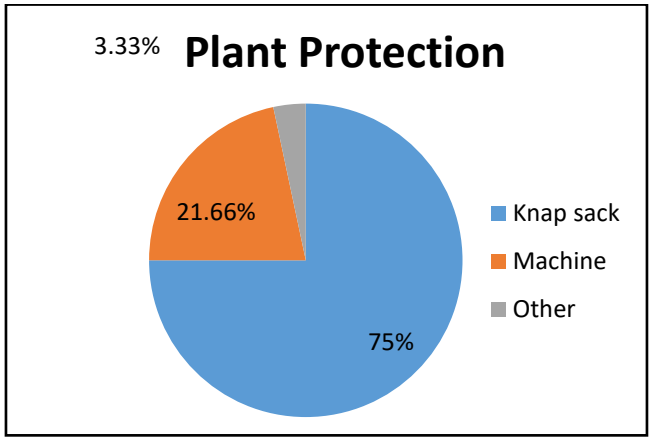

Fig.4: Plant Protection for chaka block

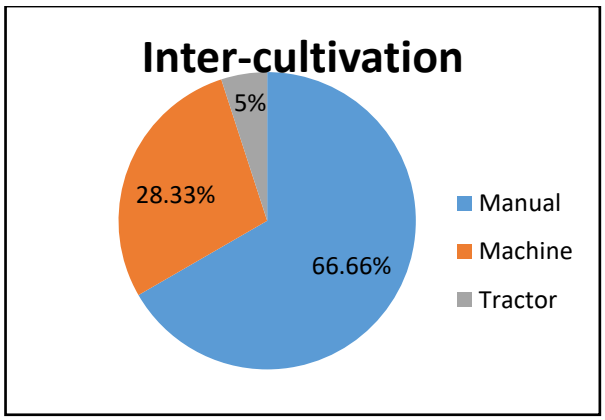

Fig.5: Inter-cultivation for chaka block

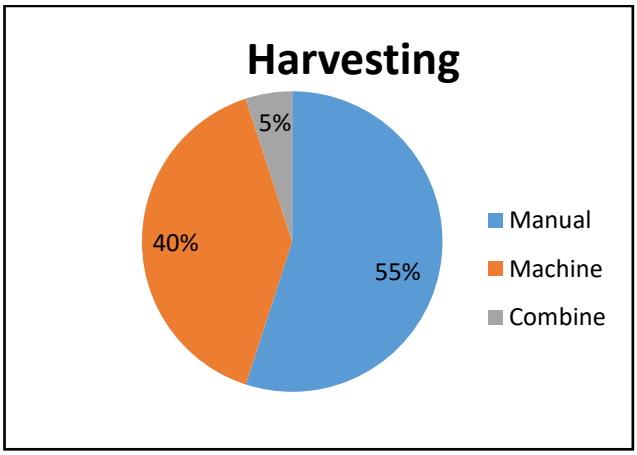

Fig.6: Harvesting for chaka block

IV. CONCLUSION

The salient findings of the survey are reported herein.

- Land holding size per household fanner was found (0,74 ha/farmer) very low. At present in Allahabad district as per survey data land holding size per house hold farmer (0.74 ha) was less than operational land holding size of the U.P. state (1.1 ha) according to statistical bulletin (Anonymos-1993). The productivity of land and percentage irrigated area are mostly higher in mechanized fanning areas like Jasra, Shankargarh etc. than other non mechanized blocks of Allahabad district.
- Manually operated improved tools and equipment available with the farmers were serrated sickle(28.33,35.71 % 25.49) plant protection equipment (sprayer 75%,80.35% & 72.54%and duster 3.33%,1.78% & 3.92%) , (16.66%,19.64% & 19.60%)winnower and chaff cutter only.
- Tractor drawn cultivator and trolley were the only equipment owned by most of the tractor farmers. The disc harrow and thresher were other common

equipment in use. The presence of tractor drawn seed cum fertilizer drill and potato planter were less but farmers enthusiastic to own them. The use of other improved implements like reaper, potato digger, harvester, ridger and cage wheel etc. were very rare.

- Power operated centrifugal pump sets were most common for under ground water pumping and its availability was highest in Chaka block 10.71% cultivated area followed by 13.33% and 11.76% vcultivated area in Jasra block and Shankargarh block respectively. But there was need for increasing the number of pump set in Allahabad district to have more irrigated area .The use of electric motor was low as compared to diesel engine due to uncertainty of availability of continuous electricity.
- Most of the farmers of entire district were enthusiastic about the use of improved farm tools and equipments. They felt that the improved farm implement and equipment have increased their production, efficiency and quality of operations and reduced the drudgery involved in conventional farm operations. Presently almost most of the farmers were using locally available improved implements and equipment on custom hiring. It was observed that tractor drawn farm machinery compared to traditional local ploughs, disk harrows and cultivators cover more than 8-10 times and ensure timeliness besides drudgery reduction.
- Survey revealed that the main problems faced by the farmers in adoption of the improved farm implements were: lack of awareness about various implements, non availability of quality implements, lack of effective field demonstration, high initial cost, difficulty in credit availability, high interest rate on credit, poor after - sale service etc.
- The immediate need of farmers of different blocks of Allahabad district with regard to farm implements were almost same. Implements like paddy transplanter, seed-cum-ferti drill, potato planter, potato digger, harvester, sprayer and weeding tool are their common need.

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Effect of use Organic Fertilizer on Yield component yield and quality of Hatri 10, Hatri 475 rice on Chau Phu a Giang, Vietnam

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Web: hatri.org

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Abstract— *The present study aimed to determine the effect of different organic and inorganic source of fertilizers on growth and performance of rice. The experiment was conducted at locations: Chau Phu A Giang province with two varieties of improved rice with HATRI 10 and HATRI 475. Each experiment has seven treatments. Treatments included a combination of organic and inorganic nutrients at seven rates (F1: 80-40-40+ organic manures 10 t ha-1; F2: 60-40-40+ organic manures 10 t ha-1; F3: 40+40+40+ organic manures 10 t ha-1; F4: 20-40-40+ organic manures 10 t ha-1. F5: only organic manures 12 t ha-1; F6: control no dose of NPK; F7: farmers used: 120-40-60+ organic manures 12 t ha-1). The experiment is arranged on the farmer's field, a split-plot in a randomized complete block design with three replications. Treatments produced significant results for plant height. Panicle length and grain yield but thousands of grains weight was not significant. Result showed that application of half of recommended 60-40-40 + organic manures 10 t ha-1 produced significantly higher value for grain yield and good for quality improve variety.*

Keywords— ***nutrition N. P. K. productivity. factors that constitute productivity.***

I. INTRODUCTION

Depending on chemical fertilizers as a source of nutrition, besides it tends to reduce soil yields causing a decrease in yield. It also reduces the quality of rice. For example the structure of rice becomes hard due to its high amylose content and low amylopectine (Jian et al., 2004).Some traditional Sabah local varieties have the potential to be grown as they can contribute to higher rice yields and their accommodation rates can be minimized by the adoption of NPK. The Serendah Merah (V3) variety received with F1 has no significant difference between the NPK fertilizer applied. So it can be recommended for farmers, the amount of fertilizer used in F1 treatment (60:30:30 kg ha-1) is the least. So it offers an economic advantage because low fertilizer costs are needed to achieve higher yields and better grain quality. (Mohd et al.,2018). A field experiment was conducted for the effect of different sources of nutrients on NPK uptake by rice at various growth periods. The NPK uptake by rice at various growth periods was significantly increased with the application of 100% NPK in combination with FYM @ 10t ha-1. However, it was on par with that of green manuring together with 100% NPK during both the years of the study (Mohana et al.,2017). Most of the landrace rice land in An Giang belongs to the group of poor sandy soils and uneven distribution of rainfall during the year. In addition to the use of low-yield genotypes (Ishag. 1980). The objective of this study is to improve the yield of landrace rice varieties and find a relationship between the dosages of N. P. K and integrated application of organic manures and inorganic fertilizers was effective for enhancing growth, yield, and the yield components of landrace rice.

II. MATERIALS AND METHODS

2.1. Two varieties HATRI 10, HATRI 475

2.2. Experimental experiments were conducted in Chau Phu An Giang provinces, with silt soil structures. Experimental soils have been cultivating seasonal rice for twenty years and in recent years have been managed in the conservation system for the seasonal rice region. Prior to the experiment the soil layer was collected in each area in layers 0 to 30 cm deep to make up the composite sample. which was used to analyze chemical indicators according to the method of (Raij et al., 2001) and particle size according to (Camargo et al., 2009) - Experimental layout: The experiment is arranged on the farmer's field A split-plot in a randomized complete block design (02 varieties, 7 experiments,3 repetitions, at 01 locations, the area of each laboratory is 25 m2).

Experimental layout method

No	Treament : (F)= (N-P-K)
1	F1 = 80-40-40 + organic manures 10 t ha-1
2	F2= 60-40-40 + organic manures 10 t ha-1
3	F3= 40-40-40 + organic manures 10 t ha-1
4	F4= 20-40-40 + organic manures 10 t ha-1
5	F5= only organic manures 10 t ha-1
6	F6= 0-0-0+ 0
7	F7= farmers (120-40-60 + organic manures 12 t ha-1)

About fertilization: only use innocuous fertilizers to assess the effect of nutrients on rice crops. experiments do not use compost and other fertilizers. Single forms of in innocuous stools are used as follows: urea (46% N). phosphate supe (16% P2O5. 20% CaO) and potassium chloride (60% K2O).

Agro-morphology Analysis

HATRI 10 varieties were planted in the field at Chau Phu . During the wet season from 2023. Seeds were sown in the raised seedbeds. and 15-day old seedlings were transplanted at one seedling per hill. Hills were established at distances of 15 x 20cm. The standard cultural management practices for rice were followed (Bui. 1986).

Quality traits

A total of HATRI 10 varieties were evaluated (Table 1) and the following quantitative traits were considered: Panicle length (cm): length of panicle at maturity measured from the base of the plant to the tip of the panicle (taken from 10 random selected primary panicles per accession per replication). Panicles per plant (number): the total number of panicles per plant (from 10 random selected primary panicles per accession per replication). 1000-grain weight (g): weight of 1000 welldeveloped grains at 14% moisture content (from 5 random selected primary panicles per accession per replication). Filled grains (number): obtained from counts of total number of filled grains per panicle (from 5 random selected primary panicles per accession per replication). Unfilled grains (number): obtained from counts of total number of unfilled grains per panicle (from 5 random selected primary panicles per accession per replication). Yield obtained from the harvested plants in each replication. Harvested grains were threshed, cleaned, drie, and weighed for each accession per replication. Moisture content per plot was determined immediately after weighing using a moisture meter. Yield = weight of harvested grain (g)/number of hills harvested x number of possible hills x MF (of the harvested grain).

Cooking and eating properties

Milled grains underwent assessment of physical traits (grain dimensions. proportion of head rice in milled rice, and chalkiness) and then a test portion of each sample was ground into fine flour (100-mesh) using a Udy Cyclone Sample Mill (model 3010–30. Fort Collins. CO). Reverse osmosis (RO) water and reagent-grade chemicals were used for the chemical analyses.

+ **Amylose content**: The AAC of isolated rice starch was analysed by using the iodine reagent method [AACC International.1999]. Briefly,exactly 25mg rice flour was gelatinized overnight in 2ml of 1.0N NaOH in a water bath set at 50°C. The solution was boiled in the water bath for 10 min and then cooled to room temperature. The cooled solution was extracted three times with 5ml of butanl: petroleum ether (1:3) to remove the lipid. After which 1.5ml of 0.4N KI was added to the solution and mixed. The AC was determined in duplicating with an ART-3 Automatic Titrator, according to the manufacturer's instruction (Hirama Laboratories. Japan) in which 1.57mM KIO_3 was titrated at a speed of 2.5µl per s to the starch solution. The titration terminal was automatically detected with a sensitivity setting of 3, and the used volume of KIO_3 was transformed into amylose content. Standard amylose solutions were prepared as checks by dissolving pure amylose and amylopectin in distilled water (Tan YF et al., 1999).

+Gelatinisation temperature

GT was determined using the alkali digestion test [Little RR et al 1958]. A duplicate set of six whole-milled kernels without cracks was selected and placed in a plastic box (5×5×2.5cm). 10mL of 1.7% (0.3035M) KOH solution was added. The samples were arranged to provide enough space between kernels to allow for spreading. The boxes were covered and incubated for 23h in a 30°C oven. The starchy

endosperm was rated visually based on a seven-point numerical spreading scale as a standard evaluation system for rice [IRRI .2013]. According to the ASV score. GT of rice grains can be classified into four groups: high (1–2), high-intermediate (3), intermediate (4–5), and low (6–7) [IRRI.2013].

+Gel consistency

Gel consistency was determined as previously described [IRRI.2013]. Rice flour (100mg) was mixed with ethyl alcohol (0.2mL) containing 0.025% thymol blue and 0.2M potassium hydroxide (2mL) and heated in a boiling water bath for 8 minutes. After heating, the sample tubes were allowed to cool in an ice-water bath and immediately laid horizontally on the table. Gel consistency was measured by the length of the cold rice paste in the culture tube held horizontally for one hour, Hard, medium, and soft gel standards such KhaoDawmali 105 are respectively included in every set.

Milling recovery

Brown rice samples of 100 g from each treatment plot were milled in a McGill-type miller no. 2 with the 685 g added weight on the pressure cover for 30 sec, followed by 30 sec without the added weight. Total milled rice weight was determined. Head rice yield was determined by sizing milled rice with a Satake testing rice grader TRG 05A using a 4.75-mm mesh indentation, weighing the brokens and whole grain fractions. Total and head milled rice yields were calculated as percent of rough rice. Head rice yield in kg/ha was calculated from rough rice yields determined at harvest of each experiment from a 5-m 2 area within each plot.

Data Analysis

Analysis of variance.

The agro-morphological data collected were initially analyzed through analysis of variance to verify genetic variation in the traits measured. The few traits with insignificant genetic variation, based on the F-test, were not considered for further analyses.

III. RESULT AND DISCUSSION

3.1. *Experimental soil properties*: The production of landrace rice grains is extremely important thanks to the structure of the soil. The soil must have a bright, light texture with good drainage system and moderately low amount of organic matter. The results of land analysis at Chau Phu locations showed that the maximum humidity reserves fluctuated from 40.8% to 41.0% for Chau Phu in order. Organic C content is not high (0.92% and 0.86%). This suggests that organic matter is not so high suitable for growing landrace rice because the soil is often porous, allowing root remove and lodging. Bright soil color reduces the color of the shell. Ensures the attractiveness of rice grains and catches the eye with the market. The soil drains well, providing air inside the soil for the root system to grow. The percentage of lightning particles is very low (1.2-2.02%) (Table 1). Mild - neutral soil (pHKCl 6.01-6.25).

Table 1: Some properties of the tested soil (0–30 cm depth) before sowing

Property	**Value assessment**	
	Before	**Ater Havested**
Component distribution (%)		
Sand	66,4	65,78
Silt	32,5	31,20
Clay	3,1	2,02
Soil texture	Sandy loam	Sandy loam
Saturation percent (S,P%)	41,0	40,8
pH (soil)	6,09	6,35
E,C (dS m^{-1}, at 25 °C)	0,42	0,55
Soil physical and chemical analysis		
Total N (%)	0,089	0,093
Available N (ppm)	28,0	47,0
P (ppm)	7,40	11,20
K (ppm)	137,6	148,5

Exchangeable cation (meq/L)		
Ca^{++}	0,90	0,98
Mg^{++}	0,38	0,29
Na^{+}	0,08	0,19
K^{+}	0,25	0,39
Organic carbon (ml/lit) -C (%)	0,92	0,86

pH_{KCl}	6,10	6,17
Các bon hữu cơ (OC - %)	0,98	1,28
N tổng số (%)	0,213	1,456
P_2O_5 tổng số (%)	0,74	0,98
K_2O total (%)	0,58	1,16
Lân dễ tiêu (mg P_2O_5/100 g đất)	5,36	7,22
Kali dễ tiêu (mg K_2O/100 g đất)	19,22	28,9
CEC (lđl/100 g đất	2,01	2,67
Mg (%)	1,09	1,02
Cát (%)	12,7	12,7
Limôn (%)	20,5	20,5
Sét (%)	2,6	2,6

3.2. Effects of fertilizers on plant height, filling and unfilling of HATRI 10 and HATRI 475 rice.

3.2.1. Analysis of the impact of fertilizers on the componts of yield composition of rice plants This analysis is based on factors: productivity and productivity composition in Chau Phu . An Giang .

Table 2. Effects of fertilizer on the development of HATRI 10 and HATRI 475

Factors (F)	HATRI 10			HATRI 475		
	Plant height (cm)	Filled grains / panicle (number)	% unfilling	Plant hight (cm)	Filled grains / panicle (number)	% unfilling
Chau Phu , An Giang						
F1= 80-40-40 + organic manures 10 t ha-1	102.7	112.5	15.2	108.2	128.3b	20.1
F2= 60-40-40 + organic manures 10 t ha-1	104.6	120.4	13.7	106.6	129.2a	15.6
F3= 40-40-40 + organic manures 10 t ha-1	112.2	118.3	18.2	103.6	119.4c	12.8
F4= 20-40-40 + organic manures 10 t ha-1	111.1	128.4	16.7	102.5	119.6d	22.4
F5= only organic manures 10 t ha-1	110.5	132.6	20.4	131.3	118.6e	20.1
F6= 0-0-0+ 0	108.3	61.5	25.7	127.5	103.7f	25.4

F7= Famers (120-40-60 + organic manures r 12 t ha-1	114.6	117.1	19.5	108.3	118.5b	16.5

3.2.2. Effects of fertilizers on yield for rice .

Treatments produced significantly different effect on all measured parameters: panicle length and grain yield, thousand grains weight at 5% level of significance . The panicle length of HATRI 10 fluctuates from 27.3-26.2cm. In the thousand grains weight there is no significance for the all treatments except not fertilization (F6) for lower weight inHATRI 10. For HATRI 475 the treatments for thousand grains weight not significantly different effect two varieties . In terms of recorded productivity like HATRI 10 for the highest productivity in the F2 test. This is recorded on HATRI 475 (F2) for a yield of 9.3tons / h.a). fertilizers application (60 -40-40 kg/ha+ organic manures 10 t ha-1) increased grain yield both HATRI 475 and HATRI 10 at Chau Phu .

Table 3. Effects of fertilizers on yield and yield components in rice

Factors (F)	HATRI 10			HATRI 475		
	panicle length (cm)	1000-grain weight (g)	yield(ton/ha)	panicle length (cm)	1000-grain weight (g)	yield(ton/ha)
F1= 80-40-40 + organic manures 10 t ha-1	27.5a	25.7a	9.2a	25.2a	24.5a	9.1a
F2= 60-40-40 + organic manures 10 t ha-1	27.3a	25.6a	9.7a	25.2a	24.5a	9.3a
F3= 40-40-40 + organic manures 10 t ha-1	27.3a	25.4a	9.2a	25.5a	24.9a	9.3a
F4= 20-40-40 + organic manures 10 t ha-1	27.3a	25.8a	9.6a	25.6a	24.9a	9.2a
F5= only organic manures 10 t ha-1	27.1a	25.6a	8.5b	25.6a	24.3a	8.9b
F6= 0-0-0+ 0	26.2b	25.4a	6.9c	25.6a	24.6a	6.5c
F7= Famers (120-40-60 + organic manures r 12 t ha-1	27.4a	25.7a	9.2a	25.5a	24.5a	8.2b

3.2.3. The effected of fertilizers on rice qualities (Cooking and eating properties) of landrace rice

Analyzing the amylose content of HATRI 10 varieties recorded fluctuations in fertilizer levels that have changed statistically. Amylose levels increased slightly when nitrogen levels were increased. In the full fertilizer treatment (F1) the average amylose (%) content is calculated for HATRI 10 (20.6). Similar to the experimental fertilization of F7 (amylose content is 21.7%). Other tests showed that lower amylose levels ranged from19.4% to 20.7%. For HATRI 475 varieties in the high amylose(22.7%) test is the F7 (fertilizer according to farmers). Thus, the amylose content has changed due to changes in the amount of fertilizer. Next Gel consistency (GC) also recorded fluctuations on both two varieties. In particular, in gelatinisation temperature (GT) there is no change in the tests for both varieties (table 4). This is also noted on the HATRI 475 . The GT popularity alone has not changed al of treatments.

Table 4. Effected of fertilizers on cooking and eating properties of improved rice

Factors (F)	HATRI 10			HATRI 475		
	Amylose content (%)	Gel consistency l(mm)	Gelatinisation temperature	Amylose content (%)	Gel consistency l(mm)	Gelatinisation temperature
F1= 80-40-40 + organic manures 10 t ha-1	20.6a	69.2a	3	22.3a	65.5c	3
F2= 60-40-40 + organic manures 10 t ha-1	20.8b	69.5a	3	22.5b	66.7b	3
F3= 40-40-40 + organic manures 10 t ha-1	20.7b	69.4a	3	22.2b	66.4b	3
F4= 20-40-40 + organic manures 10 t ha-1	20.6b	69.3a	3	22.6c	67.5a	3
F5= only organic manures 10 t ha-1	20.2b	69.2a	3	22.5c	67.2a	3
F6= 0-0-0+ 0	19.4c	69.5a	3	22.2c	66.2b	3
F7= Famers (120-40-60 + organic manures r 12 t ha-1	21.9a	69.2c	3	22.5a	65.6c	3

2.4. The effected of fertilizers on the milled qualities of rice

Analyzing the rate of milling on rice varieties with different levels of fertilizer recorded in terms of the ratio of head rice, brown rice and the ratio of white rice both varieties HATRI 10 and HATRI 475 on two points of statistical significance. Analysis of brown rice ratios showed that the F2 treatment gave a high percentage of head rice on both varieties (HATRI 10) and 53.5% on (HATRI 475). The treatment had the lowest percentage of head rice in the F7 (49.7%). The same of HATRI 475 (49.2%). HATRI 10 had much higher head rice at treatment F2 (60-40-40 + organic manures 10 t ha-1). (Table 5)

Table 5. Effect of fertilizers on milled rice content for rice

Factors (F)	HATRI 10			HATRI 475		
	Brown rice (%)	White rice (%)	Head rice (%)	Brown rice (%)	White rice (%)	Head rice (%)
F1= 80-40-40 + organic manures 10 t ha-1	81.5c	76.4d	50.8d	82.4a	76.4b	51.2b
F2= 60-40-40 + organic manures 10 t ha-1	82.6b	76.4a	55.7a	80.6c	77.1 a	53.5a
F3= 40-40-40 + organic manures 10 t ha-1	81.3c	76.5e	54.b	81.6b	75.1a	50.4c
F4= 20-40-40 + organic manures 10 t ha-1	84.4a	74.6c	53.5c	82.7a	76.5b	50.7c

F5= only organic manures 10 t ha-1	82.4b	72.5e	53.3c	81.6b	76.5b	50.6c
F6= 0-0-0+ 0	82.5b	73.4d	53.2c	80.3c	76.2b	50.5c
F7= Famers (120-40-60 + organic manures r 12 t ha-1	80.7d	75.2b	49.7e	81.2b	74.5c	49.2e

3.3. Discussion

The nitrogen -deficient tree will be elongated in Chau Phu experiments for both HATRI 10 and HATRI 475 varieties in the treatment F 6. The HATRI 475 variety had a higher plant height, compared to the HATRI 10 variety. This is in the same with Dobermann and Fairhust's (2000) comments.The application of nitrogen fertilizer can increase the height of the plant, the number of panicles.According to (Spargo et al., 2013). "the desired pH range of 6.0 to 7.0 of most crops but acidity reduces the availability of nitrogen. Phosphorus and potassium. P deficiency of this nutrient can lead to a decline in plant growth; weak root system, and seed quality, low yield. Phosphorus plays an important role in the development of roots, promoting early flowering and ripening and resistance to disease and drought. In table 2 with treatment 5 and 6 lack of phosphorus the plant height is the same with other treatment but the yield is very low table 3. Phosphorus deficiency can delay the maturation of rice crops and increase sensitivity to rice disease (Fageria et al., 2003). Potassium-deficient plants cannot use nitrogen and water more efficiently and are more susceptible to disease." Low to moderate soils require fairly reasonable management (Belachew and Abera. 2010). Furthermore. The proper application of potassium is closely related to the dependence of cell walls, bundles and growth intensity of the trunk, which enhances resistance to the tree against reclining beans (Kong et al., 2014). Rice plants that are deficient in potassium will often have high cases when the disease enters which can lead to the incidence of the disease. Therefore, this study was conducted to assess the effect of different levels of NPK fertilizer on the growth and productivity of landace rice varieties. Soil organic matter is the local biodegradation that affects soil structure and porosity. The rate of penetration of water, humidity, the diversity and biological activity of soil organisms and the availability of nutrients (Bot and Benitez 2005). Soil structure affects soil fertility and how air and water move through the soil (Macie. 2013). The results revealed no interaction effects of NPK fertilizers and rice varieties on the physiological characteristics, lodging incidence characteristics and yield component. There were significant different observed on the plant height, panicle number, percentage of filled grains and 1,000 grains weight of different rice varieties. Different levels of NPK exerted significant effect on yield and component yield such as 1,000 grains weight and grain yield. This can happen because compost has high nutrients. The tallest plant height is affected by a combination of compost and a fertilizer recommendation N. P. K but does not differ significantly when compared to other experiments except by combining compost and fertilizer n. P. K. at 80 N consciousness (F1). The increase in the height of landrace rice may be related to the full availability of water in the test area during the test period. However, Tri Ton and Tinh Bien are mainly based on heavenly water, so the disruption of water sources affects the development of rice crops. In this experiment at Chau Phu with HATRI 10 and HATRI 475 had a slight increase in amylose levels in the F7 test. Which was consistent with previous reports (El-Kadyet al.,1999) reporting that the application of nitrogen fertilizer slightly increased amylose content.

Fertilization depending on the rice variety with the level of 120 N / ha significantly increased the proportion of whole rice decreased in the F7. The yield is also reduced due to the landrace and the leaves are more likely to fall when applying high nitrogen fertilizer. These productivity trends also to explain that limping alone cannot serve to reach the maximum potential of acidic soils. Thus suggesting that depleted soils N and K. Which clearly affect crop performance as were observed when these modifications (fertilizer P) were applied in combination with manure (Farag and Zahran. 2014). Organic sources along with chemical fertilizers have improved the productivity and quality of improved varieties on the F1 test also recorded in Table 4 . Therefore, it can be inferted that potassium manure along with K released from straw, increases the availability of this nutrient in complexes and in soil solutions, allowing for better absorption of nutrients as evidenced by the nutritional status of the crop. In many physiological and metabolic processes, including photosynthesis, osmosis, nutrient transport, carbohydrate transport and storage, nitrogen absorption and protein and starch synthesis (Hawkesford et al.. 2012; Raza et al.. 2014).Given the importance of nitrogen fertilization for the yield in grains from rice crops, it is necessary to know the best dose for each variety as well as its effect on productivity components and other agrocological

parameters such as cycle yield, plant height and yield composition of the plant. Increasing the rate of nitrogen fertilizer can increase productivity but reduce particle quality on the F7 test. On the other hand, there are many factors that play a huge role in the quality of rice. The quality of cooked rice and its taste, which is important to consumers. The most important factor that can affect the quality of cooked rice is the amylose (AC) content, which is part of the starch. Other factors such as gel consistency (GC) and gelatinization temperature (GT). In general, the AC in rice grains will determine the softness and hardness of the grain after the cooking process. GC is the mucus ratio during cooking. In fact. GT is the water temperature of starch particles at an irreversible expansion (Zamani and Alizade. 2007). Dong et al. (2007) showed that nitrogen intake had a profound effect on the quality of cooking and the nutritious value of rice, with an increase in GC but a decrease in AC. Young Lee (2006) in this study concluded that there was a negative correlation between the amount of nitrogen and amylase in rice on Table 4 and the rate of milling quality also decreased table 5. Dong et al. (2007) showed that nitrogen in take had a profound effect on the quality of cooking and the nutritious value of rice, with an increase in GC but a decrease in AC. Young Lee (2006) in this study concluded that there was a negative correlation between nitrogen and amylase levels in grains.

IV. CONCLUSION

Integrated application of organic manures and inorganic fertilizers was effective for enhancing growth, yield, and the yield components of HATRI 10 and HATRI 475 . The increase in the rate and dosage of N. P and K from the F1 treatment has significantly increased plant height. Panicles per plant (number. 1000-grain weight (g). Filled grains / panicle). The interaction between genotypes and fertilization of nitrogen, phosphate and potassium had a significant effect on all agricultural and crop yield indicators at both test sites. The genotype of the HATRI 10 gives the value of the above indicators higher than that of the HATRI 475 . The increase/decrease in fertilizer intake has had a significant and statistically significant effect ($p \leq 0.05$) on the yield and quality of l rice grains in both all treatments the exception of 1000-grain weight. HATRI 10 and HATRI 475 received with F2 had significant different between NPK fertilizer applied.Therefore it can be recommended to farmers. The amount of fertilizer used in treatment F2 (60:40:40 kg ha-1+ organic manures 10 t ha-1) is the least. Thus, it gives an economical advantage as low fertilizer cost is required to achieve higher yield and better grain quality.

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Challenges and Solutions in D-Amino Acid Production Methods

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Abstract— *D-amino acids play a vital role in the pharmaceutical, food, and chemical industries for the manufacture of antibiotics, fertility drugs, sweeteners, and drugs against neural disorders. Examples include; they can be components in analgesics, antistress agents, antidiabetics (e.g., nateglinide), and anticoagulants. Therefore, industrial production of D amino acids is required, and owing to this, research has progressed while aiming towards improving yield and reducing the cost of production. However, to optimize large-scale production, challenges faced in currently used production methods must first be exposed for possible solutions. In this review, we provide a background of recent methods utilized, challenges in these methods, and solutions to enhance the production of D-amino acids.*

Keywords— ***D-amino acids, pharmaceutical industry, Industrial production, Production methods, Challenges and solutions***

I. INTRODUCTION

Amino acids except Glycine have a chiral center (Alpha Carbon) onto which is attached an amine group, carboxyl group, R group, and hydrogen atom. The arrangement of these four groups around the chiral center forms two enantiomers; L-amino acids and d-amino acids. L-amino acids are the most dominant and functionally common enantiomer and these universally provide building blocks for proteins made in all living organisms. Recently, Researchers have gained interest in D-amino acids. Among these, D-Alanine, and D-Glutamic acid have been identified for years as fundamental blocks in microbial physiology where they are key constituents of the peptidoglycan. (Hancock, 1960) The existence of D-amino acids in the cell wall of bacteria (Peptidoglycan) offers a protective role against destruction from some proteases since D-enantiomers would go unrecognized by the enzymes. (unnatural forms)

Bacteria, such as Vibrio Cholera and Bacillus Subtilis secrete D-amino acids other than D-Glutamate and D-Alanine; these are collectively called Non-Canonical D amino acids (NCDAAs) as they are not involved in peptidoglycan layer synthesis although some have been found to regulate its structure.

It was found by Lam et al (2009) that stationary phase supernatant fractions of wild-type *V. cholerae* consisted of four amino acids: Met, Leu, Val, and Ile D-forms, and not L-forms, of these amino acids. This shows their role in bacterial stress adaption and competing against non-D-amino acid-producing bacteria. It has also been researched that D-amino acids not only take part in the peptidoglycan layer synthesis but also regulate spore germination and biofilm dispersal in certain species. (Bucher et al., 2015)

D-Amino acids are produced following a reversible stereo-inversion of groups around the chiral carbon and such a reaction is catalyzed by a group of enzymes called racemases. Two types exist in bacteria; highly specific and

broad-spectrum racemases (Bsr). (Hernández and Cava, 2016) Bsr can catalyze racemization reactions of a wide range of amino acids although their activities may be lower than the specific racemases. It is interesting to note that most gram-negative bacteria occupying a wide range of environments contain Bsr such as soil, marine water, or animals. (Espaillat et al., 2014) This suggests a possible role of racemases in colonizing hush ecosystems by rendering bacteria the ability to utilize D-amino acids as Carbon and Nitrogen sources present in such environments. (Kubota et al., 2016)

II. APPLICATIONS OF D-AMINO ACIDS

The use of D-amino acids in antimicrobial agents is an obvious role owing to their ability to suppress bacterial growth and biofilm dispersion. Li et al (2016) found out that optimized concentration of a mixture of D-methionine, D-tyrosine, D-leucine, and D-tryptophan greatly enhanced THPS biocide treatment of two recalcitrant biofilm consortia containing sulfate-reducing bacteria (SRB), nitrate-reducing bacteria (NRB), and fermentative bacteria. In the latter case, it is noted that the D-amino acids were utilized as Biocide enhancers hence reducing bacterial biocide resistances.

D-amino acids also find significant roles in neural system health, where low or high levels would trigger disease; therefore, correcting such levels within human bodies has been seen as a potential therapy. Several reports have come out about the role of particular D-amino acids in improving emotion, memory, and cognitive ability, and as medication in neural conditions such as Parkinson's disease. An example of this is low concentrations of D-Serine (25% decrease compared to normal) in cerebrospinal fluid and significant reduction of serine racemase in the hippocampus and frontal cortex of schizophrenia patients. (Bendikov et al., 2007)

Tsai et al (1998) studying the treatment of schizophrenia found out that the symptoms improved through orally administrating D-Ser 30 mg/kg daily. At the end of the 6-week trial, a 17% reduction had occurred in the positive symptoms and a 21% reduction had occurred in the negative symptoms of the disease. D-Serine is a strong endogenous co-agonist of N-methyl-d-aspartate receptors present in the hypothalamic structure, and schizophrenia arises following hypofunction of NMDAR evident with a reduction in D-Serine concentration. It should also be noted that stimulation of NMDAR by D-amino acids (D-serine and D-Alanine) plays a role in the analgesic effect and protection of the nervous system. (Shi et al., 2022)

It has been reported by Hartman et al., (2015) that D leucine has a positive effect on seizures by attenuation of the long-term potentiation (LTP) in the hippocampus. This gives D-Leucine an advantage over other medications which completely inhibits LTP and this may impair learning and memory. (Hartman et al, 2015)

D-amino acids also find possible application in the food industry as sweeteners particularly D-valine, D-phenylalanine, and D-tryptophan owing to having a sweeter taste and flavor compared to the L-enantiomers. Of particular interest is Alitame, an artificial dipeptide sweetener containing L-aspartate and D-Alanine having over 10 times sweetening power when compared to aspartame and six times when compared to saccharin. (Chattopahyay et al., 2014)

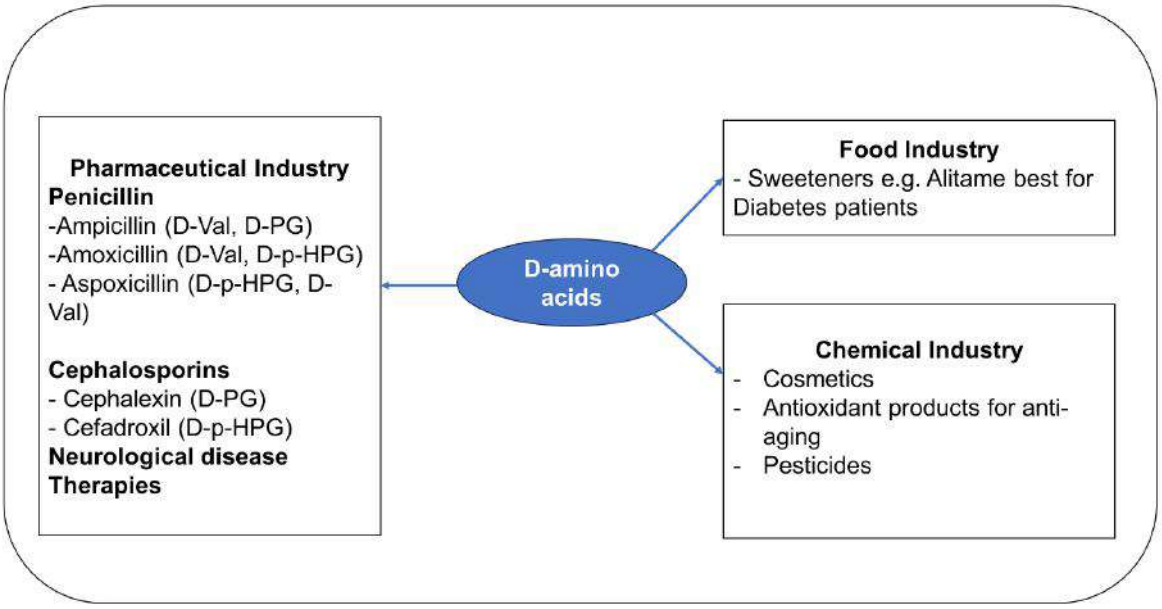

Fig 1: Examples of fields in which synthesis of D-amino acids is of great importance. In antibiotics, illustrated D-amino acids function as a core in the chemical structure.

III. BIOCATALYTIC METHODS FOR PRODUCTION OF D-AMINO ACIDS AND CHALLENGES INVOLVED

Several enzymes are utilized in biotransformation to form D-amino acids from a variety of substrates; D, L amino acid mixtures, N-acyl D, L amino acids, D, L-hydantoin, D, L-amides, α–keto acids and L-amino acids. Among these, L-amino acid substrates are the cheapest sources since they can be readily available from fermentation processes of engineered strains starting from cheap carbon and nitrogen sources. Biocatalysis can be both in-vivo; where whole cells are engineered to synthesize and secrete D-amino acids, or in-vitro where enzymes are extracted, purified, and modified to catalyze reactions in vessels, solutions, or surfaces.

1.) D-amino acids production from Alpha keto acids

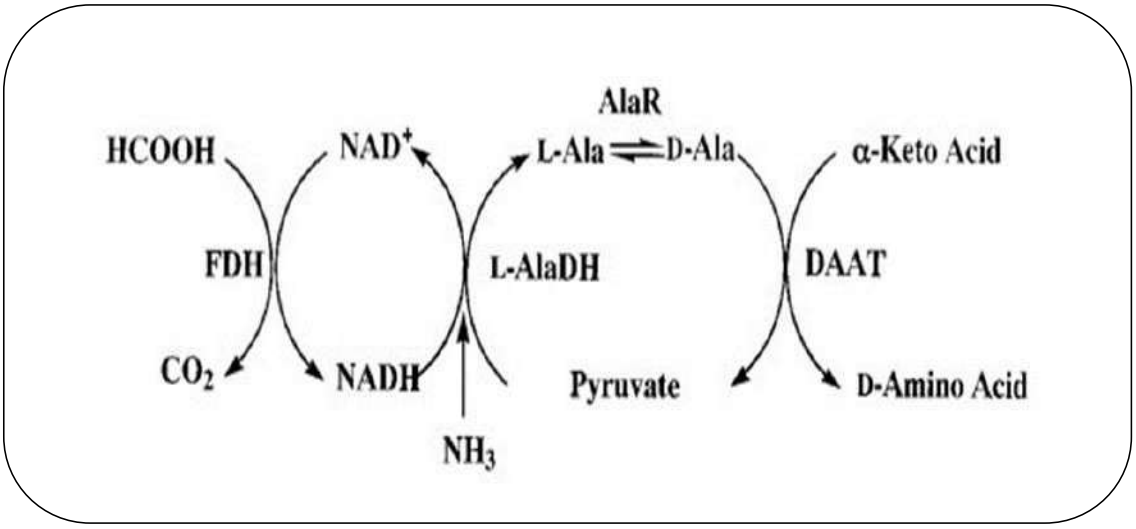

Fig 2: (Galkin et al., 1997) illustration of one of the earliest proposed methods for the production of D-amino acids particularly from corresponding alpha-keto acids.

Fig 1 demonstrates a scheme composed of Formate dehydrogenase (FDH) that generates NADH, L-Alanine Dehydrogenase which incorporates ammonia to pyruvate to generate L-Alanine, Alanine Racemase (AlaR) that catalyzes interconversion between L and D-Alanine, and lastly, D-amino acid aminotransferase (DAAT) that catalyzes transfer of amino group from D-Alanine to an alpha-keto acid to form corresponding D-amino acids. L-Alanine dehydrogenase (L-AlaDH) functions to regenerate L-Alanine from pyruvate by incorporation of ammonia. Galkin et al (1997) reported the production of D enantiomers of glutamate and leucine at high optical purities and high conversion rates obtaining a concentration of about 44g/l D-glutamate following heterologous expression of FDH, AlaDH, DAAT, and AlaR genes in E. Coli.

However, the main problem faced while using the strategy was the production of D-amino acids as racemic mixtures probably as a result of catalysis by L-amino acid aminotransferases to form L amino acids from the provided alpha-keto acids. Economically, alpha-keto acids are also expensive, limiting the application for large-scale profit production.

Such a racemic result problem could be solved by using the L-amino acid oxidase enzyme which oxidizes only L-amino acids leaving behind D-enantiomers for further purification. However, it has been investigated that L-amino acid oxidase generates hydrogen peroxide which limits the enzyme activity. Sang et al (2022) utilized Aromatic Amino acid decarboxylase (AADC) from Bacillus atrophaeus as an alternative to L-amino acid oxidase for the purification of aromatic D amino acids from racemic mixtures. This enzyme offered an advantage owing to catalytic decarboxylation of a variety of aromatic L-amino acids to respective mono-amines without the generation of Hydrogen peroxide. Results from enantioselectivity studies of AADC also showed that the enzyme offers efficient kinetic resolution without undesirable loss of D-amino acid.

2.) Production via expression of hydantoin racemase, d-hydantoinase and *N*-carbamoyl-d-amino acid amidohydrolase in E-coli (In vivo Hydantoinase process)

Hiroyuki et al (2005) utilized a system for enzymatic production of D-amino acids in whole cells by co-expression of three genes encoding for D-specific hydantoinase from Microbacterium liquefaciens AJ3912 (DHHase), *N*-carbamoyl-D-amino acid amidohydrolase (DCHase), and hydantoin racemase (HRase) from Flavobacterium sp. AJ11199 in *E. Coli*. The system works

in such a way that substituted 5-monosubstituted L-Hydantoin molecules corresponding to a particular amino acid are converted first to 5-monosubstituted D-hydantoin molecules, which are then linearized by hydrolysis catalyzed by DHHase forming an N-carbamoyl-D-amino acid derivative. The latter molecule is then hydrolyzed by DCHase forming the corresponding D-amino acid. Using this platform, D-Phe, D-Tyr, *O*-benzyl-D-Ser, D-Leu, D-norvaline, and D-norleucine proceeded efficiently, achieving a 98% molar yield after 48 h and an optical purity of more than 99% e.e. (Hiroyuki et al., 2005)

The whole-cell biocatalyst's advantage was the efficient production of a wide range of D-amino acids from corresponding 5-monosubstituted-L-hydantoins owing to the broad specificity of the three enzymes used. However, the challenge was the need for the production and purification of the substrate (Substituted Hydantoin derivatives) which could be expensive for commercial production of the D-amino acids. Also, the production of the substrate requires potassium cyanate which is a toxic chemical compound. (Suzuki et al., 2005)

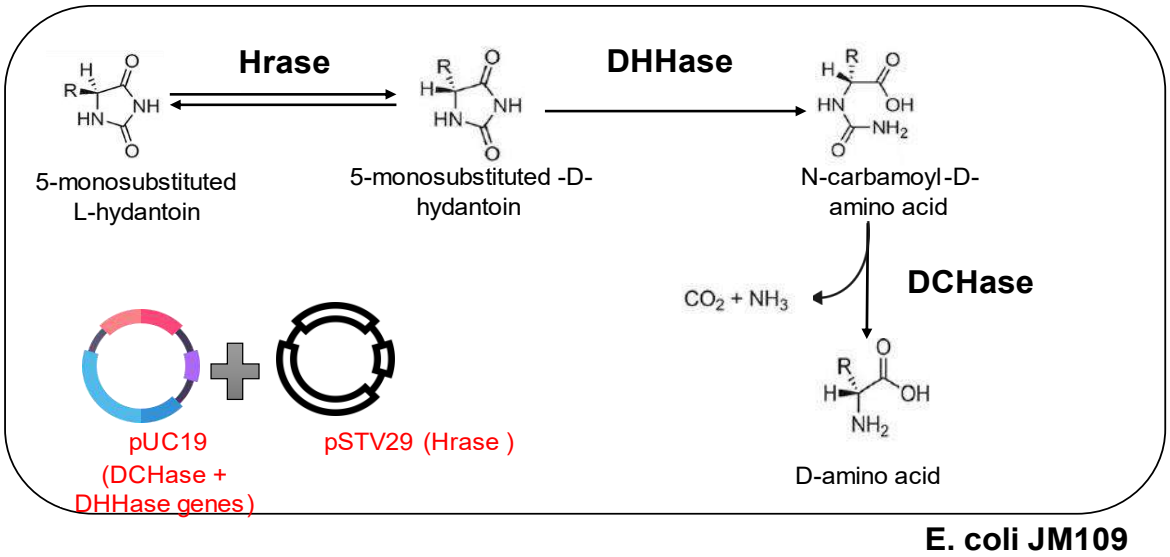

Fig 3: illustration of biotransformation platform utilized by Hiroyuki et al (2005); enzymes DCHase, and DHHase were expressed via pUC19 plasmid, while pSTV29 plasmid was used to express Hrase enzyme both utilizing trp promoter. The co-expression was carried in E. coli JM109 cells.out

In-vitro Hydantoinase process for production of D-amino acids

Similar to the whole cell (in-vivo) hydantoinase process employed by Hiroyuki et al (2005) optically pure D-amino acids can also be produced by hydrolysis of their respective L-5-monosubstituted hydantoins in a similar classical three-enzyme cascade reaction, (dynamic kinetic resolution cascade process) carried out in-vitro as illustrated in figure 2. (Yafei et al., 2017) This method offers no need for intermediate removal and purification since it offers 100% theoretical conversion. (Yamaguchi et al., 2007) Although theoretically strong, 100% bioconversion cannot be achieved owing to the thermal instability of enzymes mainly D-Carbamoylase, Insolubility of enzymes, and sparingly soluble Hydantoins owing to these challenges, several research efforts towards searching for highly compatible and thermostable enzymes have been ongoing. Yafei et al (2017) identified a d-carbamoylase from Arthrobacter *crystallopoietes* (*Ac*HyuC) through genome mining and screening of Carbamoylases with pH compatibility, high activity, enantioselectivity, and solubility to establish a DKR cascade for efficient production of d-Trptophan from l-indolylmethylhydantoin. With this enzyme used in the cascade, 80 mM L-indoylmethylhydantion could be completely converted into d-tryptophan within 12 h in a 0.5-L system, reaching a yield of 99% and productivity of 36.6 g L^{-1} d^{-1}.

Chemical methods combined with biocatalysis have been widely used for the synthesis of D-amino acids; however, such methods are so expensive requiring high starting material, low specificity, and high need for purification from toxic chemicals. To solve this problem, microbial production of D-amino acids is seen as a potential solution through stereoisomeric conversion of L-amino acids to D-amino acids. L-amino acids are mostly generated by fermentation from inexpensive and renewable natural sources, therefore providing a cheaper way.

3.) Preparation of D-amino acids from DL mixtures via Selective L-amino acid Degradation

Recent studies have shown that microorganisms use mainly L-amino acids although they may also have the ability to utilize D-amino acids as Carbon and Nitrogen sources. Some strains showed asymmetric degrading activity against DL-amino acids and degraded only the L-enantiomer. (Takahashi et al., 1997) Zhang et al (2015) isolated and characterized Candida maltose DLPU-zpb which could degrade L-valine effectively but not D-valine, and based on this, a preparation method for D-valine was suggested from DL-valine mixture using the yeast strain, Candida maltose DLPU-zpb as a biocatalyst.

Zhang et al (2015) noted that DL-valine is produced commercially at a low cost via racemization of L-valine and therefore suggested that Candida maltose DLPU-zpb biocatalyst provides a cheap option for industrial production of D-valine.

However, we noted that although the L-isomer was completely degraded within 72 h under the conditions of 30°C and at pH 6, the final yield was dependent on the D-amino acid concentration in DL-amino acid mixtures. Therefore, this method only purifies but does not change the initial concentration of D-amino acids as produced by upstream racemization procedures. L-enantiomers' degradation leads to a high loss of resources since several methodologies utilize L-amino acids as starting raw materials for D amino acid synthesis. Therefore, we suggest that such a method requires a recycling process instead of only degradation to minimize the loss of useful L-enantiomers.

To prevent wasteful loss of L-amino acids, acetylation of the DL-amino acid mixture can be utilized together with *N*-acyl-D-amino acid amidohydrolase which selectively removes the acetyl group from only N-acyl-D-amino acids to form a corresponding D-amino acid. (Wakayama and Moriguchi, 2001) The un-converted N-acyl-L-amino acid can then be racemized to N-acyl-DL-amino acid for continued reaction.

4.) Biosynthesis via in-vitro dynamic kinetic resolution of N-succinyl amino acids

D-Phenyl alanine, D-Tryptophan, and D-Valine can be easily synthesized from corresponding N-Succinyl-L-amino acids or racemic mixtures of N-Succinyl-DL-amino acids via in-vitro catalysis by purified N-Succinyl amino acid racemase (NSAR) and D-Succinylase (DSA) (Sumida et al., 2018) In the suggested one-pot dynamic kinetic resolution, NSAR catalyzes the interconversion between D and L forms of N-Succinyl amino acids while DSA catalyzes the removal of Succinyl group from N-succinyl-D-amino acids to form an unconjugated D-amino acid form. Using this strategy, Sumida et al (2018) were able to convert 100 mM *N*-succinyl-DL-tryptophan, N-Succinyl-DL Phenylalanine, and N-Succinyl-DL-Valine to corresponding D-amino acids at 81.8% yield with 94.7% ee under optimized concentrations of DSA and NSAR

However, it is reported that the chiral purity of the D- amino acids is dependent on the ratios of DSA and NSAR, and adjusting such a concentration ratio is not feasible for industrial applications. Therefore, despite the promising industrial application, genetic engineering of DSA is required to improve the optical purity of the D-amino acids by minimizing side chain reactions with N-Succinyl-L-amino acids that would lead to the formation of L amino acids.

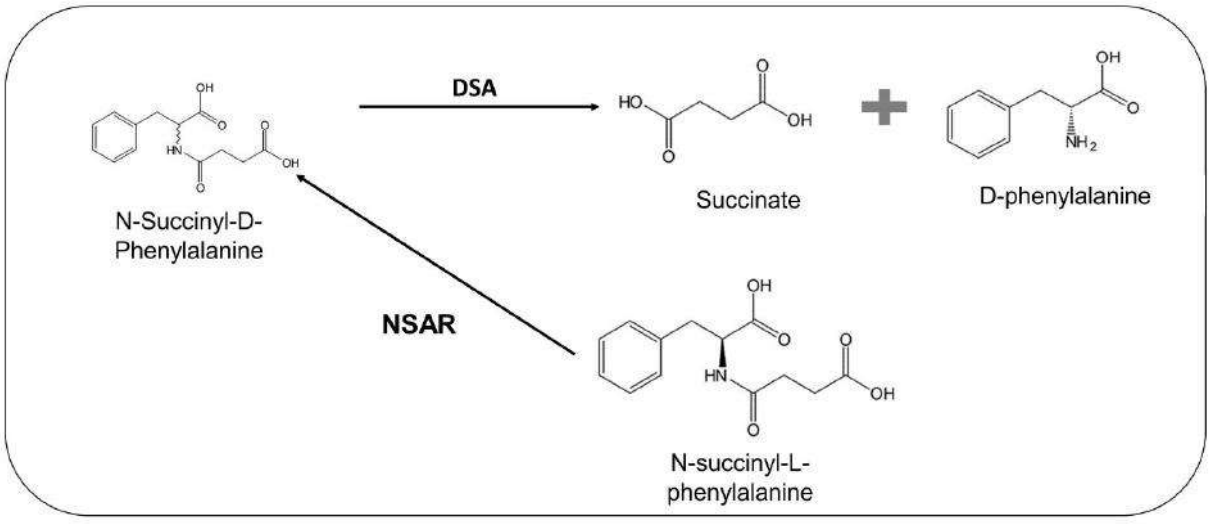

Fig 4: illustration of bioconversion of N-succinyl-D-phenylalanine to D-phenylalanine by DSA enzyme. NSAR functions to generate the D-enantiomer from N-succinyl-L-phenylalanine for further formation of D-phenylalanine.

5.) D-Tryptophan synthesis via engineered D-amino acid aminotransferase in a one-pot biocatalytic system

It has been investigated that complete stereo inversion of the L-Phenyl alanine could be achieved by combining oxidative deamination of L-amino acids using Proteus mirabilis LAAD (PmirLAAD) with transamination of D-amino acids using an engineered DAAT from Bacillus sp. YM-1 (DAAT-T242G) (Walton et al., 2018) Using this background, Parmeggiani et al (2019) combined mutations; V33G, S240G, and T242G with the idea that these will further increase the size of the binding pocket of DAAT. Out of the three combinations, DAAT-V33G/T242G) was the most active variant showing a 35-fold improvement to kcat/KM against D-tryptophan compared with the wild-type towards stereo inversion of L-Tryptophan to D-Tryptophan.

To further expand such an improved activity, a one-pot biocatalytic system utilizing Salmonella Tryptophan synthase (TrpS), LAAD from Proteus myxofaciens, and the engineered DAAT-V33G/T242G was developed in which D-Tryptophan was synthesized from indoles. With optimized substrate and reaction conditions, around 5g/l of D-Tryptophan was obtained with a percentage yield of 66%

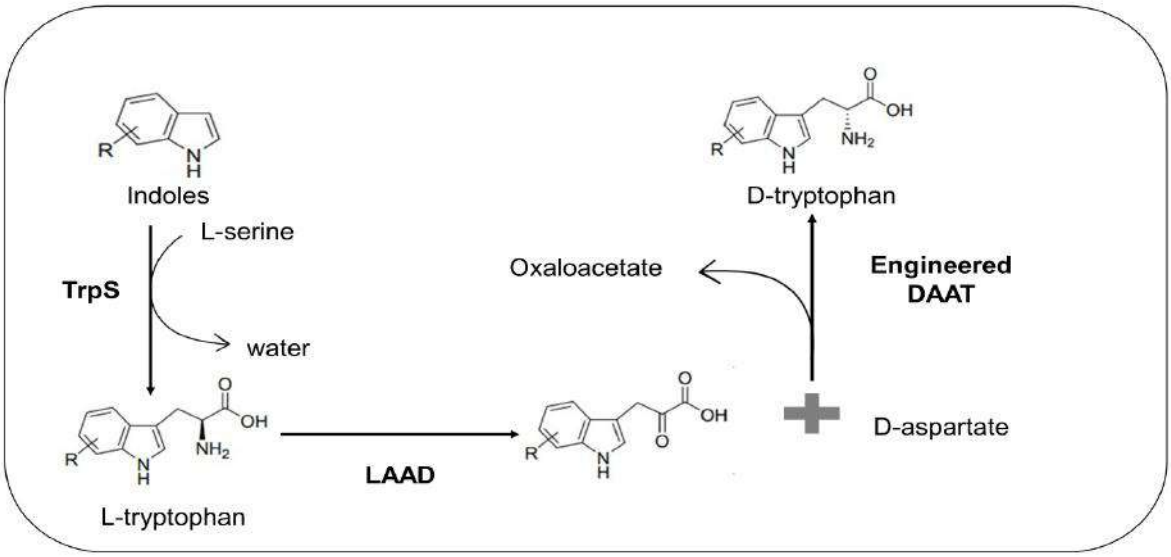

Fig 5: illustration of D-Tryptophan synthesis from indoles. It should be noted that R in the illustration represents Hydrogen atom in the case of Tryptophan. However more substituted D-tryptophans can be synthesized using the platform and for this case, R can be Fluoride, Chloride, Methyl, or Methoxy groups.

There are a few challenges with the one-pot biosynthesis of D Tryptophan from indoles; the DAAT enzyme requires a D-amino acid as an amino donor, the best substrate being D-glutamate which is expensive. Although the enzyme can also utilize D-Aspartate which is cheaper, a lower activity was reported as compared to using D-glutamate. This problem has been solved by the utilization of *meso*-diaminopimelate dehydrogenases which catalyze reductive amination of 2-keto acids such as pyruvic acid to generate d-amino acids in up to 99% conversion and 99% enantiomeric excess using inorganic ammonium ions. (Zhang et al., 2019; Gao et al., 2012) The second problem noted in this platform that may contribute to a reduction in yield is the utilization of separate enzyme systems; LAAD and DAAT are expressed in separate E. coli BL21(DE3), and TrpS is expressed in vitro. This means the conversion rate will be dependent and limited by the translocation of reaction intermediates between the media and the cells. The third challenge is that the production of L-Tryptophan intermediate from indoles is dependent on the presence of L-Seine in the reaction media, which therefore has to be provided. Scaling up such a platform to an industrial level would be economically challenging owing to the need for both L-Serine and D-Aspartate as mixture components.

6.) One Pot Biocatalytic stereo inversion cascade (In-vivo and in-vitro biocatalytic components)

Zhang et al (2019) utilized recombinant Escherichia *coli* BL21(DE3) cells for expression of L-Amino acid deaminase from *Proteus mirabilis* (PmLAAD) and purified *meso*-diaminopimelate dehydrogenases (DAPDHs) in a cascade route for the synthesis of D-amino acids from L-amino acids. This model was used to efficiently catalyze the transformation of L-phenylalanine into D-phenylalanine.

Studies showed that pmLAAD enzyme is membrane-bound and so a whole-cell biocatalyst with intact membranes

produces higher reaction than when utilized within in-vitro reactions. (Hou et al., 2016) Through optimization procedures, pmLAAD (whole cell biocatalyst) was found to have no activity on D-phenylalanine and Phenylphyruvic acid, however, with the ability to convert L-phenylalanine to Phenylpyruvic acid PPA at a yield of 58.7% 100 mM of substrate. Another advantage of pmLAAD enzyme is the ability to catalyze deamination without the generation of Hydrogen Peroxide, which would be toxic to the cells.

Screening results from various DAPDH enzymes showed StDAPDH/H227V mutant to have the highest activity towards the conversion of Phenylpyruvic acid to over 99% optically pure D-Phenylalanine, with no activity on D and L phenylalanine which means that the direction of reaction (equilibrium) is towards the formation of D-Phenylpyruvate. Since DAPDH/H227V is dependent on NADPH, *Burkholderia stabilis* formate dehydrogenase (BsFDH) produced and purified from recombinant E. coli was included in the cascade to generate the required co-factor. PmLAAD whole-cell biocatalyst (100 mg mL^{-1}), StDAPDH/H227V (4 mg mL^{-1}), and BsFDH (0.35 mg mL^{-1}, 1 U) were assembled to construct the one-pot stereo inverting cascade. Subsequently, the D-Phe was obtained in a yield of 76.2% by the addition of 30 mM L-Phe, 90 mM NH_4Cl, 60 mM sodium formate, and 3 mM $NADP^+$ to the 50 mM Tris–HCl (pH 9.0) reaction buffer during the cascade reaction at 45 °C (Zhang et al., 2019)

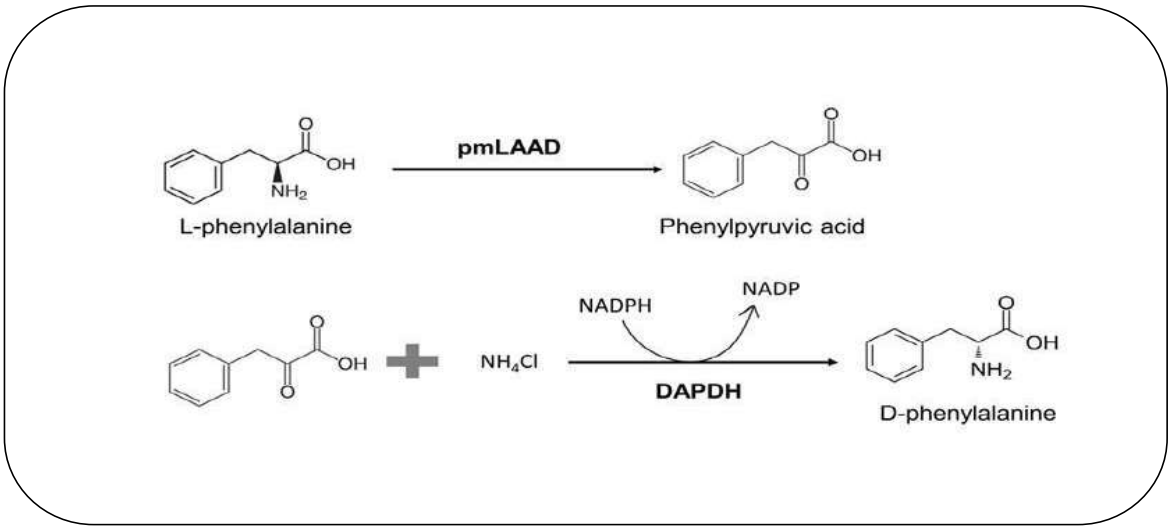

Fig 6: illustration of a two-step stereo inversion proposed by Zhang et al., NADPH is regenerated from NADP by BsFDH enzyme in the presence of Formate ions.

The biocatalytic stereo-inversion cascade also showed biotransformation efficiencies of 70%, 100%, 75%, 45%, 100%, 100%, 100%, and 100% for L-leucine, L-Glutamic acid, L-Norvaline, L-Tyrosine, L-Phenylalanine, L-Homophenylalanine, 2-Chloro-L-phenylalanine, 3-Chloro-L-phenylalanine and 4-Chloro-L-phenylalanine showing a reasonable versatility. Possible challenges with the cascade include the need for purification steps of BsFDH and StDAPDH/H227V enzymes and proper optimization of the reaction conditions, considering all three biocatalysts.

Also, the Phenylpyruvic acid intermediate from pmLAAD needs to be transferred through the cell membrane for the connection between the two necessary steps of the stereo-inversion reaction, and this would affect the conversion efficiency of the entire biocatalytic system. This challenge has been solved by incorporation of the three enzymes in a single cell for whole-cell biocatalysis as described by Zhang et al (2021)

7.) Whole-cell in-vivo Stereoinversion (pmLAAD, DAPDH, and BsFDH co-expression in E. coli)

To modify the previous one-pot stereo inversion, Zhang et al (2021) developed an in vivo cascade cell factory by co-expression of the three enzymes; L-amino acid deaminase from *Proteus mirabilis* (LAAD), *meso*-diaminopimelate dehydrogenase from *Symbiobacterium thermophilum* (DAPDH), *and* formate dehydrogenase from *Burkholderia stabilis* (fdh) into *E. Coli*. By using the obtained *E. coli* pET-21b-MBP-*laad*/pET-28a-*dapdh-fdh* whole-cell biocatalyst under optimized conditions, L-Phenyl alanine was stereo-inverted to D-Phenyl alanine with high conversion efficiency and optical purity.

Appling the whole cell (*E. coli* pET-21b-MBP-*laad*/pET-28a-*dapdh-fdh*) biocatalyst for the transformation of a variety of aliphatic L-amino acids as substrates, only L-Leu, L-norvaline, L-Glu, and L-Met were transformed to

corresponding D-amino acids obtained in quantitative conversion. (Zhang et al., 2021)

However, it has been reported in recent studies by Yanqi et al (2023) that high concentrations of some D-Amino acids inhibit bacterial growth. This was significantly observed with D-arginine, D-Leucine, D-methionine, D-valine, D-Cysteine, and D-glutamate. Therefore, this approach is likely to be limited to the production of low concentrations as the whole-cell biocatalyst would seem to be inhibited at high D-amino acid concentrations. Since Zhang et al (2023) reported high conversion efficiency, the yield can be improved by utilization of the acetylation strategy described in the next section as reported by Yanqi et al (2023)

IV. CURRENT STRATEGIES UTILISED FOR ENHANCED D-AMINO ACID PRODUCTION

a) Reduction of toxicity via D-amino acid production in E-Coli as Acetylated forms

For microbial cells to be used successfully as D-amino acids production factories, the toxicity of D-amino acids towards the host cells has to be overcome. Yanqi et al (2023) used an acetylation strategy by expressing both a racemase and D amino acid N-acetyltransferase enzymes into E-coli. The use of acetyltransferase enzyme presented the importance of driving the reaction equilibrium between L and D amino acids towards the formation of the D type and also rendering D-amino acids unreactive hence reducing toxicity towards the development of the host cells.

To demonstrate the role of acetylation of D-amino in the reduction of toxicity to cells, Yanqi et al (2023) cultured BW25113 (wild type, WT) E-Coli cells in an M9 liquid medium containing nineteen natural D-amino acids at 10 g/L, and results showed significant growth inhibition from 15 D-Amino acids, however, it is D-Asp, D-Glu, D-Pro, and D-His, did not cause growth defects at this concentration. When a similar procedure was repeated with acetylated forms of D-Arginine, D-Leucine, D-methionine, D-phenylalanine, D-Valine, and D-Serine included in the growth medium, there was a general increase in OD at 600nm showing that acetylation of D-amino acids offers a protective role against D-amino acid toxicity.

It was found that Yeast cells can continue growth even in the presence of D-amino acids. Yow et al (2006) found out that the enzyme; D-amino acid-*N*-acetyltransferase (DNT) in Yeast was essential in the detoxification of D-amino acids via acetylation. To reach this conclusion, results showed that a yeast mutant with the DNT gene (HPA3) knocked out was far less tolerant to D-amino acids as compared to the wild type, while over-expression of the DNT gene via p426Gal1 plasmid reversed the first result with over 100 times activity of DNT as compared to the wild type. It was also found that the production of N-acetyl D-amino acids was intracellular and then secreted to the medium. This suggests that Yeast cells are incapable of utilizing the acetylated form of D-amino acids as a nitrogen source. (Yow et al., 2006)

It is on this basis that Yanqi et al (2023) selected the heterologous expression of HPA3 from S. cerevisiae into E-Coli strain BW25113 for the production of D-amino acid transferase enzyme, an enzyme reported for the selective acetylation of a wide range of D-amino acids in vitro. (Yow et al., 2004) First, an HPA3 bioconversion assay was performed on nineteen D-amino acids to test its efficiency in E. coli. Plasmid pZElac-*hpa3* was introduced into strain BW25113 to create strain RN1 along with a negative control which were later cultured with 10g/l D-amino acids. Results showed bioconversion to acetylated D forms for 12 amino acids; Ala, Asp, Cys, Glu, Gln, Ile, Leu, Met, Phe, Ser, Thr, and Val with Ser, Glu, and Phe having the highest bioconversion yields.

A variety of Racemases are available for inter-conversion between L and D-amino acids. In E-Coli, two Ala racemases (*alr* and *dadX*), one Glu racemase (*murI*), and an Asp/Glu racemase (*ygeA*) are present and among these, YGEA has broad-spectrum catalytic activity with the ability to produce non-canonical D-amino acids other than D-Ala and D-Glu. A broad specific racemase also exists in Bacillus Subtilis encoded by the RacX gene and has a similar function to ygeA both having low catalytic activity. (Miyamoto et al., 2017) The isoleucine 2-epimerase (ILEP) from *Lactobacillus buchneri* has been previously characterized as a pyridoxal 5′-phosphate (PLP)-dependent racemase and can epimerize nonpolar L-amino acids into their D-forms (Mutaguchi et al., 2013, 2018). Additionally, the amino acid racemase BSRV from *V. cholerae* enables the bacteria to synthesize noncanonical D-amino acids (Espaillat et al., 2014). To test the efficiency of the broad specific racemases, Yanqi et al (2023) transformed Plasmids pZElac-*hpa3-alr*, pZElac-*hpa3-ygeA*, pZElac-*hpa3-bsrV*, and pZElac-*hpa3*-ILEP into strain BW25113 and results showed ILEP transformants had the ability of racemization of a wide range of amino acids; L-Ala, L-Cys, L-Gln, L-Ile, L-Leu, L-Met, L-Phe, L-Ser, L-Thr, L-Val, L-Phenylglycine, L-Norleucine, L-2-Aminobutyrate giving rise to a considerable measurable (g/L) of corresponding D-Acetyl-D-amino acids

From these results, it can be noted that some L-amino acids such as Try, Arg, and Lys are poor substrates for the ILEP enzyme. Therefore, screening of other Racemases for increased activity and directed evolution should solve this problem. Also, despite that N-acetyl transferase expression

saves E-Coli cells from D-amino acid toxicity, over-expression of the enzyme causes growth inhibition owing to acetylation effects of other proteins that could be essential in cellular activities.

b) Screening of Strains that tolerate D-amino acid toxicity

D-amino acids are toxic to E-coli cells as reported by Yanqi et al (2023) and it is on this basis that screening of other bacterial strains would improve their production without the need for acetylation strategies. It has been observed that toxicity depends on the form of D-amino acid, and the host strain. An example of this is that although the majority of D-amino acids inhibit the growth of E-coli, D-His, D-Asp, D-Glu, and D-Pro do not affect growth, while D-Leu, D-Arg, D-Met, D-Val, and D-Cys cause the highest growth inhibition. D-Ala, D-Asn, D-Ile, D-Lys, D-Phy, D-Ser, D-Thr, and D-Trp moderately inhibit growth in E-coli.

However, it was found by Stäbler et al (2011) that Corynebacterium glutamicum tolerates D-Thr, D-Arg, D-Lys, D-Ser, and D-Ala although D-Asn and D-Met inhibited the rate of growth in all cases, maximal growth (at OD 600) was attained. This was different from results observed with E-coli as maximal growth could not be attained in the presence of D-Ser, D-Val, D-Met, D-Phe, D-Leu, and D-Arg.

Comparing the susceptibility effects of E-coli and Corynebacterium glutamicum shows that the latter could tolerate the accumulation of D amino acids. It is expected that screening will unveil the existence of more strains with the ability to tolerate toxicity associated with the accumulation of D-amino acids and hence their possible use as hosts in D-amino acid production.

c) Enhanced transfer rates of substrates across the cell wall

Some precursors such as Hydantoins utilized in the synthesis of D-amino acids have low solubilities which thereby reduces biotransformation efficiencies following an increased mass transfer resistance across cell walls. Overexpression of D-carboxypeptidases A and B is reported to have a disruption on the peptidoglycan structure without affecting growth. This strategy was utilized by Yang et al (2019) by co-expression of the D-hydantoinase (Hase), N-carbamoyl D-amino acid amidohydrolase (Case), and D-carboxypeptidases A and B. Optimisation of such a system resulted in the production of 23.4 g/L of D -p - hydroxyphenyl glycine (D-p-HPG) in 32 h from DL p-Hydroxyphenyl hydantoin.

d) Enzyme engineering for improved catalytic properties

A number of enzymes involved in production of D-amino acids have been engineered for improved thermostability, broadened substrate specificity, improved catalytic activity, changing of the reaction site, and few reports on enantioselectivity. Here we have discussed a few engineering strategies identified in recent research aimed at improving D amino acid production.

i) Engineering of N-carbamoyl D-amino acid amidohydrolase enzyme for improved thermostability

Although Yang et al (2019) reported the production of 23.4 g/L of D-p-Hydroxyphenylglycine (D-p-HPG) in 32 h with 100% conversion, a low space-time yield (STY) of 0.7 g/(L·h) was obtained probably because of the low thermostability of the Case enzyme. Zhang et al (2024) utilized salt bridge engineering to improve AkDCase (Agrobacterium sp. strain KNK712) thermostability and to demonstrate this, a complex crystal structure of AkDCase with N-carbamoyl-D-p-hydroxyphenylglycine (Cp-HPG) was constructed in which 87 salt bridges were found. Out of these bridges, three were not conserved; D30-K34, E87-K84, and E135-K134. Following the identification of these three non-conserved salt bridges, D30, E87, and E135 were replaced by neutral (A) or basic residues (K/R) via virtual mutation. Studies of these mutants revealed the D30A and E87R mutation as the best variants showing the highest thermal stability. Of the two, D30A was taken as the combination of the two mutations results in negative steric effects against each other. Comparison RMSD from Molecular dynamics simulation showed that AkDCaseD30A was stable and lower than that of WT at 40 and 60 °C. Co-expression of AkDCaseD30A with Hase from Geobacillus stearothermophilus in a single Escherichia coli cell, 29.53 g/L D-p-HPG within 12 h, was produced with a 97% conversion and a 2.46 g/(L·h) space-time yield which was higher than that previously reported by Yang et al (2019).

ii) Engineered DAPDH in the synthesis of D-p-HPG

Xu et al (2021) expressed a highly stereoselective DAPDH variant from Corynebacterium glutamicum ($CgDAPDH^{BC621}$ containing five mutations: R196M/T170I/H245N/Q151L/D155G) in E. Coli together with Laad, Hmas (4-hydroxy mandelate synthase), and MDH after successful invitro biotransformation. However, biotransformation results showed that conversion of HPGA to D-P-HPG was the rate-limiting step and low activity of $CgDAPDH^{BC621}$ would result in low HPG with accumulation of 4-hydroxyphenylglyoxalate. To increase the activity of $CgDAPDH^{BC621}$, a directed evolution strategy was employed to reduce the hydride transfer distance during

the enzyme catalytic reaction. Three mutations were introduced in the CgDAPDH^{BC621} enzyme and when combined into CgDAPDH$^{BC621/D120S/W144S/I169P}$, a specific activity of 5.32±0.85 U·mg−1 ·protein which was 37-fold higher than that of CgDAPDH^{BC621} was obtained and therefore, CgDAPDH$^{BC621/D120S/W144S/I169P}$ replaced CgDAPDH^{BC621} in the biosynthetic cascade. To regulate the four enzyme expressions and activity, Tac and T7 promoters were utilized in pETDuet-1 plasmid to control the expression of PaMDH and CgDAPDH$^{BC621/D120S/W144S/I169P}$ respectively. For pmL-AAD and SambHmaS, T7 promoters were used in the pACYCDuet-1 plasmid. Both plasmids were then co-expressed in E. coli BL21 (DE3) and under the optimal induction and transformation conditions (0.7-mM NADP+, 0.5-mM CoSO4, 20-mM Tris– HCl buffer (pH 8.5) and 30 °C), 42.69-g/L D-HPG was obtained in 3-L fermentation using 20 g/L (wet cell) of resultant E. coli from 50-g/L L-tyrosine in 20 h with 92.5% conversion and>99% ee.

Tan et al (2023) engineered DAPDH from Prevotella timonensis by broadening the size of the active site to form PtDAPDH^{M4} (W121V/H227I/R181T/S72D/S160R) that exhibited a catalytic efficiency of 26.75-fold higher than the wild type toward 4-hydroxyphenyl glyoxylate to form D-p-HPG. PtDAPDH^{M4} was co-expressed with E. coli aromatic amino acid aminotransferase and Bacillus megaterium glucose dehydrogenase catalysing transamination to form 4-hydroxyphenyl glyoxylate (HPGA) and regeneration of NADPH respectively. Under optimum conditions, conversion reached 98.8% with a D-HPG titer of 19.76 g/L in 10 h from 20 g/L L-HPG conducted at a scale of 100mL.

iii) Engineering of Alanine Racemase (Alr) for D-Arginine synthesis

Willies et al (2012) utilized error-prone PCR to construct a mutant library that included three significant mutations; I195T, N223D, and I374N. All of these mutations were identified in a strain that was characterized to have a 20-fold reduction in catalytic efficiency and a 5-fold increase in K_m towards racemization of L to D Alanine. When the mutant was tested for racemic activity on L-Lys, L-Arg, and L-Leu, it exhibited a 1300% increment in activity towards the formation of D-Arg from L-Arg. However, the strain had no activity on L Lysine and Leucine. For the latter, a reason was obtained from docking studies which showed that the point mutation of I374N existed in the active site and thereby changed the polarity from hydrophobic to hydrophilic, thereby favoring attachment of polar substrates such as L-Arg and not L-Leucine. Based on this reason it was expected that L-Lys would also exhibit an increment in activity, however, this was not the case, hence suggesting more mutations that would favor L-Lys binding.

When the mutant was tested on other amino acids, it showed a 107% increment in activity towards the racemization of L-Ornithine and a +14% increment towards the racemization of L-Glu. However, L-Ala, L-His, and L-Ile activities significantly decreased. These results show the use of directed evolution of racemases towards broadening substrate activities although native activities may be lost. (Willies et al., 2012)

iv) Engineering via signal peptide deletion of Pseudomonas taetrolens Arginine Racemase for increased production of D-Lys, D-Ser, D-Orn, and D-Arg

Arginine Racemase is a broad substrate racemase containing a signal peptide of 23 amino acids t the 5’ end of its gene. Matsui et al., 2009 characterized Arginine racemase to have a high racemic activity on L-Lys among other substrates of L-Arg, L-Orn, and L-Ala. Sequencing of the amino-terminal end of ArgR revealed a possibility of processing of the enzyme with its signal peptide cleaved off. Results obtained by Stäbler et al (2011) through heterologous expression of Arginine Racemase showed that the racemase with signal peptide (ArgR-sp) was absent in the cell extract of Corynebacterium glutamicum while expression with signal peptide cleaved off (ArgR) revealed the presence of the racemase in the cell extract. These results are in agreement with the fact that ArgR-sp is localized in the periplasm and therefore, since racemization takes place in the cytosol, engineering by cleaving off the N-terminal signal peptide improves enzymatic catalysis in host cells.

When ArgR-sp and ArgR were separately expressed in Corynebacterium glutamicum strains engineered for accumulation of L-Lys, L-Ser, L-Arg, and L-Orn, no corresponding D-enantiomers were detected in strains carrying ArgR-sp plasmids. However, strains carrying ArgR were able to accumulate D enantiomers in the extracellular medium. The only challenge was the presence of both L and D enantiomers at equimolar concentrations, therefore more research is required toward uni-directional racemization. Finally, these results showed how the location of an enzyme within a cell could affect its catalytic activity, and therefore engineering of localization signals could provide a way to improve enzyme catalytic properties. (Stäbler et al., 2011)

e) Reduction of Degrative pathways

Some bacteria, mainly gram-negative strains can utilize both L and D amino acids as sources of Carbon and Nitrogen and it appears that two distinct degradative pathways for L and D-amino acids exist which means that racemases play a key role in linking the two pathways by interconversion of L and D forms. In Bacteria, an enzyme

called D-amino acid dehydrogenase (DAD) exists which mediates the oxidation of free neutral D-amino acids to their corresponding α-keto acids leading to energetic catabolism. It was found by Naganuma et al (2018) that a strain A25 isolated from an ordinary river exhibited a higher growth rate with D-Glutamate than with L-Glutamate. Gene expression studies by reverse-transcription quantitative PCR (RT-qPCR) for target genes involved in Glutamate metabolism; encoding D-amino acid dehydrogenase (DAD; EC 1.4.99.1), glutamate racemase (EC 5.1.1.3), D-glutamate oxidase (EC 1.4.3.7 or EC 1.4.3.15), and UDP-N-acetyl-α-D-muramoyl-L-alanyl-D-glutamate ligase (EC 6.3.2.9) showed enhanced expression for DAD gene confirming its degradative role in D-amino acids. (Naganuma et al 2018)

Therefore, attention to D-Amino acid metabolism in host bacterial cells is crucial for the heterologous expression of interested synthetic pathways. Two main D-Amino acids are essential in E. coli; D-Alanine and D-Glutamate. For the former, two types of Alanine Racemases exist; anabolic alanine racemase (Alr), and catabolic alanine racemase (dadX). Alr appears to be expressed constitutively and functions in the production of D-Alanine primarily for cell wall synthesis. However, dadX is only activated by a high L-Alanine concentration in the growth medium, and functions to produce D-Alanine from L-Alanine for further breakdown into pyruvate by D-Amino acid dehydrogenase (dadA). It should be noted that even though dadA is most active on Alanine, it can act on several other D-amino acids such as D-methionine, D-serine, and D-proline but to a lesser extent. In Bacillus Subtilis, D amino acid transferase (DAT) occurs and this appears to catalyze the transfer of an amino group from D-amino acids of alanine, leucine, aspartate, glutamate, aminobutyrate, norvaline, and asparagine to alpha-keto acids generating mainly D-Alanine and D-Glutamate. It is on this basis that two main degradative enzymes may interfere with the yield of some D-amino acids if synthesis by heterologous expression is to be utilized; dadA, and DAT, and therefore depending on the D-amino acid of interest, there would be a need for downregulation of these two genes in host cells.

f) Broad-specificity Racemases in D amino acid production

Several racemases have been characterized to have racemic activity towards amino acids however, there are limited studies and research about the utilisation of racemases in the production of D-amino acids yet they can catalyze a one-step stereo inversion of L to D enantiomers.

The main reason for few reports can be explained by the low enantioselectivity of Racemases and therefore engineering of native racemases towards the improvement of the stereospecificity for D-amino acids production is seen as the only solution. It is also reported that racemases can be engineered for broad substrate activity. In both cases, the available knowledge dictates the directed evolution method to be utilized; either random mutagenesis or the use of structural and bioinformatics-supported considerations.

There is limited information present on substrate-specific racemases catalyzing the interconversion of some amino acids, particularly Tryptophan, Arginine, and Lysine, therefore, screening of broad-specific racemases that would have racemic activity including the three amino acids would be a solution to diversify biotransformation. Below we have identified a few racemases with possible application towards D amino acid production.

Broad specificity amino acid racemase from Pseudomonas Putida (bar)

This racemase enzyme catalyzes the interconversion of L-lysine and D-lysine, and L-arginine and D-arginine at optimum conditions of 37^{o}c and pH of 9.0. Kino et al (2007) found a low activity of the enzyme for racemization between L-Tryptophan and D-Tryptophan. To increase the latter activity, random mutagenesis was carried out and it was found that the substitutions at Y396H and I384M increased the tryptophan-specific racemization activity and the racemization activity for overall amino acids**.** It was reported that mutant I384M resulted in a yield of 1.8g/l of D-Tryptophan and although it was lower compared to when the Hydantoin process is used, L-Tryptophan as a substrate is cheaper hence affordable production. This demonstrates how the directed evolution of enzymes could be a solution for enhancing the catalytic activity of broad-specific racemases towards bioconversion of L to D amino acids.

Broad specificity amino acid racemase from Vibrio Cholerae (bsrV)

bsrV amino-acid racemase is able to utilize a broad range of mainly basic amino acid substrates. It reversibly racemizes ten of the 19 natural chiral amino acids known, including both non-beta-branched aliphatic amino acids (Ala, Leu, Met, Ser, Cys, Gln and Asn) and positively charged amino acids (His, Lys and Arg). Among these substrates, bsrV racemizes lysine and arginine best. It is also able to catalyze the racemization of several amino acids that are not typically incorporated into proteins such as ornithine and norleucine. It Is not active on negatively charged (Glu and Asp) or aromatic (Tyr, Trp and Phe) amino acids and displays minimal activity towards beta-branched aliphatic (Ile, Val, and Thr) substrates.

Vibrio Cholerae accumulates D-Arginine using bsrV: D-Arginine is one of the most dominant Non-canonical D-amino acids (NCDAAs) synthesized by Vibrio cholerae during the stationary phase and it functions to display

toxicity towards competing bacterial species meaning that D-Arginine limits the growth of several bacterial species. (Alvarez et al., 2018) It was found that no D-Arginine can be produced by V. Cholerae mutants without bsrV enzyme thereby confirming that the enzyme is responsible for the accumulation of D-Arginine in the extracellular environment. (Alvarez et al., 2018)

Broad specificity amino-acid racemase RacX from Bacillus Subtilis

Amino-acid racemase can utilize a broad range of substrates. Preferentially catalyzes the epimerization of LL-diaminopimelate, as well as the racemization of D-lysine, L-arginine, L-ornithine, L-lysine, and D-arginine. Has lower activity against D-ornithine, L-histidine, L-alanine, L-tyrosine, L-phenylalanine, L-serine, L-glutamine, L-methionine, L-asparagine and L-homoserine. Has weak activity against L-norleucine, L-aminobutyric acid, and L-norvaline. Has no activity toward nine L-amino acids (Thr, Glu, Asp, Val, Leu, Ile, Trp, Cit, and Aad) (Miyamoto et al., 2017)

Broad-spectrum amino acid racemase (Alr) of *Pseudomonas putida* KT2440

Previous studies on Pseudomonas putida showed its high efficiency in utilizing D-amino acids as sole sources of Carbon and Nitrogen. Radkov and Moe (2013) identified a putative biosynthetic alanine racemase (Alr) in Pseudomonas and invitro enzyme assays showed that the enzyme has broad substrate specificity, exhibiting measurable racemase activity with 9 of the 19 chiral amino acids. Among these amino acids, activity was the highest with lysine. Radkov and Moe (2018) reported Alr enzyme to be a link between the independent pathways that utilize L and D lysine by enabling the bacteria to convert L form to D form for further breakdown. This was confirmed by results obtained when Δ*alr* strains accumulated no D-Lysine in the media as compared to wild-type strains when the two were cultured on LB medium containing L-Lysine. Also, through analysis of the media supernatant, it was found that Δ*alr* strains accumulated L-Lysine in the growth medium despite the high concentrations of L-Lysine present as a substrate. This further shows that Alr is crucial in the catabolism of L-Lysine by first converting it to D-Lysine which is further broken down through the L-pipecolate pathway. (Radkov & Moe, 2018)

Broad specific Isoleucine-2-epimerase (ILEP)

ILEP Racemase enzyme from *Lactobacillus buchneri* was utilized by Yanqi et al (2023) and compared to Alr, ygeA, and bsrV, it showed the highest D-amino acid titer (g/l) and was able to racemize a great deal of L amino acids. Awad et al (2017) characterized the structure of ILEP enzyme and found out that it catalyzes the pyridoxal 5'-phosphate (PLP)-dependent racemization and epimerization of a broad spectrum of nonpolar amino acids from L- to D-form and vice versa, in particular isoleucine. This enzyme was the first epimerase to be identified from Lactobacillus and is responsible for the accumulation of D-leucine, D-allo-leucine, and D-valine in the culture medium of Lactobacillus. (Mutaguchi et al., 2013)

It should be noted that the overall 3D structure among fold-type I PLP-dependent enzymes is conserved although residues in the binding pocket must be different to offer distinctive topologies to accommodate specific substrates. Among conserved residues is Arginine at position 408 which is responsible for the formation of a salt bridge with the amino acid Cα carboxylate. Awad et al (2017) characterized potential residues in the active site that could be responsible for specific binding and orientation of the branched nonpolar amino acid substrates. These were Ala54, Tyr142, Met159, Tyr84, Leu307, and Thr309 and they provided hydrophobic patches within the active site. Following substrate recruitment, the ILEP enzyme utilizes conserved catalytic residues that may be accountable for its amino acid racemization reactivity, notably the lysine at position 280, and the tyrosine 142 pair in deprotonation/protonation reactions.

Insilco studies show a possibility of Racemase engineering toward stereospecificity

There are few reports about the engineering of Racemases towards L enantiomer specificity and it is on this basis that we further used molecular docking and molecular dynamics simulation on an already characterized structure of Isoleucine-2-epimerase enzyme for racemization of L and D Leucine to suggest residues responsible for specific enantiomer substrate binding and thus demonstrate a possibility of enzyme engineering towards enantiospecificity thereby exploring the findings by Awad et al (2017)

There were two suggested approaches towards active site characterization; the first was the docking of L and D Leucine with PLP complexed ILEP (5LL3), and the second was the docking of external PLP aldimines of both L and D Leucine with Apo ILEP enzyme (5LL2) (Awad et al., 2017). We tested the two approaches using Autodock Vina (Trott & Olson, 2010). However, to our surprise, we found no docking poses showing signs of substrates interacting with the PLP for the first approach.

We then constructed external PLP-Leucine aldimines for both enantiomers using MolView software (Smith, 1995), and using Apo ILEP, we performed docking. From docking, the first poses with the lowest energy were considered for Molecular dynamics simulation using GROMACS, and results were analyzed using VMD and PyMOL software

tools. Here we assumed that the enzyme should change conformation following the formation of an internal aldimine with the PLP of the Holoenzyme. This meant that docking with an external aldimine would make the enzyme achieve similar conformation as the case of real substrate binding to the PLP-bound Holoenzyme.

Analysis of MD simulation results showed Lys280 and Tyr142 residues to be associated with both L and D PLP-L/D Leucine aldimines throughout the simulation as illustrated in Fig 7 and 8. This is in line with previous research carried out by Awad et al (2017) and suggests that racemization of both L and D Leucine utilizes Lys280 and Tyr142 catalytic pair. Of the potential residues responsible for stereospecificity, (Ala54, Tyr142, Met159, Tyr84, Leu307, and Thr309) Awad et al (2017), Tyr84 was associated with PLP L-Leucine aldimine, while Met159 was associated with PLP D-Leucine aldimine. In addition to Met159, Ser145 was found to make polar contacts as illustrated in Fig 8. This probably suggests that the binding of L and D Leucine causes two different conformational changes of the enzyme although the position of the catalytic pair relative to the substrate is conserved. The residues Tyr84, and Met159, and Ser145 could be responsible for the orientation of L-Leucine and D-Leucine respectively in the active site of ILEP. It is at this point that such results provide an example of residues that could be mutated to disable the proper binding and orientation of one of the enantiomers to favor stereo inversion in a particular direction.

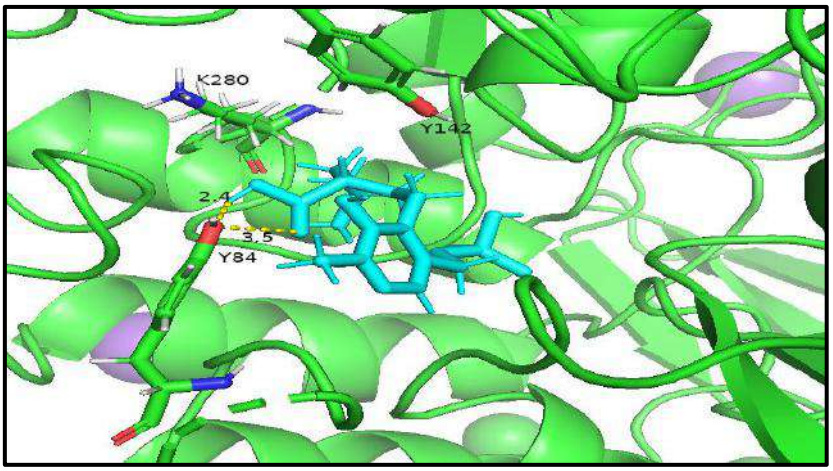

Fig 7: Molecular dynamics simulation of ILEP with an external PLP-L-leucine aldimine shows Tyr 84 forming polar contacts with L-leucine possibly suggesting a role in substrate stabilization in addition to Tyr 142 and Lys 280 catalytic pair.

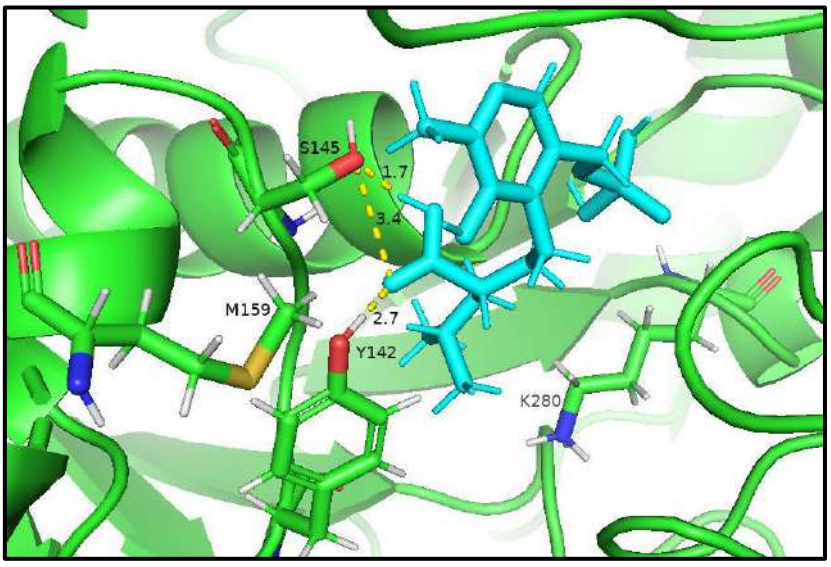

Fig 8: Molecular dynamics simulation of ILEP with an external PLP-D-leucine aldimine shows Met 159 and Ser 145 with the later forming polar contacts with D-Leucine.

V. CONCLUSION

This review summaries recent methods and challenges in the synthesis of D amino acids. These challenges identified mainly include the high cost of some raw materials such as alpha-keto acids, toxicity of D amino acids to host cells, and low yield probably coming as a result of degradation of the D amino acids within the host cells and reaction intermediates translocation. We have tried to identify solutions utilised to these challenges primarily focusing on toxicity through acetylation and screening of host cells, enhancement of yield via reduction of degradative pathways and engineering of enzymes involved in D amino acid production. Furthermore, since racemases catalyze stereo inversion of both L and D amino acids, we suggest that enzyme engineering towards enantiomer specificity could solve the problem. To show the applicability of this solution, we utilized the PDB structure of alloisoleucine-2-epimerase and external PLP-L/D-leucine aldimines in docking and molecular dynamics simulation. From simulation results, we suggest that out of Ala54, Tyr142, Met159, Tyr84, Leu307, and Thr309 residues identified by Awad et al (2017), Tyr84 and Met159 could be involved in substrate binding and orientation of L and D Leucine respectively hence highlighting the two possible residues that could be utilized in ILEP engineering. The later findings provide opportunities for using site-directed mutagenesis of Racemases toward one direction L to D stereo-inversion in a one-step reaction with an overall increase in yield.

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The Analysis of the Spatio-temporal Evolution of the Heat Island Effect and its Influencing Factors in Huadu District, Guangzhou

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Abstract— *This study aims to explore the urban heat island intensity and change characteristics of Huadu District, Guangzhou. Landsat 8 was used as the data source, and an atmospheric transmission model was used to invert the surface temperature of Huadu District, Guangzhou, in winter 2013 and 2021, extract the urban core area, and divide the heat island effect area. The results show that the urban heat island of Huadu District in Guangzhou has changed greatly in 2021. Compared with 2013, the proportion of low-temperature areas and sub-low temperature areas in urban areas has decreased in 2021, while the high-temperature area, sub-low temperature area, and middle-temperature area have increased. In terms of space, the high-temperature area of Huadu District, Guangzhou, is mainly distributed in the built-up area in the south and north of the city; the sub-low temperature area is distributed on the edge of the built-up area; the medium-temperature area occupies most of the district of Huadu District, distributed in the city's central area and the edge of the low-temperature area; and the low-temperature area is distributed in the water body and dense vegetation area.*

Keyword— ***Atmospheric Correction Method; Surface Temperature Inversion; Urban Heat Island (UHI); Spatio-temporal Evolution, Normalized Difference Vegetation Index（NDVI）***

I. INTRODUCTION

The urban heat island (UHI) effect refers to the phenomenon that the temperature in the city is significantly higher than that in the outer suburbs due to a large number of artificial heating systems, the increase of high heat storage bodies such as buildings and roads, the decrease of green space, and the decrease of wind speed affecting heat transport. The measure of its strength is heat island intensity. Human beings have long discovered that the atmospheric environment in cities has different characteristics from those in rural and mountainous areas. The British man Lake Howard first documented in 1833 the phenomenon of higher temperatures in the center of London compared to the suburbs, and Manley first proposed the concept of Urban Heat Island (UHI) in 1958. It is now widely believed that the urban heat island effect refers to the phenomenon where, when a city develops to a certain scale, the temperature of the city is significantly

higher than that of the suburbs due to changes in the nature of the urban underlying surface, air pollution, and the emission of artificial waste heat, forming a phenomenon similar to high-temperature islands.

With the rapid urbanization process and the rapid expansion of urban built-up areas, the urban heat island effect is intensifying. Materials such as asphalt, concrete, and cement transform natural surfaces into the underlying surface of the building, reducing transpiration rates, rainwater retention, and altering the reflectivity of the surface [1], "resulting in undesirable thermal effects." Urban land absorbs solar radiation during the day and slowly re-radiates the absorbed heat at night. This results in the generation of heat islands in urban areas, with higher temperature values compared to neighboring areas [2]. During the year, the increase in summer surface temperature contributes to the increase in atmospheric temperature, which seriously affects the comfort of the human body surface. The severe haze weather in winter will also have a synergistic effect with the heat island effect, leading to the deterioration of the winter environment.

Huadu District of Guangzhou is located in the central part of Guangdong Province, the north of the Pearl River Delta, and the north of Guangzhou, Guangdong Province. In 2021, the main goal of the economic and social development of Huadu District in the next five years is to achieve an average annual GDP growth rate of about 6% and build a new industrial pattern of "one core leading, three belt linkages, and six functional areas supporting." Under the grand goal of rapid construction and harmonious rise, the expansion of urban scale will accelerate, and the heat island effect will be particularly significant. Therefore, studying the spatio-temporal evolution characteristics of the heat island effect in Huadu District of Guangzhou has important theoretical and practical significance for the protection of the urban geological environment, disaster prevention and reduction, and the realization of sustainable urban development.

II. STUDY AREA AND DATA SOURCE

2.1 Study Area

Huadu District is located in the north of Guangzhou. Its climate is a subtropical monsoon climate. It has the climatic characteristics of high temperatures, high precipitation, long summers and short winters, and a long frost-free period. The average annual temperature is 21.8 degrees, the average annual rainfall is 1753.9 mm, the average relative humidity is 76%, the dominant wind direction is southeast throughout the year, the frost-free period is 365 days, and the groundwater level is about 1.2–3.5 meters. There are obvious seasonal changes in temperature, humidity, precipitation, wind direction, and wind speed [3] (Figure 1).

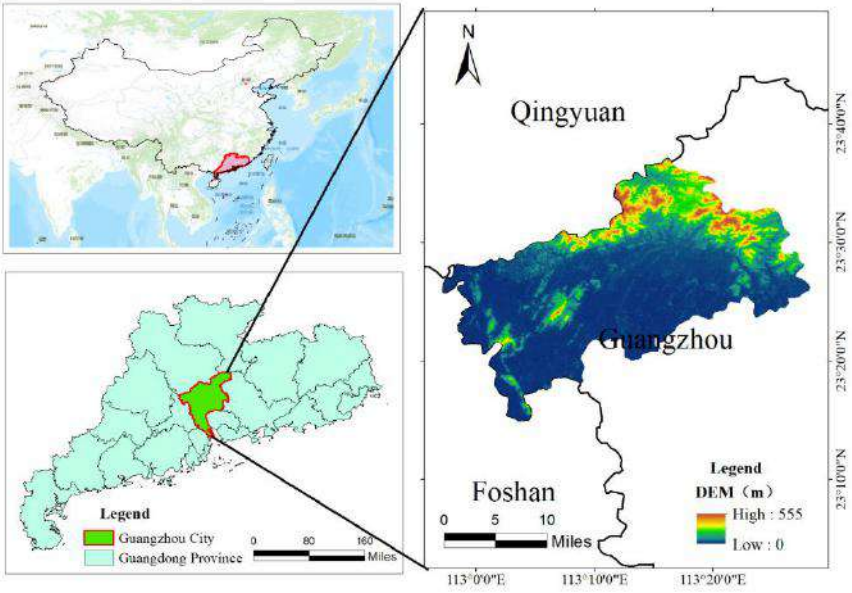

Fig.1 Location and Topographic Map of Huadu District in Guangzhou

2.2 Data Sources

This paper takes the Huadu District of Guangzhou (E113°1313 ', N23°2414 ') as the research object. The data is obtained from a geospatial data cloud (http://www.Gscloud.cn) with good image quality and basically no cloud. Use two Landsat 8 images from the winter of 2013 and 2021 for analysis and comparison (Table 1). Landsat8 is the eighth satellite of the United States Land Satellite Program (Landsat), launched on February 11, 2013, initially known as the Landsat Data Continuity Mission (LDCM). Landsat 8 carries an Operational Land Imager (OLI) and Thermal Infrared Sensor (TIRS), and many studies on surface temperature inversion use this satellite image data.

Table 1 Remote sensing image data sources

Item	Imagery	
Date	2013-12-31	2021-02-20
Data identification	LC81220442013365LGN01	LC81220442021051LGN00
Sensor type	Landsat8_OLI and TIRS	
Spatial resolution	OLI 30m/ TIRS 100 m	
`Strip number	122	
Line number	44	

III. RESEARCH METHODS AND PROCESSES

This study adopts the remote sensing surface temperature inversion method, and the main principle is that after the solar radiation reaches the surface, part of the radiation energy is reflected, and the other part of the energy is absorbed by the ground, so that the ground temperature rises and then is measured by the instrument. At present, the use of remote sensing technology to retrieve land surface temperature has become mainstream; it has a lot of advantages, and the main land surface temperature inversion algorithms are as follows: atmospheric correction method, single window algorithm, and split window algorithm [4].

In this paper, the atmospheric correction method is used to invert the surface temperature of Huadu District. The atmospheric correction method, also known as the radiative transfer equation method, is based on the principle of subtracting the influence of the atmosphere on the surface radiation from the total thermal radiation received by the satellite sensor and converting the remaining radiation value into the corresponding surface temperature. The heat radiation value of the atmosphere, the energy reflected by the atmosphere down to the sensor through the surface, and the energy transmitted to the sensor through the atmosphere after the surface absorption constitute the total heat radiation of the satellite[5]. The advantage of the radiative transfer equation is that there is no limit to the thermal infrared band, but it needs four parameters: atmospheric transmittance, atmospheric up-going radiation value, atmospheric down-going radiation value, and surface-specific emissivity.

Based on the above principles, this study uses Landsat 8 images as materials. After image preprocessing, NDVI （Normalized Difference Vegetation Index）extraction, vegetation coverage calculation, and surface radiance calculation are performed. Combined with the 10th band image, radiance calculation and temperature inversion are performed. Finally, the spatiotemporal changes are analyzed, and the conclusion is estimated (Figure 2).

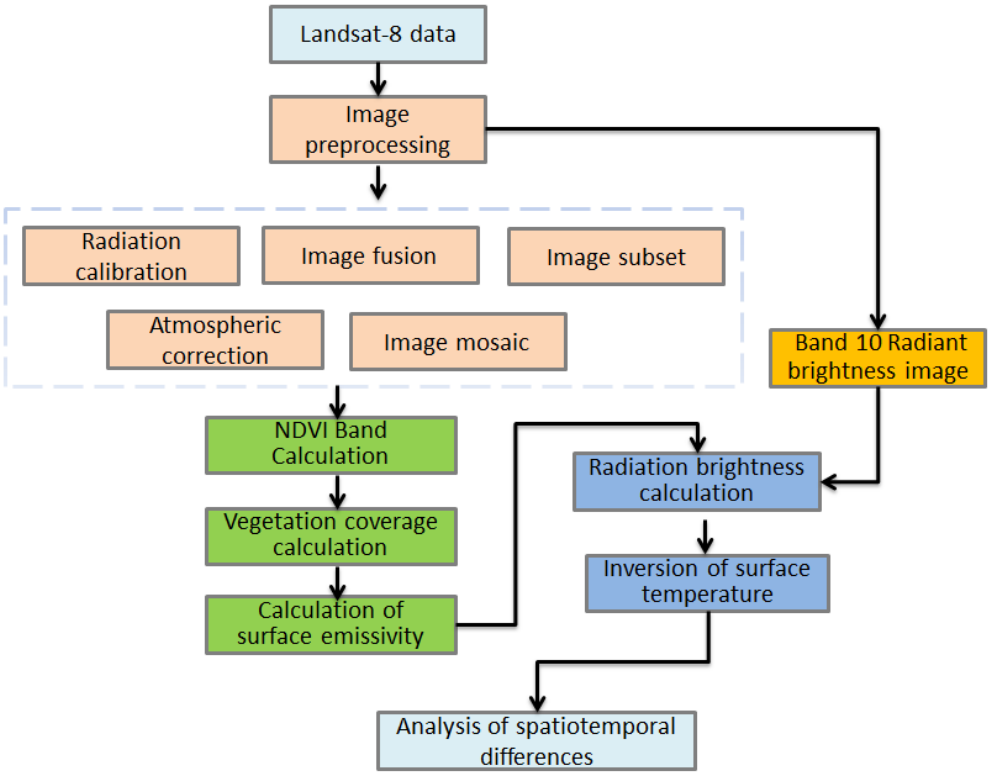

Fig.2 Technical roadmap

3.1 Radiometric Calibration

The surface radiation brightness and surface temperature in the thermal infrared band cannot be directly recorded by the satellite sensor, which is usually stored as a gray value (DN value), and the DN value recorded by the sensor needs to be converted to obtain the actual spectral information of the ground object. We call this conversion process radiation calibration[6]. Usually, a linear relation is used to complete the conversion of the gray value and the radiation brightness value of ground objects, and the formula is as follows:

La = Gain * DN + Offset (1)

In the formula, L represents the radiation brightness value of the ground object, and its unit is W/(m'·um·sr). Gain is the transformation value, and its calculation formula is as follows:

Gain = (Lmax a –Lmina)/2552.2) (2)

In the formula, Lmax a represents the maximum radiation luminance value, and Lmin a represents the minimum radiation luminance value. Offset is the deviation transform value, offset = Lmin a, and the calibration parameters are obtained from the Landsat image metadata to calculate the radiometric calibration data by selecting high-gain bands.

3.2 Calculation of NDVI

The formula for NDVI is (NIR-R)/(NIR+R) (3).

NIR is the reflection value in the near-infrared band. R is the reflection value in the red light band. The NDVI is one of the important parameters reflecting crop growth and nutritional information. Based on this parameter, the nitrogen demand of crops in different seasons can be determined, which plays an important guiding role in the rational application of nitrogen fertilizer.

1. The application of NDVI: detecting vegetation growth status, vegetation coverage, and eliminating some radiation errors;

2. -1 visible light high reflection; 0 represents the presence of rocks or bare soil, and NIR and R are approximately equal; A positive value indicates vegetation coverage and increases with increasing coverage.

3. The limitation of NDVI is that it enhances the contrast

between NIR and R reflectivity through nonlinear stretching. For the same image, when calculating RVI and NDVI separately, it can be found that the rate of increase in RVI value is higher than that of NDVI, indicating that NDVI has a lower sensitivity to high vegetation areas.

This study with the NDVI of the ENVI Toolbox, select the cut multi-spectral OIL band data and calculate the NDVI by using the short red and near infrared bands corresponding to the 4th and 5th bands of OLI in the NDVI calculator parameters dialog box.

3.3 Calculate the Vegetation Coverage

Vegetation coverage reflects the density of vegetation in a region and is also an important parameter of ecological environment assessment. The minimum value of vegetation coverage is 0, and the maximum value is 1. The inversion formula is as follows:

VFC = (NDVI – NDVI*soil*)/(NDVI*veg* - NDVI*soil*) (4)

(NDVI*soil* represent the NDVI value of bare land, and NDVI*veg* represents the NDVI value of pure vegetation cover.)

The above two values are determined according to the actual situation of this paper. For example, the confidence range of 5%~95% is adopted, and VFC*max* and VFC*min* are estimated according to experience. Tool input ((b1 lt NDVI*soil*)*0 + (b1 gt NDVI*veg*)*1 + (b1 ge NDVI*soil* and b1 le NDVI*veg*)* (b1-NDVI*soil*)/(NDVI*veg*-NDVI*soil*)[7]. Using ENVI's band math, the NDVI*soil* and NDVI*veg* values are obtained (Table 2).

Table 3 NDVI values of bare land and pure vegetation

Year	*2013*	*2021*
NDVIsoil	*-0.145098*	*-0.082353*
NDVIveg	*0.364706*	*0.317647*

3.4 Calculation of Specific Emissivity

Surface-specific emissivity is an important parameter of land surface temperature inversion that directly determines the accuracy of the inversion results. In order to obtain more accurate inversion results, this paper uses the surface-specific emissivity algorithm to divide the surface into water bodies, natural surfaces, and urban areas. The specific emissivity of the water body is equal to that of the black body (0.995), and the specific emissivity of the natural surface and urban area is calculated as follows [8]:

Natural surface-specific emissivity: εsurface =0.9625 +0.0614FV -0.0461FV (5)

Urban area-specific emissivity: εbuilding = 0.9589 + 0.086FV - 0.0671FV (6)

3.5 Estimation of Atmospheric Parameters

The radiation parameter information of Huadu District in 2013 and 2021 can be obtained from the NASA website as follows (Table 4):

Table 5 Radiation parameter information

Year	Atmospheric transmittance	Atmospheric upward radiance	Atmospheric downward radiation brightness
2013	0.93	0.50	0.85
2021	0.82	1.34	2.22

3.6 Calculation of Blackbody Radiance

Surface brightness temperature refers to the temperature of the blackbody with the same radiation brightness as the surface, referred to as the surface brightness temperature. That is, the radiation temperature of the surface object itself is the radiation intensity observed by the sensor at the height of the satellite. Surface brightness temperature can be calculated by the formula

D(t)=[Q-q2-a(1-x)q1]/ax (7)

which is converted to Bandmath as follows:

(b2-q2-a*(1-b1)*q1)/(a*b1) (8)

b1 in the formula is the surface specific emissivity, b2 refers to the radiation brightness value of the 10th band obtained at the beginning, and then the inversion is obtained according to the transmittance a of the atmosphere in the thermal infrared band, the atmospheric upward radiation brightness q2, and the atmospheric downward radiation brightness q1.

3.7 Inversion of Surface Temperature

The true surface temperature can be obtained by the Planck function, as follows:

Ts = K2 /ln(K1 /B(Ts) + 1) (9)

Where, Ts is the true surface temperature, K1 and K2 are constants, and the values of K1 and K2 are different for different sensors. The specific values of K1 and K2 are shown in the Table 4 [4]:

Table 6 Different sensor K1, K2 value standard

Sensor type	K1	K2
Landsat8_OLI	774.89	1321.08
Landsat7_ETM+	666.09	1282.71
Landsat5	607.76	1260.56

IV. ANALYSIS RESULT

4.1 Divide Urban Heat Island Effect Areas

Due to the different imaging times of remote sensing images, the difference between the maximum and minimum values of the inverted land surface temperature is large, and the existence of abnormal temperature values is not convenient for comparative analysis, so the images are normalized. The current normalization method is normalized according to the maximum and minimum values, which is not suitable for the division of heat islands in the region due to the existence of outliers and the accidentality of this method, and the other method is to normalize the mean-standard deviation. In this paper, the mean-standard deviation method was used to statistically analyze the heat island effect. The study area is divided into five temperature range classes, and the mean-standard deviation classification rules and heat island classification are shown in Table 5[9].

According to the above principles, after reclassification, the heat island of surface temperature is divided into five levels, including the low-temperature areas, sub-low temperature area, middle-temperature area, sub-high temperature area, and high-temperature area. Meanwhile, we complete the visualization of surface temperature classification zoning maps for 2013 and 2021 (Figure 3 and Figure 4).

Table 7 Mean-standard deviation temperature classification

Heat island level	Temperature zone level	The difference in average temperature between urban core and non-core areas
Cold island	Low temperature area	$T_s < \mu - \text{std}$
Green Island	sub-low temperature area	$\mu - \text{std} \le T_s < \mu - 0.5\text{std}$
Normal area	Middle-temperature area	$\mu - 0.5\text{std} \le T_s \le \mu + 0.5\text{std}$
Subheat island	Sub-high temperature area	$\mu + 0.5\text{std} < T_s \le \mu + \text{std}$
Island of intense heat	High-temperature area	$T_s > \mu + \text{std}$

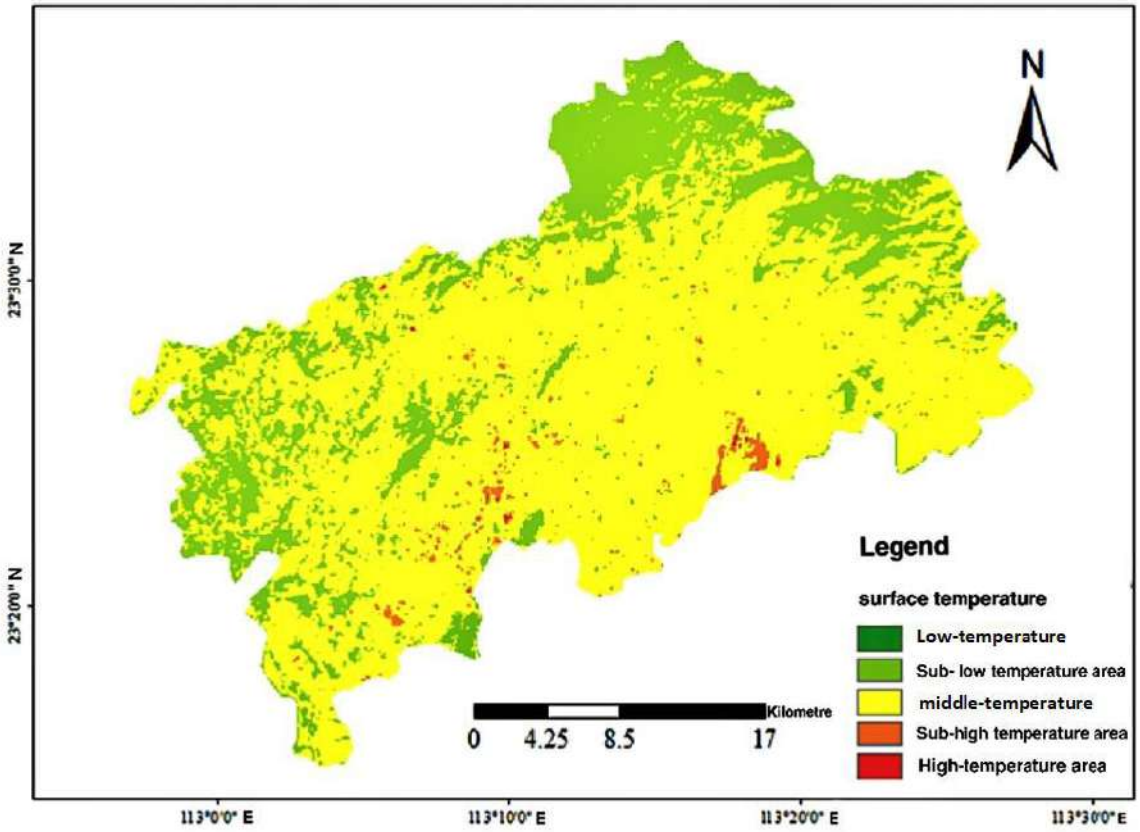

Fig.3 Classification map of surface temperature in Huadu district in 2013

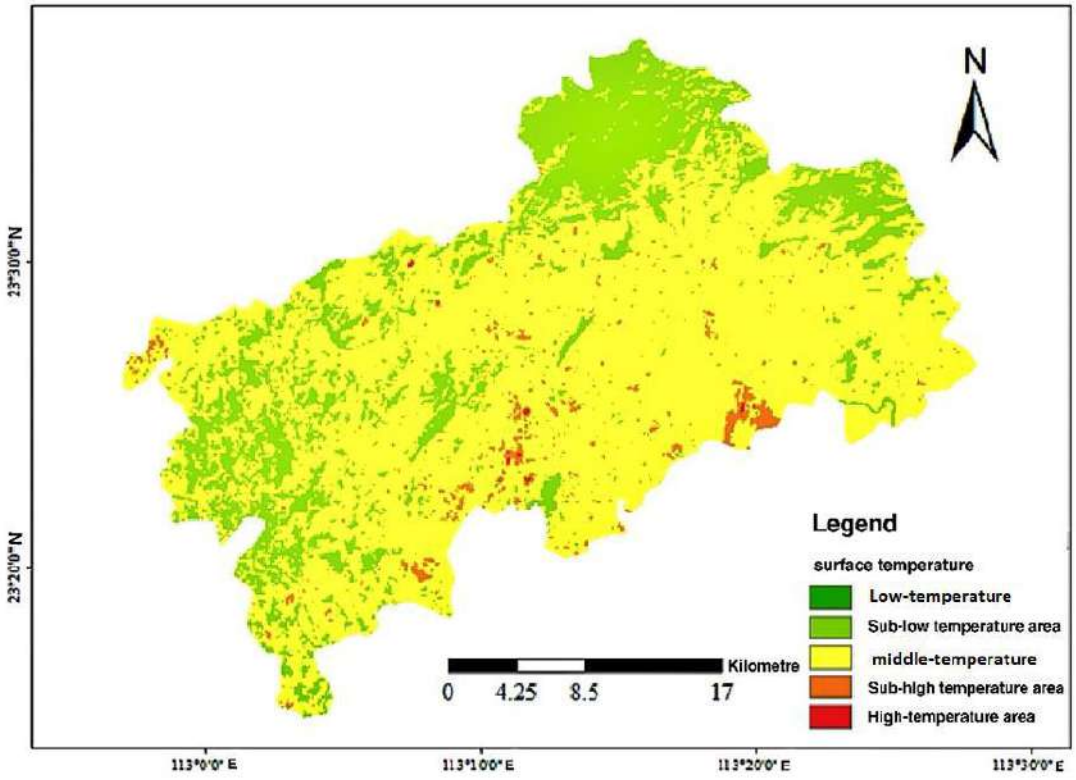

Fig.4 Classification map of surface temperature in Huadu district in 2021

4.2 Analysis of Spatio-temporal Changes of Heat Island Effect

Due to the different imaging times of remote sensing images, the difference between the maximum and minimum values of land surface temperature and the existence of abnormal temperature values are not accurate for comparative analysis. However, it can still provide a reference value for recognizing the trending patterns in the difference in temperature change. From the analysis results, the intense heat island area of Huadu District in 2013 and 2021 was located in the southern part of the city (Figure 5 and Figure 6), and distributed in built-up areas. The sub-heat island area is distributed along the edge of the high-temperature area, and there is no obvious rule. Normal areas and middle-temperature areas occupy most of the whole urban area of Huadu District and are mostly distributed in the surrounding areas of Huadu District. Low-temperature areas are mainly distributed in water areas, lakes, rivers, and other low-temperature areas.

Overall, the spatial distribution of urban heat islands in Huadu District has clearly changed in the past 8 years. The area of the strong heat island area has increased and expanded along the northern and eastern parts of the city, while the area of the sub-heat island area has decreased and shifted to the high-temperature area, while the distribution of the normal area and the green island area has not changed greatly. However, most of the low-temperature areas in the western part of Huadu District have been transformed into sub-low temperature areas and medium temperature areas.

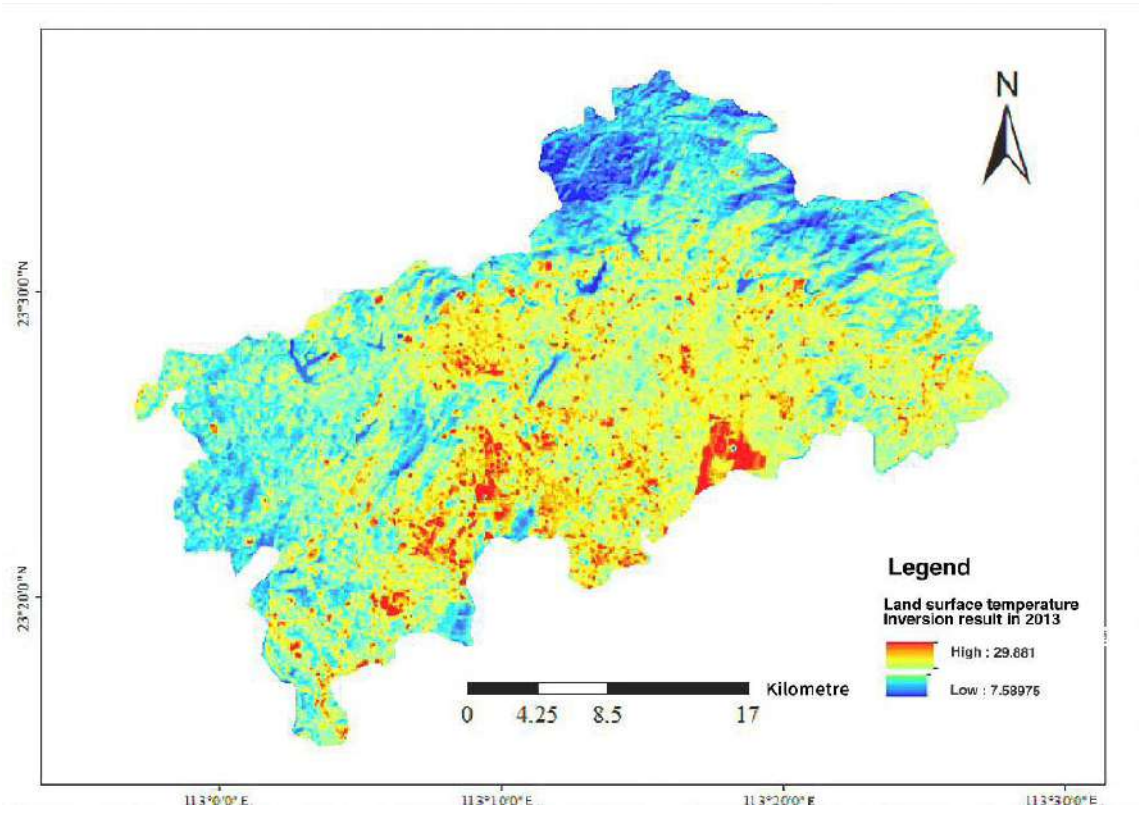

Fig.5 Inversion results of surface temperature in Huadu, Guangzhou in 2013

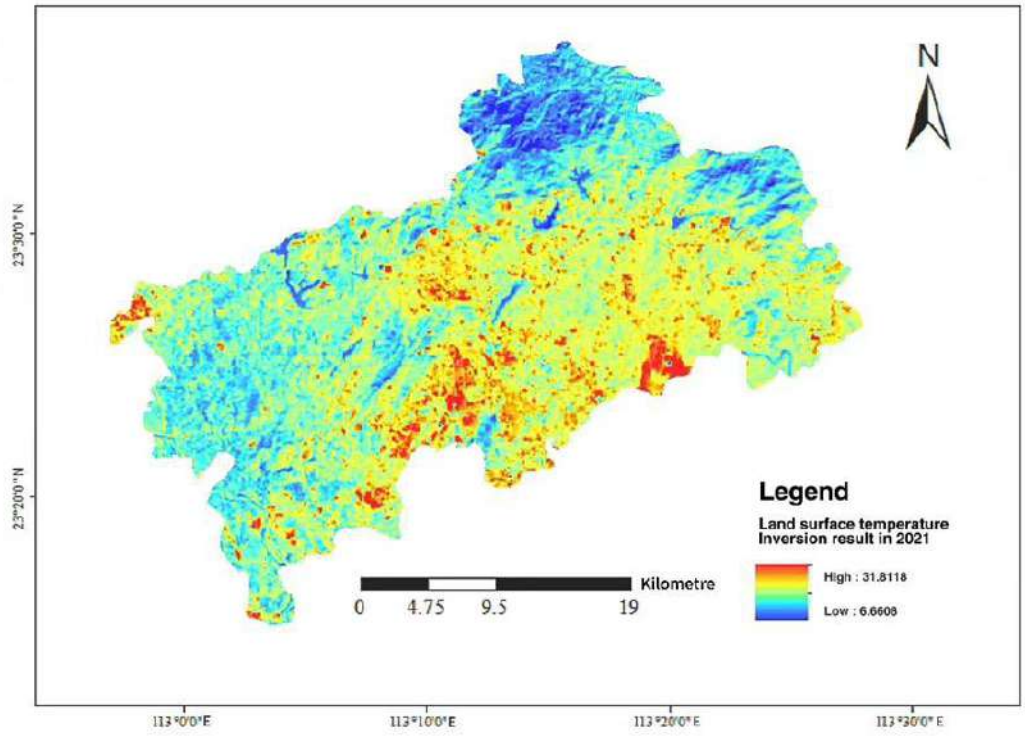

Fig.6 Inversion results of surface temperature in Huadu, Guangzhou in 2021

V. CONCLUSIONS

In terms of spatio-temporal analysis of land surface temperature, it is found that the urban intense heat island area of Huadu District is distributed in the south and east, and the small built-up area is in the northwest. The sub-low temperature area has no obvious distribution pattern, and most of them are located at the edge of the middle-temperature area. The sub-low temperature area and low-temperature area occupy a small part of Huadu District and are distributed in the northern fringe area, while the low-temperature area is distributed in the water body and the lush vegetation area. In the past 8 years, the strong hot island area has shown an expanding trend along the east-west direction of the parallel.

The increase in building area is the main reason leading to the expansion of the strong heat island area, and water and vegetation are the main factors leading to the decline of the heat island effect. Therefore, urban construction should be rationally planned, and the protection of vegetation areas should be strengthened in the process of urbanization. According to research, if the urban green coverage rate is greater than 30%, the heat island effect can be significantly alleviated. The coverage rate is greater than 50%, and the mitigation effect of green space on heat island is extremely significant. In addition, roof greening also has a certain effect on improving the urban climate. Every inch of urban land is valuable. The popularization of roof greening can effectively increase the urban green area and play a role in heat insulation and rainwater storage. At the same time, it is suggested to increase the area of urban water bodies and wetlands. Urban water bodies and wetlands have a huge heat capacity and are a cold source in summer, which can absorb a lot of waste heat from the urban environment.

The change in urban surface albedo is also an important cause of urban heat islands. Improving urban reflectivity can reflect part of solar radiation outward, reduce urban heat transfer to solar radiation, and alleviate the urban heat island effect. Therefore, in the process of urban planning and design, materials with high reflectivity are selected or the building surface is painted so that more solar radiation is reflected out of the urban space. For example, the use of light-colored materials on roofs, roads, parking lots, etc. can effectively slow down the urban heat island effect[10]. Chicago, USA, has strengthened albedo management in

urban planning and developed building codes to improve roof reflectivity, which has proved to be beneficial to alleviate the urban heat island effect.

In addition, in recent years, the construction thinking of sponge cities has also become an important strategy for future urban development. It is a concept that promotes green building construction, low-carbon city development, and the formation of innovative ecological environments in smart cities. It is also an organic combination of modern green technologies and various factors such as society, environment, and culture in the context of the characteristics of the new era.

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Effect of Storage Period on Seed Germination in different Promising Lines of Bambara Groundnut (*Vigna subterranea* (L.) Verdc)

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Abstract— *Bambara Groundnut seeds produced in this season often stored for some period of time until the next planting season. Storage period is one of the factors determine the quality of seeds while seeds was stored. This research conducted to determine the effect of storage period on seed germination. Seed viability and seed vigour were observed in 6 promising lines of Bambara Groundnut on 2 different storage period. Experiments was conducted in Agronomy Department, Faculty of Agriculture, Brawijaya University, Malang Indonesia, from May to June 2022. Six promising lines of Bambara groundnut produced from different seasons were stored in cold storage for one year and two years. Each promising line that was stored on respective years was tested with between paper method (BP) according to ISTA. Data collected from the observation of germination, viability characters, vigour characters. Experiments arranged in completely randomized design with combinations of 2 storage period; 1 year, 2 years and 6 promising lines of Bambara Groundnut; CCC 1.6, PWBG 6, PWBG 5.2.1, SS 2.4.2, BBL 1.1, TVSU 86. Results showed that Storage period affect seed viability and vigour. Longer period of storage decreasing some germination characteristics. Seeds stored after 2 years have lower vigour index, final germination percentage and dry weight of shoot and root. Lowest final germination percentage in 2 years storage recorded in PWBG 5.2.1 (70%). Seeds stored on 1 year storage period have higher vigour index (30%-60%), higher germination rate and higher dry weight of shoot and root.*

Keywords— ***Bambara groundnut, germination, promising lines, seed quality, storage period***

I. INTRODUCTION

Bambara groundnut (*Vigna subterranea L.*) is an underutilized legume species originated in Africa and have great potential in terms of their adaptability to climates and high nutritional content [1]. Specifically, Bambara groundnut contain 18-24% protein with high methionine and lysine, 4-12% crude oil content, 51-70% carbohydrates, 3-12% fiber, and 3-5% ash [2]. Bambara groundnut seed have a very hard texture, smooth surface, usually round and vary in size, can reach 1.5 cm in diameter [3]. The yield of Bambara groundnut is strongly influenced by the time of planting. Optimal planting time is in early November to produce high yields and late planting time is around the end of December to January [4]. In Indonesia Bambara groundnut are planted once a year and the seeds are greatly available from March to June [5]. Bambara groundnut takes 4 to 6 months to mature and consumed fresh after boiled or sold directly in the market, while the rest is dried and stored for later [6]. The stored seeds will be planted in the next planting season which can take 6 months to 1 year and during the period of storage deterioration of seeds and damage usually begins to occur.

Storage period is one of the factors determine the quality of seeds while seeds was stored. Seed deterioration occurs mostly due to oxidative processes and aging [7]. Deterioration promotes the breakdown of proteins

(including enzymes), lipids, and then will damage cellular membranes, RNA, and DNA. And all these things adversely affect the cellular integrity and metabolism of seeds and seedlings [8]. When the seed's metabolic system starts to break down, seed ability to germinate decreased and some seeds unable to germinate properly and affect seed viability and vigour. Some seed viability and vigour characters including final germination percentage, germination rate, maximum growth potential of seed and vigour index could greatly decrease after some period of storage.

In the tropical country like Indonesia, high humidity and high temperature can be challenging factor for the storage of orthodox seed storage. Many species of legumes seeds including Bambara groundnut are orthodox seeds. According to the USDA Orthodox seeds are seeds that survive drying and freezing in ex situ conservation and are long-lived seeds. Most seeds remain in good quality for about two to three years but some deteriorate within a year depend on the storage environment. At the time of storage, humidity, temperature, and the proportion of oxygen are the main environmental factors that influence seed deterioration [9]. Storing seeds longer than 1 year becomes a challenge even where seeds are saved, stored, and planted on short annual cycles [10].

Bambara groundnut seeds often stored for one year or more than one year. During period of storage the quality of the seeds in the term of their viability, vigour, and ability to germinate is unknown and there's limited study about effect of period storage on Bambara groundnut. The study of effect of storage period on Bambara groundnut seeds is important to obtain the information about maximum storage period for Bambara groundnut seed and also gaining details about desirable and appropriate temperature and relative humidity for storing Bambara Groundnut seeds.

II. MATERIAL AND METHODS

The experiment was conducted at laboratory of Department of Agronomy, Agriculture Faculty, Brawijaya University, Indonesia from May to June 2022. The study was carried out on six promising lines of Bambara Groundnut. The objective of this study was to evaluate the effect of different storage period on Bambara groundnut germination characters including viability and vigour of the seeds. Seeds used in the experiment obtained from Prof. Dr. Kuswanto M.P. from Department of Agronomy Brawijaya University and produced from different season. All the seeds were stored in the plastic bag under same condition in the cold storage with temperature (24ºC - 26ºC) and relative humidity 40%. Laboratory experiment was conducted with Complete Randomized Design (CRD) with three replicates in multifactorial experiment. The first factor was period of storage (i.e., 1 year and 2 years of storage period) and the second factor was six promising lines of Bambara Groundnut (i.e., CCC 1.6, PWBG 6, PWBG 5.2.1, SS 2.4.2, BBL 1.1, TVSU 86.

Germination test was carried out using rolled paper method in accordance from ISTA recommendations. All seeds and tools were sterilized to prevent fungal contamination before seeds being placed on the surface of the paper. Seeds were soaked in 1% sodium hypochlorite for 5 minutes and rinsed with water afterwards. 10 seeds used for each replication so there's total 30 seeds on each treatment. The paper used to germinate was straw paper substrate. Each paper is cut with size of 26 x 15 cm. Paper substrate was sterilized using oven for 2 hours at 100°C. Three Sheets of straw paper was moistened with distilled water and placed on the prepared plastic sheet. The seeds then planted on straw paper and covered with another 3 sheets of moistened straw paper. Paper then rolled up with plastic paper and placed in the controlled germinator in a standing position. The temperature inside the germination chamber was 24ºC-27ºC. Seeds were observed for 14 days with first count at 7th day and final count at 14th day [5].

Studied germination characters were final germination percentage, seed moisture content, germination rate, vigour index, maximum growth potential of seeds and dry weight root and shoot. Seed moisture content was determined by oven drying method. 5 grams of Bambara groundnut seeds grinded until it forms fine granules and put in the oven for 1 hour at 130ºC.

Final germination percentage (FGP) was observed by calculating the total normal seeds geminate at first count and final count with formula given in [11].

$$FGP\ (\%) = \frac{\text{Germinated Seeds in First Count + Final Count}}{\text{Total Number of Seed Tested}} \times 100\%$$

Germination Rate (GR) is determined by calculating the number of days it takes for the radicle or plumule to appear over a certain period of time calculated by the formula given in [12].

$$GR\ (\%/day) = \frac{N1T1 + N2T2 + \ldots + NxTx}{\text{Total Number of Seed Tested}} \times 100\%$$

Vigour Index (VI) is the ratio between the number of normal germinated seeds on the first count and the total number of seeds planted [13], the total number of normal germinated seed on 7th day (first count) observed on each treatment and calculated by formula given in [14].

$$VI\ (\%) = \frac{\text{Number of Normal Germinated Seeds in First Count}}{\text{Total Number of Seed Tested}} \times 100\%$$

Maximum Growth Potential of Seeds (MGP) determined by calculating the total of both normal and abnormal seedling observed on day 14th (final count) by formula given in [15].

$$MGP\ (\%) = \frac{\text{Number of Normal and Abnormal Seed}}{\text{Total Number of Seed Tested}} \times 100\%$$

Dry weight root and shoot (g) observed on the last day of germination on day 14th (final count) and all normal germinated seeds on the rolled paper were cleaned from the cotyledons and then oven-dried at 80 ^{o}C for 72 hours.

Recorded data analysed using R studio program to perform ANOVA and continued with the Duncan Multiple Range Test (DMRT) test at a significant level of 5%.

III. RESULTS AND DISCUSSION

The results of analysis shows that Bambara groundnut seeds stored for one year and two years indicated that storage period affecting the quality of the seeds both in their viability, vigour and the physical quality of the seeds (Table 1). All the studied characters i.e. seed moisture content, final germination percentage, germination rate, maximum grow potential of seeds, dry weight root and shoot significantly decreased over some period of time.

Seed Moisture Content

All seeds from the promising line that have been stored for 1 year have slightly higher seed moisture content (Table 2) ranged between 12% to 14% with the highest water content recorded is TVSU 86 at 14.38% and the lowest water content is PWBG 6 at 12.94%. Moisture content in seeds stored for 2 years, ranged between 11% to 13% with the highest seed moisture content in seeds stored for 2 years recorded in BBL 1.1 (13.35%). All seeds from six lines of Bambara groundnut both stored after one year and two years has relatively high seed moisture content. High water content of seeds during storage can be caused by the relative humidity around the seeds. Seed is a hygroscopic entity and absorbs moisture from the surrounding, and any change in temperature and relative humidity of the environment, affects moisture contents and quality of the seeds [16].

Table 1. Analysis of Variance from studied parameters

Observation Parameters	Treatment		
	Period of Storage (P)	Promising Line (L)	P x L
Final germination percentage (%)	*	ns	*
Seed moisture content (%)	*	ns	ns
Germination rate (%/day)	**	ns	ns
Vigour index (%)	**	**	**
Maximum growth potential of seeds (%)	**	ns	ns
Dry weight root and shoot (g)	**	**	ns

Note: Note: *: $p < 0.01$; **: $p < 0.05$; ns: not significant

Table 2. Means of studied parameters

Treatments	FGP (%)	SMC (%)	GR (%/day)	VI (%)	MGP (%)	DW (g)
Period of Storage (P):						
1 Year (P1)	94^{a}	13.53^{a}	8.11^{a}	43^{a}	97^{a}	0.31^{a}
2 Years (P2)	87^{a}	12.93^{b}	7.44^{b}	22^{b}	86^{b}	0.18^{b}
Bambara Groundnut						
Lines (G):						

CCC 1.6 (G1)	88^{ab}	13.12^{b}	7.77	25^{b}	88^{ab}	0.25^{ab}
PWBG 6 (G2)	90^{ab}	12.74^{b}	7.94	22^{b}	90^{ab}	0.25^{ab}
PWBG 5.2.1 (G3)	83^{b}	13.19^{ab}	7.69	22^{b}	85^{b}	0.23^{b}
SS 2.4.2 (G4)	90^{ab}	12.93^{b}	7.68	25^{b}	90^{ab}	0.28^{a}
BBL 1.1 (G5)	97^{a}	13.31^{ab}	7.75	55^{b}	97^{a}	0.22^{b}
TVSU 86 (G6)	97^{a}	14.10^{ab}	7.83	48^{a}	97^{a}	0.21^{b}
P X L:						
P1xG1	93	13.57	8.07	40	93	0.32
P1xG2	97	12.94	8.29	30	97	0.33
P1xG3	97	13.16	8.48	40	97	0.28
P1xG4	93	13.89	7.89	47	93	0.35
P1xG5	93	13.26	7.96	60	100	0.28
P1xG6	93	14.38	7.95	43	100	0.28
P2xG1	83	12.66	7.47	10	83	0.18
P2xG2	83	12.53	7.59	13	83	0.17
P2xG3	70	13.23	6.91	3	73	0.18
P2xG4	87	11.96	7.46	3	87	0.21
P2xG5	100	13.35	7.53	50	93	0.16
P2xG6	100	13.83	7.70	53	93	0.15

Note: SMC= Seed moisture content; FGP= Final germination percentage; VI= Vigour Index; MGP= Maximum growth potential of seed; DW= Dry weight of root and shoot; GR= Germination rate.

Other factor affecting moisture content of the seed is improper drying at post-harvest process could result in high moisture content in seeds and later promotes deterioration of the seeds. Study at flowering dogwood seed [17] also showed that seed dried to 14% moisture content germinating poorly after 2 years and failing to germinate after 3 years in storage. SS 2.4.2 stored for 2 years is the only lines which has the lowest moisture content and close to the recommended moisture content for storage (10% or below) as reported by Bonner [18] that orthodox seeds can be stored for relatively long periods if their moisture contents are about 5 to 10%.

Final Germination Percentage

Final germination percentage was observed at first count (7th day) and final count (14th day). After one year period of storage all lines of Bambara groundnut seeds showed higher germination percentage. Higher percentage indicated that the quality of the seed remained high (Table 2). Final germination percentage of six lines are above 90%. PWBG 6 and PWBG 5.2.1 showed highest germination percentage and the rest showed the same result for their germination percentage (93%). During observation all seeds able to germinate to normal seedlings (Figure 1) and no abnormal seedling was recorded. High germination percentage highly associated with high viability of the seeds and recent harvested seeds also tend to have high viability. The viability of seeds is a critical and important factor for seed quality and closely related to resistance to biotic and abiotic stress, germination percentage, and plant performance [19]. For seeds stored after 2 years showed significant decrease of final germination percentage in one of the lines. PWBG 5.2.1 showed lowest final germination percentage (70%) and some seeds grow abnormally (Figure 2). Normal seedlings are seeds that able to germinate with complete structure with all of their essential structures well-developed (Figure 1). Abnormal seeds recorded in PWBG 5.2.1 with some essential structure failed to develop. After 14 days plumule failed to develop and root formed a loop. According to Vujosevic [20] abnormal seedlings are less likely to develop into normal plants. Lower germination and higher abnormality of seedling indicated that deterioration of seeds in Bambara groundnut most likely begin to occur on 2 years period of storage. [21] also stated higher number of abnormal seedlings indicated the deterioration of the seeds.

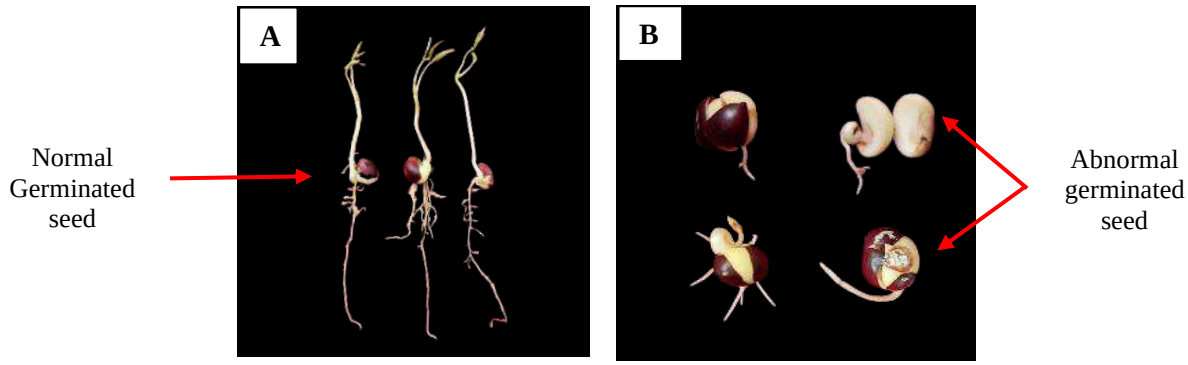

Fig. 1: Seed Germination in Bambara Groundnut: a) Normal germinated seed of Bambara Groundnut with complete essential structure. b) Abnormal growth of Bambara groundnut seed with deformed shape

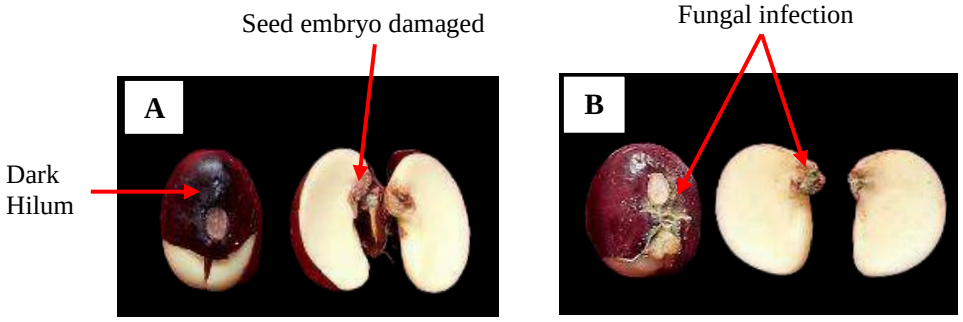

Fig. 2: Dead seeds of Bambara Groundnut: a) Bambara groundnut (PWBG 5.2.1) that unable to germinate stored on 2 years period of storage. b) Damaged embryo of seeds of Bambara Groundnut (PWBG 5.2.1). Stored on 2 years period of storage.

Germination Rate

The results showed significant different in germination rate between Bambara groundnut seeds stored on one year and two years (Table 1). Higher germination rate recorded in all lines of Bambara Groundnut stored for one year with highest germination rate are CCC 1.6 (8.07 %/day), PWBG 6 (8.29 %/day) and PWBG 5.2.1 (8.48 %/day) and SS 2.4.2, BBL 1.1 and TVSU 86 are 7.89 %/day, 7.96 %/day, 7.95 %/day respectively. Faster germination rate on seed stored on one year period of storage indicated that seeds most likely still remained in high viability and vigour and less deteriorated. Siregar [22] also reported, seed with high germination rate associated with decent seed viability.

After 2 years storage, seeds of Bambara groundnut showed slower germination rate with average 7.44 %/day. The rest of the lines showed germination rate above 7 %/day, CCC 1.6 (7.47 %/day), PWBG 6 (7.59 %/day), SS 2.4.2 (7.46 %/day), BBL 1.1 (7.5 %/day) and TVSU 86 (7.70 %/day). Lowest germination rate recorded in PWBG 5.2.1 (6.91 %/day). PWBG 5.2.1 also showed lowest germination percentage and this is the only lines that shows most visible decline of the quality of the seeds compared with all other types. Slower germination rate is one of the symptoms of deteriorated seed because of natural seed aging. Garoma (2017) also reported some parental seed of maize stored on longer duration resulted in delayed onset decreased the germination rate as well as germination index, slow seedling emergence and low weight of seedling traits [23].

Period of storage also show influence to vigour index of the seeds of Bambara groundnut and showed results of highly significant different between 2 period of storage (one year and two years). Vigour index observed by calculating the number of normal seed that able to germinate on the first count (7^{th} day). Vigour index of the seeds stored for 2 years shows most visible decline in most lines of Bambara groundnut. Vigour index ranges from 3% to 50% with lowest vigour index recorded in PWBG 5.2.1 (3%) and SS 2.4.2 (3%) followed by CCC 1.6 (10%) and PWBG 6 (13%). Low vigour can be caused by the storage environment of the seeds. Seed storage is very important to secure good quality seeds for planting materials. Seed longevity, vigour and viability depend on genetic and physiological factors as well as storage conditions. The

most important factors that influence storage are temperature, moisture, seed characteristics, micro-organism geographical location and storage structure [24]. Six promising lines of Bambara groundnut studied stored in plastic bag inside the fridges with temperature 14.4ºC and 40% RH**.** For long term seed storage these conditions could promote rapid seeds deterioration after 3 or 4 years because of the current relative humidity and the temperature. The temperature of fridges is cold, but it also very humid. Factors which influence the longevity of stored seed include harvesting condition, drying process and storing process but the most important is storage conditions which is temperature and relative humidity [25]. As seed stored in suboptimal storage condition and went through ageing phase, seeds physiologically changes including membrane damage and loss of enzymatic activity [26] these physiological changes later contribute to decrease seed vigour

Maximum Growth Potential of Seeds

In this study period of storage does not greatly affecting the maximum growth potential of seeds. Low maximum potential of seeds caused by the high number of seeds that unable to germinate because of severe damaged embryo, hard seed coat and prevent imbibition or fungal infection (Figure 2b) this is linked to post-harvest process and materials used for storing the seeds. Improper post-harvest process and materials to store the seeds may cause seeds damage and further invested with fungi. Taghfir (2018) also reported, the seed container or packaging has an effect on maximum growth potential, germination, growth speed and seed vigour index [27]. Six lines of Bambara groundnut studied, stored in a plastic bag. This material has possibility to create small hole and can be an entry point for pests in seed storage or may promotes fungal infections on the seeds. In order to prevent this, seeds better to be stored in a closed container that is not easily perforated or damaged and it also keep seed humidity low and prevent the seed water content from increasing due to increased humidity [28]. Seed must be stored in moisture-proof containers and stored dry and kept dry to keep the longevity, viability, and vigour of the seeds.

Dry Weight Root and Shoot

Average dry weight of root and shoot from six lines of Bambara groundnut also have different results between two period of storage. Normal germinated seed on one year period of storage recorded 0.307 (gr) and higher than average dry weight of normal germinated seed on two years period of storage (0.176 gr). This result is likely influenced more by seeds vigour and germination rate of the seeds linked with accumulation of dry matter. According to Siregar (2018) Normal seedling biomass indicates abundant nutrient content stored in the plant. High vigour in the seeds contribute to supply energy or the seeds and allow seeds to have enough energy to germinate and supply nutrient stock during the germination process [22], [29], [30].

Physical Change on Seeds

Figure 3a show example of Bambara groundnut seed stored for one year (one year period of storage). The surface structure of the seeds was smooth and the colour of the seeds did not show any change. The colour of seed hilum appears bright and clean. Some of Bambara groundnut seeds stored after 2 years show a little change physically but there were also many damaged seeds found.

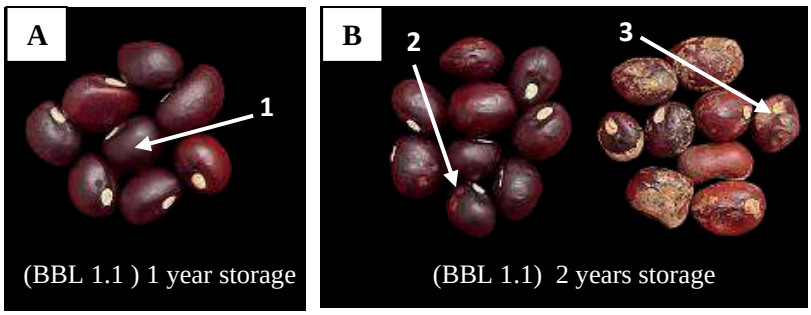

Fig. 3: Bambara Groundnut Seed Stored After 2 Years. Smooth surface of the seed. Discoloration on the surface of the seed (2). Seed Completely damaged (3).

Figure 3b show example of Bambara groundnut seeds on two years period of storage. Seeds shows little change in the surface structure of the seeds, The surface structure of the seed begins to change with the appearance of wrinkles and discoloration in some parts of the seeds but the colour of the hilum was still bright and clean. Figure 3b show example of damaged Bambara groundnut seeds on two years period of storage. Damaged seeds experienced changes in the colour of the seeds to brownish. The seeds become wrinkled and the surface of the seeds becomes rough. The colour of the hilum becomes brown and the seed coat begins to peel. Over period of times Bambara

groundnut seeds went to some natural aging phase and begin to show symptoms of deterioration both physiologically and physically and influence decline in the quality of the seed in terms of their viability, vigour and their ability to germinate. Copeland (2001) also explained that some processes occur in seed deterioration are colour changes in the seeds and the number of died seeds [31], [32].

Pearson Correlation

Pearson correlation between each studied parameter shown in Figure 4. Showed positive correlation among parameters (SMC=seed moisture content, FGP=Final Germination percentage, VI=Vigour Index, MGP=Maximum growth potential, DW=dry weight root and shoot, GR=Germination rate).

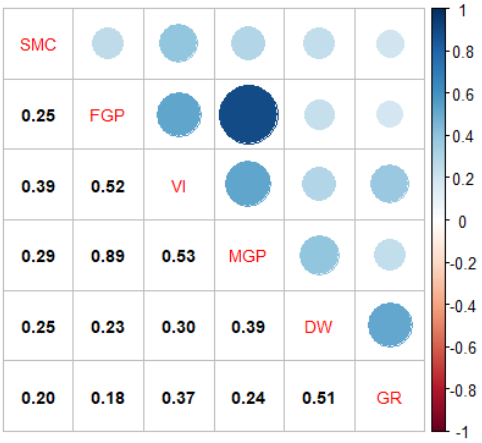

Fig. 4: Pearson Correlation Analysis for Each Parameters

The observation made in this study showed that final germination percentage have positive correlation with seed moisture content (r = 0.25), vigour index (r = 0.52), dry weight root and shoot (r = 0.23), germination rate (r = 0.18) and highly positive correlation with maximum growth potential (r = 0.89). Vigour index shows positive correlation with seed moisture content (r = 0.39), final germination percentage (r = 0.52), maximum growth potential, dry weight root and shoot, (r = 0.30) and germination rate (r = 0.37). Weak correlation found between seed moisture content and germination rate (r = 0.20), final germination percentage with germination rate (r = 0.18), and final germination percentage with dry weight root and shoot (r = 0.23). The further findings in this correlation analysis explain why the decline of germination capacity may have also caused decline in vigour index, germination rate and dry weight root and shoot as there's a positive correlation was found between the parameters. This result describes and emphasized that negative effect of seed deterioration will have an impact on other physiological quality of the seeds.

Germination percentage of seeds highly correlated with maximum growth potential of seed as both parameters observed similar thing but maximum growth potential observed both normal and abnormal seedling. Germination rate have weak correlation with most of the parameters showed that germination rate may not be influenced only with the decline of other parameters but mostly influenced by environmental factors where seed were grown. Temperature and moisture levels are proven to influence the rate of germination [33].

IV. CONCLUSION

Six promising lines of Bambara groundnut stored for 1 year period of storage showed better germination in their ability, viability, vigour and still maintained optimum seed quality. For planting materials seed stored for 1 year suitable for cultivations. This result followed by seeds stored for two years. After two years seeds of six line of Bambara groundnut seed still maintained the quality of the seed but showed some decline in some germination characters and greatly affecting their vigour and germination rate. After 2 years stored, seeds began to show symptoms of deterioration both in their germination ability and also physical deterioration symptoms also occurred. This decline and deterioration process may cause by high moisture content of seeds when seeds stored and later accelerate because of the environment condition of the storage chamber (low temperature but high relative humidity).

In conclusion Bambara groundnut seeds can be stored for 2 years while still maintaining their quality with some seeds showed symptoms of deterioration. Maximum storage period for Bambara groundnut may reached 3 years or more if the seeds stored in lower temperature and lower relative humidity so it can delay or prevent accelerate severe seed deterioration.

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Effect of intercropping wheat (*triticum aestivum.L.*) with mustard (*brassica juncea*) on yield and economics under organic system of cultivation

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Abstract— *Wheat is the most important food grain among the cereal grain crops. In India, wheat demand increases because of an increase in population and diet changes among the people.and wheat occupies 2nd place over the rice. The research was conducted with the aim of checking out the suitable row proportion while wheat was intercropped with mustard under different organic manures and biofertilizers. The field experiment was conducted at the farm of Lovely Professional University, Phagwara, on wheat (Triticum aestivum.L) intercropped with mustard (Brassica junecea.L) during the rabi season in the years 2020–2021. A split plot block design was used with three row proportions of wheat + mustard intercropping (3:1) (2:2) and sole wheat (the main crop),and four treatments of different manures and biofertilizers, and three replications. Based on the complete analysis of the experimental results, it is concluded. that different treatments affected the wheat + mustard intercropping, there was a significant increase in growth parameters, yield attributes, and yield. Among the geometries, G1 (sole wheat) showed good results in biological, economical, straw yield, and harvest index in M4 (vermicompost 5 t/ha + Azospirillum + PSB) in all geometries. There is no significant increase in harvest index in all geometries and treatments. Higher gross returns were absorbed in (3:1) wheat + mustard intercropping in the treatment (G3M4 Vermicompost 5t/ha + PSB + Azospirillum), and the highest net returns were increased was observed in Geometry 2 (2:2 wheat + mustard) of treatment no:3 (M3 poultry manure 5t/ha + PSB + Azospirillum). The highest benefit-cost ratio (2.27) was observed in Geometry 2 (2:2 wheat + mustard intercropping) of treatment M3 (poultry manure + azospirillum + psb)).Hence, research outcomes (2:2) of wheat + mustard intercropping by applying M3 poultry manure along with biofertilizers are beneficial to farmers.*

Keywords— ***Economics, organic manures, biofertilizers, Row ratio, wheat + mustard intercropping, sole wheat, geometries, beneficial, farmers.***

I. INTRODUCTION

Intercropping plays a major role in organic cultivation. this type of cultivation, intercropping involves between two or more crops with row arrangements and different types of patterns at the same time and same field. Intercropping is noted as a useful agronomical practice to increase the yields and quality of the environment in the area through agricultural production. Intercropping of wheat and mustard is an old age practice mainly in the northern part of India for the purpose of stability and the necessity of oil and grains both. now adays intercropping may be raise due to produce higher yields than monocropping or sole cropping.Row ratios in intercropping play a major role in wheat and mustard. In recent times, row intercropping has produced higher yields than mixed cropping. recommended row ratio to be preferred for the farmers to attain higher yields from available resources more effectively on a sustainable basis. With variation in row ratio, growth and development of both

crops are also being diverted,which eventually affects the yield attributes or yield,but aparticular crop ratio LER and yield profit surely increase. Thus, recommendations have been made by scientists for various locations or areas. Due to the change in various weather conditions, climatic changes may vary from place to place, as may the types of cultivation practices and cultivation varieties. The research analysis is satisfied with wheat + mustard intercropping in relation to management of irrigation, fertiliser recommendations,genotypes, and crop geometries. Hence, the upcoming research must focus on studies of crop competition on row ratios,growth and development studies on intercropping, and mainly yield attributes and advantages of intercropping by using agro-techniques to get more yields. In organic farming, manures plays a major role. Manures are nutrient-rich plant and animal wastes. During decomposition, they emit nutrients. the art of gathering and using waste from livestock, human, and 14 vegetable sources to improve crop production. Manures are organic materials made from animal, human, and plant waste that produce diverse plant nutrients.

II. LITERATURE REVIEW

Intercropping is also described as the cropping method in which two or more crop species are grown simultaneously in the same area, with crops having the same growing season [Ofori and Stern, 1987].

Intercropping systems avoid the risk of various pests and diseases. It also enhances the absorption of sunlight, fertilisers, and water absorption. The rate of absorption of sunlight, water, and fertiliser is greater in amounts when compared to a sole crop. Intercropping also helps in overcoming the unfavourable climatic conditions in areas with uneven environmental conditions. (Rathi and Verma, 1979)

Intercropping is more beneficial than sole crops, as sole crops do not make use of all the available resources. In the intercropping system, the selected crop species are sown in parallel in order to increase crop production. There will be an upsurge in crop production with space utilisation and time management. (Ahlawat and Sharma, 2002).

In order to obtain greater yield benefits, a suitable row ratio and distance should be required in the management of intercropping in wheat and mustard, as aerial competition arises between these two crops when they are grown together at various sowing amounts. (Bora,1999)

In order to increase the yield in intercropping, the agronomic study of the arrangement of plants and also the total number of plants and number of plants in component crops is required (Willey, 1979b).

In addition, Saini et al. (1989) also reported that growing one row of mustard with eight or nine rows of wheat increased the yield of mustard seeds by about 2–3 quintals per hectare without damaging the yield of wheat.

III. MATERIALS AND METHODS

An experiment was conducted at the field Department of Agronomy, school of Agriculture, Lovely Professional University, Phagwara, on wheat + mustard intercropping during late November in the year 2020-2021 with the title —"To study the effect of intercropping wheat (Triticum *aestivum*) with mustard (Brassica *juncea*) on yield and economics under an organic system of cultivation".

The experiment was laid out in a split-plot design comprising three row proportions of wheat + mustard intercropping, i.e., 3:1, 2:2, and sole wheat, in main-plots, and four different treatments of manures, along with two biofertilizers of farmyard manure,poultry manure,vermicompost, azospirillum, and phosphate solubilizing bacteria (PSB) in sub-plots. Thus, total twelve {(3 main-plots x 4 sub-plots)} treatment combinations were formed. The treatments were allocated to each plot randomly using a random number (Fisher and Yates, 1963) and replicated three times. The main plot and subplot treatments, as well as the sole crop (wheat), were also randomized separately using the said random number. Details of the treatments and their combinations used in the experiment are given in the below table.

Main plot:(crop ratio)

S. No	GEOMETRY
G1	(100% Wheat)sole wheat
G2	50 % wheat + 50% mustard **(2:2)**
G3	75 %wheat + 75% mustard **(3:1)**

Sub plot: Four different treatments of manures along with two biofertilizers

Sub plots	Treatment
M1	Control
M2	FYM + azospirillum & psb
M3	Poultry manure + azospirillum & psb
M4	Vermicompost + azospirillum & psb

Experimental site location:

Experiment was conducted at the farm of lovely professional university Phagwara district kapurthala late forth november in year 2020-2021. The farm is

situated at ^{0}3122'31.81'' North latitude and ^{0}7523'03.02‖ East longitude with 252m average elevation above mean sea level. It is at 350 km distance from capital of India (Delhi) in Punjab fall under sub tropical region in central plane of state agro climatic zone.

Weather and climatic condition:

Region of experimental site comes under sub tropics with cool weather in winter season, hot weather in summers and distant rainfall period in month of July, August and September. South west monsoon is main source of rainfall in this region. During winter season the temperature never goes below zero degree especially in the month of December and January. Highest temperature reached 42^{0}c during summer month April, May and June.

1.Sole Wheat

2. (Wheat + mustard intercropping (2:2) Ratio

3. (Wheat + mustard intercropping (3:1) Ratio

Meteorological data of growing season

Month	Temperature			Relative Humidity (%)	Rainfall (mm)
	Min	Max	Avg		
Nov (2020)	11.5	25.1	18.3	69.2	15
Dec (2020)	7.5	20.6	14.05	69.8	21
Jan (2021)	6.1	18.5	12.3	75	48.4
Feb (2021)	8.5	22.2	15.35	67.7	74
Mar (2021)	14	29	21.5	61.5	62
Apr (2021)	18	34	26	38.5	38

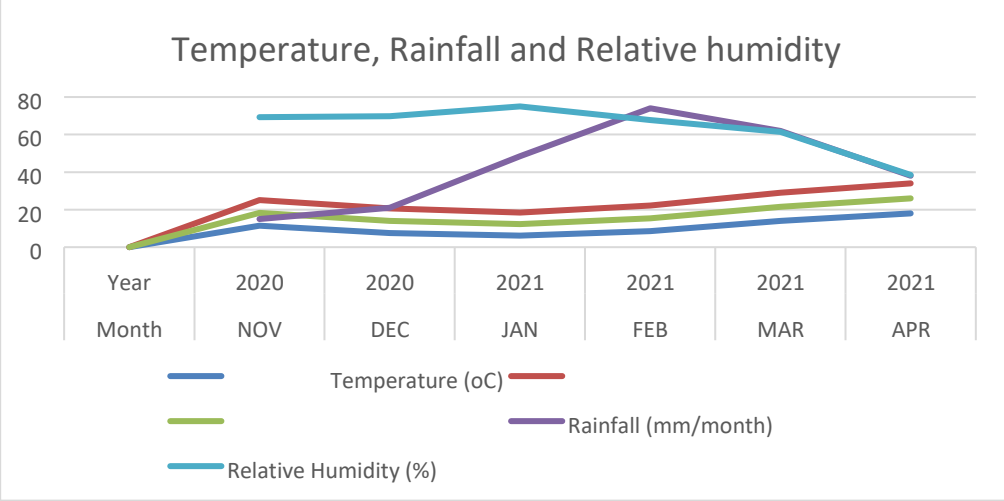

Fig.1 Temperature rainfall and rainfall and relative humidity

IV. RESULTS & DISCUSSION

Table.1 Effect of row proportions, and different types of manures on yield of wheat.

GEOMETRIES	Biological yield	Grain yield	Straw yield	Harvest index
Row proportions				
Sole wheat	126.2	53.45	72.40	42.25
2:2 wheat + mustard	73.16	30.85	42.30	42.20
3:1 wheat + mustard	88.58	37.84	50.74	42.80
SE(m±)	1.426	0.749	0.204	0.942
CD (P=0.05)	5.75	3.01	4.85	NS
Treatments				
Control	77.55	32.92	44.10	42.60
Fym + azospirillum+psb	98.0	41.21	56.78	42.07
poultry + azospirillum +psb	101.4	43.21	58.23	42.56
vermicompost+azospirillum+psb	107.0	45.52	61.47	42.43
SE(m±)	1.091	0.728	0.257	0.997
CD (P=0.05)	3.26	2.18	3.76	NS

Grain yield (q/ ha^{-1})

A critical examination of the data on wheat grain yield was presented in the above table 1. While comparing the treatments,the grain yield was higher in (M4) (Vermicompost + azospirillum + Psb) than in (M_3)(poultry manure + azospirillum + Psb) & (M_2)(fym + azospirillum + psb).Hence, all the treatments recorded significantly higher grain yields than the control treatment. Different row proportions exhibited perceptible variation in the wheat grain yield in Intercropping of wheat + mustard in (G_1) (sole) row proportion significantly recorded maximum grain yield over the both row proportions of (3:1)(wheat + mustard) & (2:2)(wheat + mustard), respectively; however, (3:1)(wheat + mustard) recorded significantly higher than (2:2)(wheat + mustard),and lower than G_1(sole). However, the lowest grain yield was observed in the (2:2)(wheat + mustard) row proportion. Hence, wheat in a pure stand recorded a significantly higher grain yield than the intercropping mean.

Table.2. Effect of row proportions, and different types of manures on gross returns, net returns and benefit cost ratio in wheat + mustard intercropping.

GEOMETRIES	Gross returns	Net returns	Benfit cost ratio
Row proportions			
Sole wheat	1,21,491.60	53,789.00	1.792
2:2 wheat + mustard	1,36,825.40	69,122.84	2.032
3:1 wheat + mustard	1,24,689.10	56,986.58	1.843
SE(m±)	1,516.76	1,516.78	0.023
CD (P=0.05)	6,115.01	6,115.09	0.093
Treatments			
Control	1,04,173.30	46,083.22	1.793
Fym + azospirillum+psb	1,29,173.70	59,933.67	1.866

poultry + azospirillum +psb	1,34,836.60	70,596.45	2.099
vermicompost+azospirillum+psb	1,42,491.30	63,251.22	1.798
SE(m±)	1,388.90	3,033.56	0.021
CD (P=0.05)	4,158.59	8,067.45	0.062

Economics

B:C Ratio

Benefit cost ratio is the key factor that denotes the how the farmer is benefited from his input application to the field for crop production and how the returns are obtained. If the returns are more as to cost of cultivation. Better the B: C ratio more the benefit resulted. Benefit cost ratio have shown significant variance among geometry treatments.

From the data obtained highest benefit cost ratio (**2.27**) were observed in Geometry 2 (2:2 wheat + mustard intercropping) of treatment no. M3 (poultry manure + azatobactor + psb)) and within the geometry's G1M3, G3M3,G2M1 and G2M2 (2.26, 2.20,2.19 and 2.10) have shown highest B: C ratio as to other treatments within the geometry's. In geometry treatments comparison of G1 (sole wheat) while comparing with control plot G1T1 (1.69) all treatments G1M3,G1M2,and G1M4 (2.26,2.05,1.83) have shown better B: C ratio. In geometry 2 (2:2 wheat + mustard) while comparing with control G2M2 and G2M4 shows lesser bc ratio.and G2M3 were heighest bc ratio among all the treatments in a geometry. In geometry 3 (3:1 wheat + mustard) control plot G3M1 (1.86) other treatments G3M3 and G3M4 (2.20 and 1.95) have shown better B: C ratio while G3T2 (1.82) have shown lowest B: c ratio as to control plot. Among total G1M1, G3M2, G1M4 and G3M1 have shown lowest B: C ratio.Hence among all geometries and treatments the lowest bc ratio were recorded in G1M1(control) and significantly highest bc ratio were recorded in G2M3(2:2 poultry + azospirillum + psb) and which was statistically par at G1M3 sole(poultry + azospirillum + psb). Benefit cost ratio of different geometry treatments were shown in above table 2.

V. CONCLUSION

By applying manures and biofertilizers in wheat + mustard intercropping, there was a significant increase in spike length, spike weight, number of spikelets spike^{-1}, number of grains spike^{-1}, number of siliqua/seed, stover yield, biological yield, yield, harvest index, and 1000 seed weight. There was no significant increase in all geometries. By applying of (M4 vermicompost+azospirillum +psb)there was a significant increase compared to the control. There was a further significant increase when applied (M3 poultry manure+ azospirillim + psb). There was no significant increase in both the geometries and all treatments of harvest index for both the crops. There was a significant increase in yield in (M4 vermicompost + azospirillum + psb) in all geometries of wheat + mustard intercropping for both crops.

Economics: Higher gross returns were observed in the (3:1) wheat + mustard intercropping (**Rs.1, 55,089 /ha**) were observed in the treatment (G3M4 Vermicompost 5t/ha + PSB + Azospirillum) and the highest net returns was increased in (**Rs.81,809/ha**) were observed in Geometry 2 (2:2 wheat + mustard) of treatment no 3 (M3 poultrymanure 5t/ha + + PSB + Azospirillum). The highest benefit-cost ratio (**2.27**) was observed in Geometry 2 (2:2 wheat + mustard intercropping) of treatment no. M3 (poultry manure + azatobactor + psb)).Hence, (2:2) wheat + mustard by applying of poultry manure along with biofertilizers is beneficial to farmers.

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The effects of brine concentrations on the drying characteristics and microbial quality of dried fillets of African Catfish *(Clarias gariepenus)*

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Abstract— *This study evaluates the drying characteristics of brined fillets of African catfish (Clarias gariepenus) at four brine concentrations (0, 10, 15 and 20%) and four drying temperatures (40, 50 60 and 70^o C). Fresh catfish samples were obtained from a farm at Ikorodu, Lagos. They were cleaned, gutted and cut into fillets of approximately 5×4×3 cm sizes and soaked in the different concentrations of salt (NaCl) solutions for about 6 hours. Thereafter, the fillets were placed on trays and dried in a cabinet dryer till no appreciable changes in weight of the samples were observed. The drying data obtained were analyzed and employed to construct the drying curves, obtain the drying rate constants, the diffusion coefficients and activation energy. Drying rate increased with drying temperatures, with the fillets experiencing greater moisture loss at the initial stage of drying. Fillets dried at temperatures higher than 60 ^{o}C acquired the aroma of cooked fish. Both the drying rate constants and diffusion coefficients increased with increasing temperature. Microbial analysis revealed that the higher the salt concentration, the lower the microbial load on each of the dried samples at the different drying temperatures. The microbial load -population of total viable count recorded in this study varied from 4.50 × 10^5 cfu/g (70^{o}C, 20% salt concentration) to 3.08 × 10^7 cfu /g (40^{o}C, 0% salt concentration). Thus, drying temperature and salt concentration has profound effects on the drying rates as well as the microbial load of the dried catfish.*

Keywords— ***catfish, drying, salt, drying rate, diffusion coefficient.***

I. INTRODUCTION

In Nigeria, the African catfish (*Clarias gariepenus*) is the one of the main fish species commonly stocked in ponds and one of the few that has demonstrated commercial viability (Olagunju *et al.*, 2007, Banjo *et al.*, 2009; Alawode and Ajagbe 2020, Ogah *et al.*, 2022). As noted by Onoche *et al.*, (2020), and Kaleem and Sabi (2021), a major production constraint is the high post-harvest losses recorded by farmers particularly during gluts. Appropriate preservation methods that significantly reduce the loss, including those occurring during distribution and marketing are therefore essential. Moisture control primarily by drying provides opportunity to prevent losses, which occur during harvesting, handling and storage.

Natural sun drying has been used for preserving fish catches, however, the limitations include lack of control of the drying process, weather uncertainties, high labour costs, insect infestation, and mixing with dust and other foreign materials. As a result, drying in which the drying heat is artificially provided is finding increasing applications for fish products. Salting and drying achieve a lowering of the water activity (a_w) of the fish flesh and hence an extension of the shelf life. Many researchers have studied conventional fish salting and drying methods (e.g Sen and Lahiry (2004) and Sen and Sripathy (2007)). Sobukola and Olatunde (2011) studied the effect of salting techniques on salt on uptake and drying kinetics of African catfish (*Clarias gariepinus*). Sankat and Mujaffar (2006) presented a report on the drying behaviour of salted fillets

of catfish (*Arius sp*). The optimal drying conditions for the tropical African catfish species do not appear to be well established yet, particularly, the kinetics of water removal from salted fish fillets. The objectives of this study were therefore to study the drying behaviour of catfish fillets dried in a cabinet oven at different temperatures ranging from 40°C to 70°C and to determine the microbial load of the fillets after drying.

II. THEORETICAL CONSIDERATIONS

Moisture ratio during drying experiment is usually calculated using the equation (Vallero *et al*.,2023).

$$MR = \frac{M_t - M_e}{M_o - M_e} \quad . \qquad \ldots\ldots\ldots\ldots (1)$$

With the drying rate calculated using equation (2)

$$Drying\_Rate = \frac{M_{t+dt} - M_t}{D_t} \qquad \ldots\ldots\ldots (2)$$

The effective moisture diffusivity is calculated by using the simplified Fick's second law of diffusion:

$$\frac{\partial M}{\partial t} = D_{eff}\nabla^2 M \qquad \ldots\ldots\ldots\ldots (3)$$

The solution of Fick's second law in slab geometry, with the assumption that moisture migration was caused by diffusion, negligible shrinkage, constant diffusion coefficient and temperature was given as (Vallero, 2023):

$$MR = \frac{M_t - M_e}{M_o - M_e}$$

$$= \frac{8}{\pi^2}\sum_{i=1}^{n}\frac{1}{(2n-1)^2}\exp\left(\frac{-(2n-1)^2\pi^2 D_{eff}t}{4H^2}\right)$$

$\ldots\ldots\ldots$ (4)

Where D_{eff} is the effective moisture diffusivity (m^2s^{-1}), H is the half thickness of the slab (m) and n is a positive integer. Since only the first term of the equation can be used for long drying time (Lopez *et al.*, 2000) the equation becomes:

$$MR = \frac{8}{\pi^2}\exp\left(\frac{-\pi^2 D_{eff}t}{4H^2}\right) \qquad \ldots\ldots\ldots (5)$$

III. MATERIAL AND METHODS

3.1 Samples preparation

Catfish *(Clarias gariepinus)* samples were obtained from the Lagos State Polytechnic fish farm at Ikorodu. Upon capture the fish were cleaned, gutted and then spitted into half and filleted. They were immediately cut into 5cm by 4cm by 3cm fillets using filleting knife. Brine solution made from food-grade sodium chloride dissolved in distilled was used. The fillets were soaked in various salt concentration range (0, 5, 10, 15 and 20%) in separate bowls for a period of 6 hours to allow adequate salt absorption and to stop normal bacteria spoilage.

The brined fillets were spread in single layer on pre-weighed shallow wire trays. Seven samples per salt concentration level were placed in different trays for each temperature setting of the Gallenkamp 300 plus cabinet oven. At regular intervals, the samples were taken out quickly from the oven, cooled in desiccators, weighed and return to the dryer. Drying was continued until moisture content reduces to safe storage moisture content. The procedure was repeated at a temperature of 50°C, 60°C and 70°C respectively. From these, the average value moisture loss as a function of time was determined and use to construct the drying curves.

A 2.5g representative sample was obtained aseptically from the muscle (the thickest part of the muscle) of the cat fish steak and crushed using a lab pestle and mortal. Serial dilutions (10^{-1}–10^{3}) of the homogenized samples were made using 25mls distilled water. Briefly, serial dilutions of the homogenized samples (10ml) in duplicate were passed through a 0.45 mm grid membrane filter, which was subsequently placed onto an absorbent pad soaked with appropriate broth in a Petri-dish, and incubated at 35°C for 24–48hrs.

IV. RESULTS AND DISCUSSION

4.1 Air drying curves

The drying data obtained were used in constructing the drying curves for salted catfish fillets dried at 40, 50 60 and 70° C. The drying rate curves for the air drying of salted catfish fillets are shown in Figures 2 (a-d). The figures revealed that drying rates were higher during the first two hours of drying, when the moisture content was greater. An increase in rate constant with increasing drying air temperature has been shown for the drying of many biological materials (Krokida *et al.*, 2004; Mujaffar and Sankat, 2005). Drying at a higher temperature resulted into a greater movement of fat from the within the muscle to the surface of the fillets. Drying at 70°C also resulted in hard and crusty fillets. At 20% salt concentration a thick layer of salt on the surface developed which impeded further drying. Salted fillets dried at 40°c appeared moist and later developed off- odours.

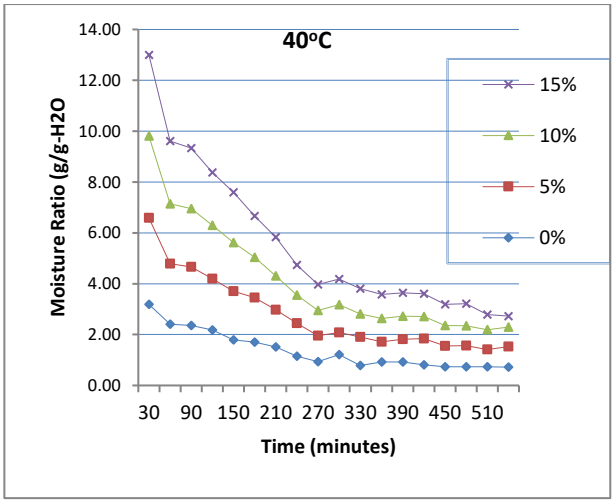

Fig 1a. Drying curves for fish at 40^{0}C and varying salt concentration

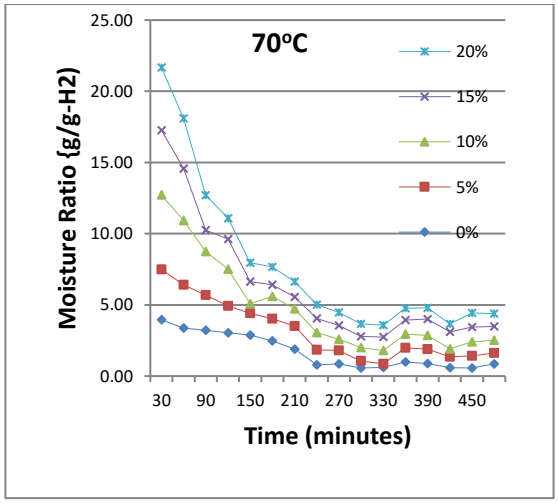

Fig 1d Drying curves for fish at 70^{0}C and varying salt concentration

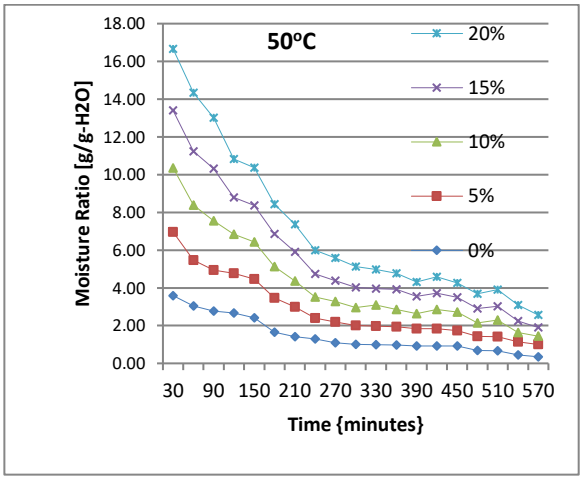

Fig 1b. Drying curves for fish at 50^{0}C and varying salt concentration

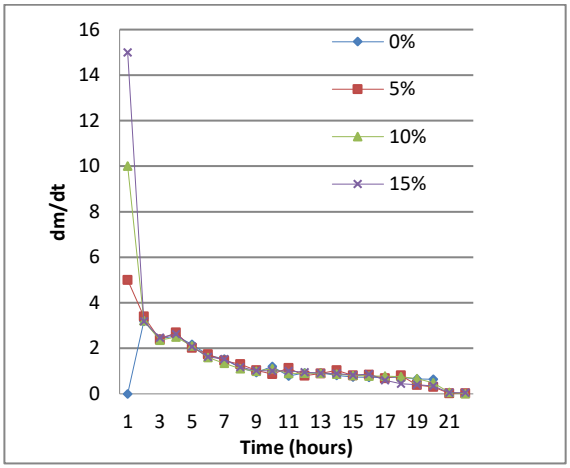

Fig 2a. Drying rate curves for fish at 40^{0}C and varying salt concentration Time (hours)

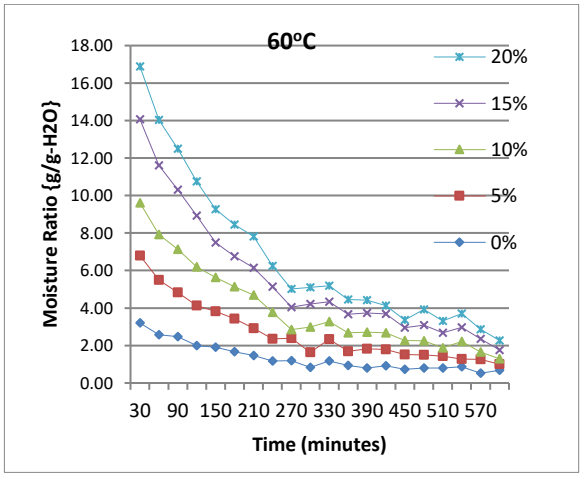

Fig 1c. Drying curves for fish at 60^{0}C and varying salt concentration

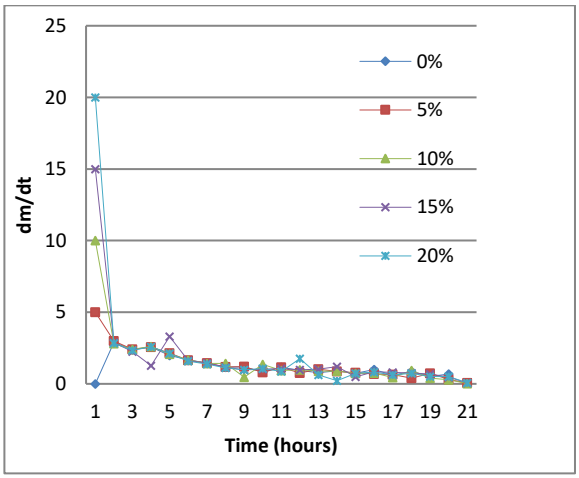

Fig 2b. Drying rate curves for fish at 50^{0}C and varying salt concentration

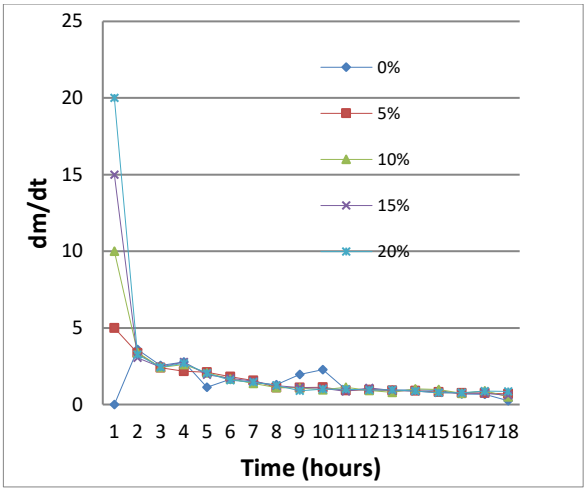

Fig 2c. Drying rate curves for fish at 60^0C and varying salt concentration

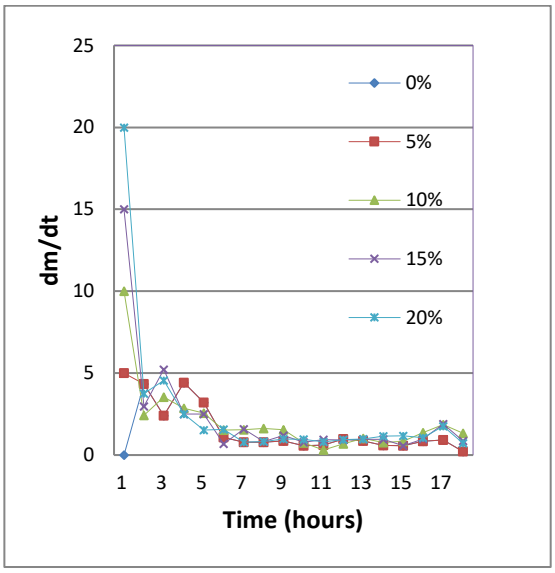

Fig2d. Drying rate curves for fish at 70^0C and varying salt concentration

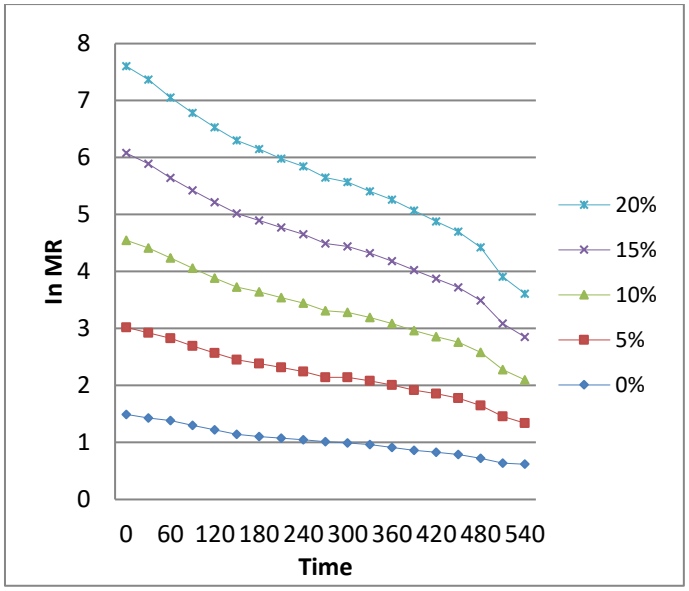

Fig.3: Plot of Ln MR against time for salted catfish dried at 60^0c

4.2 Drying constants, diffusion coefficient and activation energy

It is generally accepted that moisture is removed from salted fish during the falling rate period during which the rate is governed by the transfer of water by diffusion. (Jason, 1998; Wheaton and Lawson 2000; Ismail and Wooton 2002). The drying constant (k) is obtained from the slope of the plot of In MR versus time and is shown in Table 1. Figure 3 presents a plot of In MR against time for 60°C.

The drying rate constants for salted fillets were used to calculate diffusion (D) using the following equation:

$$A=\pi^2 \, D/L^2$$

where A is the diffusion area. The temperature dependence of the D-values for salted fillets and the activation energies were estimated from a plot of In D versus I/T using an Arrhenius type equation:

$$\mathrm{In}D = \frac{-\mathrm{E_a}}{\mathrm{RT}}$$

The values obtained are presented in Table 1 above.

Table 1: Values of drying constants, Diffusion coefficient and activation energy at different salt concentrations and drying temperatures

Drying Temp	Salt concentration	K-value $\{\frac{Y_1 - Y_2}{X_1 - X_2}\}$	Diffusion coefficient $\{D = \frac{4KL^2}{\pi^2}\}$	Activation energy calculated from $In\ D = \frac{-Ea}{RT}$
40°C	0%	0.0013	0.013	12384.3
	5%	0.0012	0.012	12612.6
	10%	0.0012	0.012	12612.6
	15%	0.0011	0.011	12860.7
	20%	0.0010	0.010	13132.5
50°C	0%	0.0013	0.013	11660
	5%	0.0013	0.013	11660
	10%	0.0012	0.012	11877.2
	15%	0.0011	0.011	12110.8
	20%	0.0010	0.010	12366.8
60°C	0%	0.0015	0.015	10926.9
	5%	0.0014	0.014	11106.5
	10%	0.0013	0.013	11299.1
	15%	0.0012	0.012	11509.4
	20%	0.0012	0.012	11509.4
70°C	0%	0.0016	0.016	11412
	5%	0.0015	0.015	12131.8
	10%	0.0014	0.014	11798.4
	15%	0.0014	0.014	11798.4
	20%	0.0013	0.013	12002.1

4.3 Microbial quality: total viable count

The total viable count (TVC) is one of the indicators for the quality of dried fish. The population of TVC recorded in this study varied from 4.50 × 10^5 cfu/g (70°C, 20% salt concentration) to 3.08 × 10^7 cfu /g (40°C, 0% salt concentration). The unsalted sample, dried at low temperature was confirmed spoilt. As stated by Broekeart *et al.*, 2011, loads of 10 7 -10 8 CFU/g make spoilage organoleptically detectable. The effects of temperature and salt concentration on the microbial load of the dried catfish was thus evident.

V. CONCLUSION

The drying characteristics of brined African catfish fillets were determined in this study. From the experimental result, under the same condition (temperature and salt variation), further increases the temperature to 70^0c resulted in case hardening of the fillets. It was also observed that as the salt concentration is increased at each drying temperature the microbial load decreases, but greatly with increasing temperature. Hence, salt serves as an inhibitor to the growth of micro-organisms and can be use in the preservation of fish. The results provide relevant information procedure for catfish processor, drying handling, modeling and designing of an over for drying fish.

NOMENCLATURE

∂M	change in moisture content
∂t	change in time
χ^2	Reduced chi-square
D	diffusion coefficients cm^2s^{-1}

D_{eff}	effective moisture diffusivity (m^2s^{-1})
D_o	Arrhenius equation constant (m^2s^{-1})
Ea	activation energy $Jmol^{-1}$
H	half thickness of sample
k	drying rate constant (h-1)
M R	moisture ratio
MC	moisture content any time (kg water/kg dry matter)
M_e	equilibrium moisture content (kg water/kg dry matter)
Mo	initial moisture content g H2O/g DM
M_o	initial moisture content (kg water/kg dry matter)
N	number of observations
R	gas constant 8.314 JK-1mol-1
R^2	coefficient of determination
RMSE	root mean square error
t	drying time (min or h)
T	process temperature K
W_r	reduced weight of samples after drying
W_s	initial weight of samples before drying
Z	number of constants in the model

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Municipal Solid Waste Landfill as a Dangerous Ungovernable Biochemical Reactor

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Abstract— *The paper discusses a critical dangerous situation regarding unequipped municipal solid waste (MSW) landfills which take place in virtually all developing economics, as well as in many richer countries. In this article, MSW landfills are considered as uncontrolled biochemical reactors that generate dangerous air, water and soil pollutants and negatively affect the health of the population for tens and even hundreds of kilometers around. To this purpose, we have comprehensively examined four operating solid waste dumps in a large industrial city. The so-called "landfill gas" is a 99% greenhouse gas (its composition is a mixture of CO_2 and CH_4), that is, MSW landfills also make a considerable contribution to global warming. In additional, self-ignition and smoldering of municipal waste inside the MSW landfills giving very dangerous pollution of the environment by their "flue gases" have been studied. Experimentally studied bacterial activity in the body of the MSW landfill which largely determines the "behavior" of the MSW landfill. Also, a technology has been developed to suppress smoldering and burning inside MSW landfills.*

Keywords— ***municipal waste, unequipped landfill, environmental pollution, biogas, toxic metals, landfill microorganisms.***

I. INTRODUCTION

A large number of countries in the world (mainly in Africa, Asia and South and Central America) dispose their unsorted MSW in unequipped landfills where is only a bulldozer that levels and compacts the MSW layers (from about the original 250 to the final 600 kg/m^3) [1]. The municipal solid waste (MSW) management is a particularly critical problem for countries with "developing" economics [2]. Ukraine, like about other 150 countries in the world, use non-equipped (at best, poorly equipped) MSW landfills. Such poorly organized landfills, and sometimes just "spontaneous" dumps everywhere arise around cities (especially large ones) around the world. The no rules for the placement and disposal of waste are not observed there: don't have preliminary sorting MSW; the bottoms of the huge open pits are not equipped; there are no protective dams around; the elementary technology of MSW storage (layer-by-layer isolation with soil) is not observed; there are no ditches for diversion of flood and rain water; "landfill gases" and poisonous filtrate are not captured, and even control wells for their analysis are not provided for; wheels of departing garbage trucks are not disinfected; etc. In addition, these landfills have constant hotbeds of smoldering These numerous and typical violations lead to serious negative consequences for the natural environment: pollution of open water bodies, groundwater, atmospheric air, fires, and the spread of infectious diseases [3]. In Brazil from 2003 to 2011 1.5 million tons per year of CO_2 (an average) were emitted into the atmosphere [4]. Mumbai (India) generates over 9000 t of municipal solid waste daily and disposes of most of it in open dumps [5]. But this problem also exists for economically developed countries. The so-called "Naples waste management crisis" of the 80s and 90s is well known, when tens of thousands of tons of MSW accumulated on the streets and outskirts of Naples in "wild dumps", many of which were set on fire [6].

For such unequipped landfills and even equipped landfills, biochemical processes and the role of bacteria in

them have been little studied. One of the most impressive studies on this problem has been described in article [7]: played the most important role Gammaproteobacteria, Firmicutes, Bacteroidetes, and Pseudomonas. Besides, over surface landfill air was found pathogenic microbes [8].

When MSW, after disposal, will press by a bulldozer, the supply of oxygen inside is weakened, therefore, anaerobic bacteria are activated [7, 9, 10, 11). First, hydrolysis cellulose less soluble compounds occur: $(C_6H_{10}O_5)n$ + nH2O = $n(C_6H_{12}O_6)$; the second stage is a biochemical decomposition of smaller compounds such as glucose into short-chained acetic or propionic acids, for example: $C_6H_{12}O_6$ = $3(CH_3COOH)$; and third stage is an anaerobic decomposition of VFAs into "simple gases": CH_3COOH = CH_4 + CO_2. The result of these biochemical processes is a production and emission of "greenhouse biogas" and other toxic gases (H_2S+SO_2, NH_3, NO+NO_2) from unequipped landfills. These "bacteria reactions" are exothermic, that increase the temperature inside MSW landfill up to 50 °C and often leads to smoldering and spontaneous combustion of an MSW [12].

In addition, to traditional toxic "flue gases", it has been shown that the maximum concentration of dioxins in the air can exceed the European Union standard of 0.1 nanogram/m^3 [13]. Therefore, MSW can cause significant damage to the environment if they are not stored in a properly engineered system. Typical problems that might occur are the following: emission of greenhouse biogas and other toxic gases, pollution of soil and ground water by highly toxic leachate [14], and also pollution of air by flue gases [15].

Thus, the purpose of this research was to provide a qualitative and quantitative estimation of the degree of environmental pollution by poorly equipped real MSW landfills as well as a theoretical and experimental study of bacterial activity inside the solid waste landfill.

II. MATERIALS AND TECHNIQUES

Note. Estimating the average inaccuracy of MSW experiments is a challenge. Firstly, the composition of MSW is heterogeneous in different places of the landfill, and secondly, it changes over time even in the same place due to biochemical processes. Therefore, in addition to taking into account the "relative error" and the "error dispersion" of the results in the series of measurements, we also added "measurement error due to changes in the measurement conditions" [16]. It must be emphasized that the real measurement error of such "undefined" mixtures as MSW is many times higher than the accuracy of the devices used for measurements.

2.1. Measurement of biogas emission for real landfills in typical industrial city 1 million population was fulfilled with the help of an individual multi-channel gas analyzer "MX-21-Plus" (France) and portable mobile ionic spectrometer "Multi-IMS" (Drōger, Germany). For that, 10 boreholes 1,5 m deep were made equidistant throughout the landfill. The average inaccuracy of measurements - 8.5%.

In order to calculate the maximum theoretical biogas production at MSW landfills, we used the following formula for first order reactions [17]:

$$\mathbf{V = \Sigma V_0 Q e^{-k\tau}} \qquad \text{(Eq. 1)}$$

where:

$\mathbf{V_0}$ – the theoretical MSW methane production potential, m^3/t (for "average" Ukrainian MSW is equal 80);

Q – the average quantity of MSW received at a landfill, t/year (see Tab. 1);

k – empirical coefficient equal 0.1;

τ - the period of landfill working, years (see Tab. 1).

2.2. The quantity of leachate ($\mathbf{V_f}$) which might be produced at the working area of the landfill (dump) depends mainly on the amount of annual precipitation **(P)** of the region, evaporation **(V),** and water absorption by landfill wastes (**W**) [18]. However, we added to this formula another summand **R**:

$$\mathbf{V_f = [(P-I-W-F)\cdot S\cdot 10^{-3}] + R} \qquad \text{(Eq. 2)}$$

where:

P – precipitation for this area, mm/y-m^2 (1 mm = 10 tons of precipitation per hectare; for East Ukraine P=500);

V - evaporation rate, mm/y•m^2 (for East Ukraine V=200);

W - water absorbed by solid waste, mm/year•m^2 (for East Ukraine W=100);

F - water drained, mm/year•m^2 (for East Ukraine F=10);

S - landfill working area, m^2;

R - water produced during MSW degradation, m^3/year, which is 0.3 m^3 (tons) of H_2O for every 1000 m^3 of natural biogas emitted.

Underground water samples for analysis were taken at the landfill border at the depth of 10-15 m. Altogether there were 8 wells: 2 at each of 4 sides. Three samples were taken from each well. The result of the analysis is an average value received for 3 samples. After that, an average value was obtained for all wells. Soil samples were taken at the distance of 500 m (sanitary zone) from the landfill border at the depth of 0.2-0.3 m also from four sides. From each side, 3 samples were taken. After that all samples were averaged through quartering and the analysis was fulfilled. Atomic

absorption spectrophotometer was used to measure toxic (heavy) metals in soil, water and ash (for that, samples of MSW were exposed to heat – see point 2.3). The inaccuracy of the analysis did not exceed 8%.

2.3. Derivatograph has been modified by us for heating of columns up to 325°C, and was used to study the thermal decomposition of MSW. MSW sample (225 g; composition is according to Tab.1, right column; the speed of air supply into column was constant, being 1 liter/min; in fact, this is a slow burning of MSW with limited access to oxygen). The tests were conducted with MSW being heated (in the thermostat) by +70°C, 120°C, 170°C, 220°C, 270°C, and 325 °C (when the temperature was higher than 300°C some of MSW components started to burn - for instance, the temperature of self-ignition of pressed paper is about 250°C).

2.4. We analyzed of soil and also toxic gases in air samples (1 m above ground) on the border of a sanitary zone (SZ) of the of the smoldering MSW landfill No. 3 (500 m from the edge of a landfill), with the help portative analyzers "MX-21-Plus" and "Multi-IMS" (samples of air and soil were selected and delivered to the laboratory for analysis of the heavy metals with help atomic absorption spectrophotometer). We have measured concentrations of toxic gases produced after MSW smoldering (burning) and total concentrations of "heavy" (toxic) metals in the ash. We measured the part of heavy metals, which transforms in more "volatile" forms and is emitted into the atmosphere together combustion gases as well as the part of heavy metals that enter the ash. Besides, we studied as a separate part of heavy metals in the ash, which is "labile" and can migrate from ash into soil. The inaccuracy of all measurements did not exceed 6%.

2.5. For more detailed examinations of MSW biodegradation, within laboratory conditions an artificial «closed MSW micro-dump» was created. A "laboratory composition" of MSW for our "artificial close laboratory MSW micro-dump" was obtained by crushing and mixing various components (food, paper, plastic, wood, glass, etc.) which corresponds to the average composition of MSW in a large city of Ukraine (see Tab. 1). We refused to use "natural" MSW as in such a case the results of experiments were badly reproduced. It is a 10 cm- layer of "laboratory composition" of MSW (180 g of dry MSW) and 20 g of "seeds" from bacteria and mushrooms (it is about 10 %, that in the sum with already available nitrogen approximately corresponds to its quantity in natural food waste) and 100 ml of water so that the "natural" humidity of MSW was about 30%) was placed in a glass jar with, its diameter being 15 cm. At the top of the layer a 2 cm soil-layer was placed. A polyethylene cap sealed the jar (not tightly), leaving a 20 cm air-space above the soil (under the cap). The number of "mesophilic aerobic and facultatively anaerobic microorganisms" (MAFAM) was calculated using the following procedure: an MSW sample was inoculated into a beef-extract (agar) and maintained at 37 °C for 24 hours. The grown colonies were counted after incubation (by means of a microscope) and reported as "colony forming units" (CFU) per 1 g of dry MSW. The capacity of the experimental chamber and weight of MSW were adjusted based on preliminary experiments so that the period of "laboratory biodegradation" of MSW was about 2-4 months.

2.6. We also studied the microbial activity in the unequipped MSW landfills. Measurement of gas emissions at real landfills was conducted with the help of an individual multi-channel gas analyzer "MX-21-Plus" (France). An average value was used, received on the basis of 3 measurements performed with an interval of 10 minutes. The analysis of gas samples of mini-lab-dump was carried out with the help of a modern gas chromatograph in accordance with its instruction.

III. RESULTS AND DISCUSSION

3.1. Gas research of 4 real MSW landfills

In fact, there are not the classic landfills, there are the large unequipped dumps because the MSW is delivered there by dump trucks and then compacted by the tractors (up to density 0.6 t/m^3). These dumps aren't equipped with any technical means for collecting biogas and leachate. Besides, the wrong storing leads to self-heating and smoldering inside the MSW, and then to spontaneous ignition of separate sites of a dump.

Table 1. Real landfills characteristics

Landfills	Years of operation	Average quantity of MSW received each year (tons)*	Working area, hectares	Depth, m (average)	Average composition (mass. %)
No. 1	47	115,000	11	25	food-26; plastic-20; paper-11;

No. 2	37	51,000	4	12	glass-6; wood-8; metal-8; textiles-4; stones-6; sweepings**-11.
No. 3	29	48,000	5	10	
No. 4	15	155,000	24	18	

**) The bulk density of incoming MSW is 0.25 t/m^3, after landfill compaction it is 0.6 t/m^3.*

***) Approximately 1/3 of sweepings is an organic matter.*

The volumes (the theoretical maximum possible) of biogas emitted from real landfills No.1-4 were calculated according to formula (Eq.1). The results are illustrated at Fig. 1.

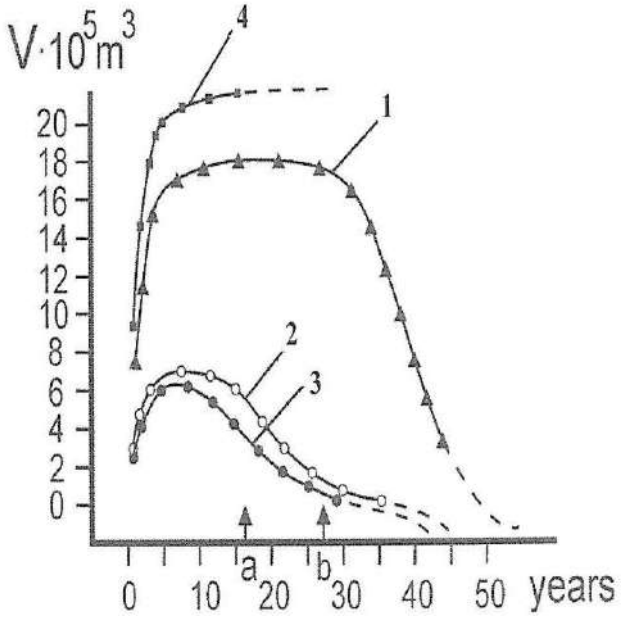

Fig. 1. The volumes (theoretical maximum possible) of biogas emitted

from real landfills No.1-4

("a" is sign when delivery of MSW to No.4 landfill was limited;

"b" is the same for No.2 and No.3 landfills)

As we can see from Fig. 1, the biogas emitted from the No.1-4 landfills during biodegradation term reach their maximum at 1/4 – 1/3 of the full working period that is connected with activity of bacteria and also alterations of pH and temperature in a landfill body (similar curves like overturned parabola were described by [19, 20]. Fig. 1 also shows that, for example, landfills No.2 and No. 3, in fact, have been already almost full 10 years ago but MSW delivery wasn't stopped there (only were limited) as this zone of the city has no other place to store MSW.

According to calculations [21], world emission of biogas (which greenhouse gas is) from 1990 to 2050 will increase by 9 times (from the real 340 Mt in 1990 up to calculate 2900 Mt) – if we will not change relation to the management of municipal waste).

At the depth of 25 m, from the bottom layers of mostly "old" No.1 landfill there have been taken samples of “residual” MSW. The age of these MSW layers corresponds to 45 years. The samples were tested for the share of organic components. The average result received on the basis of three samples is the following: the share of organic components - 13.5% (the initial share 45 years ago was about 75% - see Tab. 1). Thus, during 45 years MSW has been considerably mineralized as a result of a deep biodegradation of organic components of MSW.

In fact, these data have shown: at such landfills as No. 1-2 the process of biodegradation has almost finished, while at No. 3 and especially No. 4 «more young» landfills (see Tab. 1) are still active. Therefore, we don't share an opinion [21] regarding "Significant amounts of biogenic carbon may still be stored within the landfill body after 100 years".

Measurements of biogas (there are, basically, greenhouse gases) emissions at 4 real landfills (from 2 m deep borehole) show the following composition of biogas (see Tab. 2). That is, MSW landfills make a considerable contribution to global warming.

Table 2. Biogas (greenhouse gases) emission from real landfills

No.	Biogas (vol. %)	
	CO_2	CH_4
1	69	31
2	67	33
3	60	40
4	55	45

Gases sampled above 1m the real landfills surfaces were tested for dust, hydrogen sulfide (H_2S), nitrogen dioxide (NO_2), ammonia (NH_3), sulfur dioxide (SO_2) and carbon monoxide (CO) - see Tab. 3. These results show that the local atmospheric concentrations above the landfills were often more the norm (especially for dust and NO_2). At landfills with smoldering waste - No. 1 and No. 2 - the share of carbon monoxide sharply increases.

Table 3. Atmosphere composition at the level of 1 m above the landfill surface (mg/m^3)

Parameter	No. 1	No. 2	No. 3	No. 4	MPC*
Dust	0.8	0.5	0.6	0.3	0.15
H_2S	0.01	0.053	0.05	0.003	0.005
NH_3	0.013	0.01	0.04	0.023	0.04
NO_2	0.09	0.05	0.06	0.052	0.04
SO_2	0.14	0.05	0.012	0.018	0.05
CO	3.1 (smoldering)	5.6 (smoldering)	1.6	0.7	3.0

**)MPC - maximum permitted concentration in air of settlements (average daily).*

However, additional research found that biogas also contains micro-amounts of highly toxic chlorides methane (less 5 ppm).

3.2. Leachate pollution

None of the four landfills have a leachate collection system. We have analyzed the leachate composition at No. 3 landfill; the data are listed in Tab. 3. We have studied the composition of underground water the samples of which were taken from the wells surrounding No. 3 landfill. The sampling was done from the depth of about 5-10 m.

Table 4. Leachate composition at No. 3 landfill

Parameter	Concentration (mg/l)	MPC*
BOD**	2130	350
Oil products	110	0.5
Ammonia nitrogen	512	10.0
SSAM***	0.3	0.01
Fe	190	0.3
Ni	0.3	0.1
Zn	11.4	1.0
Pb	4.1	0.03
Cd	0.06	0.001
Cr	0.4	0.05
Hg	0.2	0.0005

**) MPC- maximum permissible concentration;*

***) BOD - biochemical oxygen demand - is the amount of dissolved oxygen needed by aerobic biological organisms in a water;*

****)SSAM - synthetic superficially-active materials.*

Data of Tab. 4 demonstrate that the concentration of toxic substances in leachate is in hundreds, and sometimes thousand times more sanitary norms (MPC), i.e. leachate is highly toxic and a very dangerous liquid.

The calculation of leachate volume produced at No. 3 landfill has been done by formula (Eq. 2). If to apply the equation to No. 3 landfill, which occupies 3.1 hectares (Tab. 1), using R = 200 m^3/y and the values shown in Tab. 7, the expected annual leachate volume will be 298 m^3/y:

$\mathbf{V_f}$ = [500 – 200 – 100 – 10] = 190 x 5 x 104 x 10^{-3} = 5890 + 300 = **298 m^3/year.**

The uncontrolled formation of such big volumes of toxic leachate should inevitably worsen ecological conditions of nearby underground water and soil.

For check of possible soil pollution on the border of a sanitary zone (SZ-border) No. 3 landfill (a concentric circle of 500 m from the edge of the landfill) were analyzed samples of soil (Tab. 5).

Table 5. The results of soil research on the SZ-border for No. 3 landfill

Parameter	**MPC*** (mg/kg)	**Real concentration**	**Outreaching**
Cd	0.2	0.78	4 times
Ni	4.0	3.3	7
Pb	6.0	1.9	3
Hg	0.05	0.3	6
Nitrates	10	82	8
Oil products	0.3	3.6	12

**) MPC- maximum permissible concentration;*

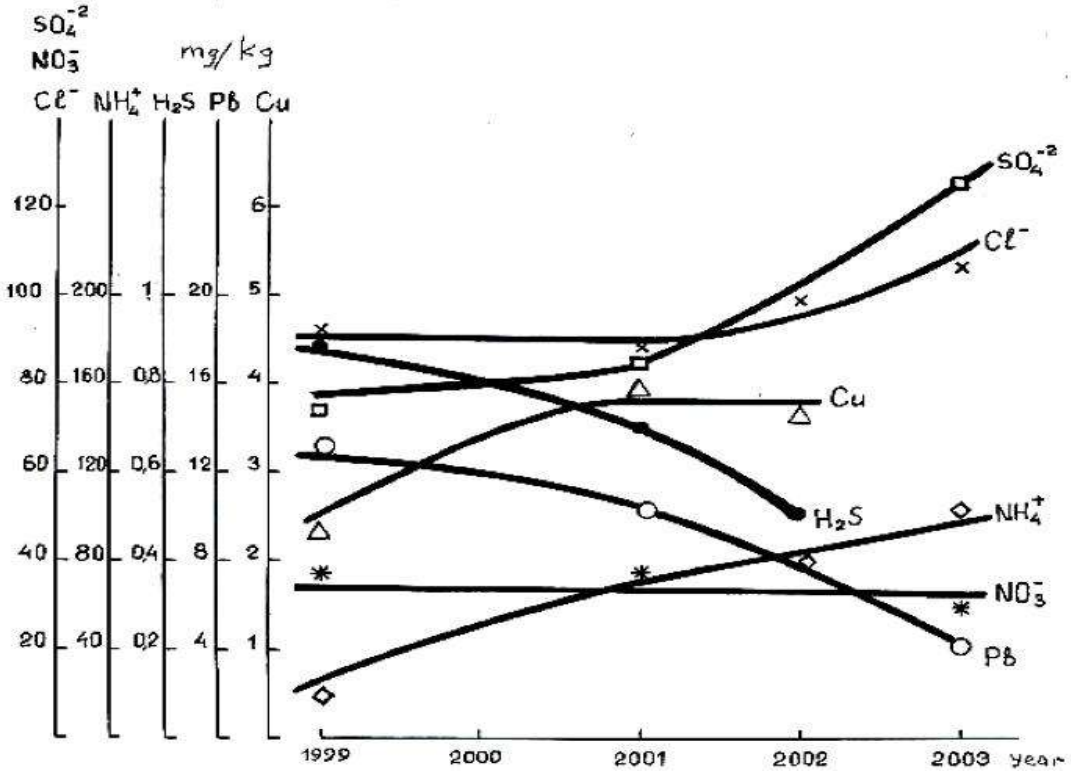

Fig. 2. Dynamics of harmful metals and ions concentration in soil at the SZ-border (500 m)

of MSW landfill No. 3

The data of Tabs. 4 and 5 and Fig. 2 confirm the worst fears regarding the high danger of leachate from unequipped MSW landfills.

3.3. The danger of MSW smoldering processes

For studying of the danger of self-heating and self-ignition of MSW stored in poorly equipped landfills, samples of MSW (in briquettes with density 0.6 t/m^3) were exposed to thermal destruction in the laboratory device by using of derivatograph as adjustable furnace at temperatures of 70-325 °C. (I remind: air was supplied to the "furnace" of the derivatograph - 1 L/min; in fact, this is a slow burning of MSW with limited access to oxygen). Results of measurements - see in Tab. 6-7.

We have measured concentrations of toxic gases produced after MSW incineration (including such super-toxic ones as hydrogen cyanide - HCN, hydrogen chloride - HCl, formaldehyde - CH_2O) and total concentrations of "heavy" (toxic) metals in the ash (with the help of a mass-spectrometer). After that, we measured the part of heavy metals, which transforms in more "volatile" forms and is emitted into the atmosphere together combustion gases as well as the part of heavy metals which enter the ash. Besides, we studied a part of heavy metals in the ash, which is "labile" (soluble) and can migrate into soils (if it will be washed out from the ash by rain). The results of the measurements are provided in Tab. 6-7.

Table 6. Concentration of emitted toxic gases after MSW incineration (mg/m^3)

Concentration of toxic gases							
CO	**SO_2**	**H_2S**	**C_6H_5OH** (phenol)	**NO_2**	**HCL**	**HCN**	**CH_2O**
678	8.8	13.7	5.7	41	0,2	0,12	19.8

Table 7. Concentration of toxic metals in initial MSW and its ash

Parameter	**Concentration of toxic metals in MSW ash (mg/kg)***						
	Pb	**Ni**	**Cr**	**Cu**	**Zn**	**Hg**	**Co**
Sample of initial MSW	511	140	190	1270	2410	3.2	46
Sample of MSW ash	288	120	180	1100	2080	0	36
Quantity of toxic metals that was washed out from the ash – imitation of rain	48.3	8.5	9.9	15.7	23.8	0	1.34

By comparing the data of Tab. 7 we can see that the ash accumulates all toxic metals, excluding mercury and lead: mercury completely evaporates into the air and lead – half-on-half. Therefore, the proposal to use ash after recycling MSW through incineration for building materials [22] causes concern. So, we have established that during the incineration of MSW the vast emission of toxic gases in the atmosphere will take place. Some parts of each of the heavy metals are taken into the atmosphere together with combustion gases, the other parts enter the ash. At the same time, some parts of heavy metals that have passed into ash are in a soluble form, i.e. they might (in case of precipitation of ash on wet soil) enter into the soil.

For check air pollution on the border of a sanitary zone (SZ-border) for smoldering No. 1 landfill (a concentric circle of 500 m from the edge of the landfill), samples of air were analyzed (see Tab. 8).

Table 8. The results of research of the SZ-border for No. 1 landfill (mg/m^3)

Parameter	**MPC***(mg/m^3)	**Amount**	**Exceeding**
NO/NO_2	0.035	0.55	16 *(times)*
H_2S	0.05	0.39	8
HCl	0.2	0.8	4
Ash	0.1	0.71	7

**) MPC- maximum permissible concentration;*

Evidently, combustion gases from the smoldering dumps have high toxicity (see Tab. 8) and high danger for environment and human health.

When researchers began to explore burning landfills, even more toxic, deadly compounds were found in the flue gases: hydrogen chloride, dioxin, and furan [23]. Unfortunately, we didn't study the smoldering dumps

concerning dioxin due to the lack of access to reliable analyzers of dioxin. Therefore, the scientific paper [24] well fills up a gap in our studying. At research of influence of the illegal burning dumps in Italy (province of Campania) on the health of local population, it was found high concentrations of dioxins (≥ 5.0 pg TEQ/g fat) in sheep and cow milk samples, and also dangerous contamination of dioxin and polychlorinated biphenyls in woman milk samples from those living in Campania (at 16.6 pg TEQ/g of fat).

The calculation of the maximum concentration limit C_{ml} (g/s), i.e. the amount of harmful substances emitted by the polluting source per unit of time which will create at the surface layer (at the height of 2 m from the ground level) the concentration equal to a maximum allowable concentration of harmful particles in the atmosphere M (considering a background concentration Cb), was calculated using the special computer program.

The calculation was done with the help of the computer software, the results are illustrated at Fig. 3. At the border of the SZ (green circle with a red flag) the concentration of one of the most toxic components of fire-hazardous gases – nitrogen oxide – exceeds MPC 16,59 times.

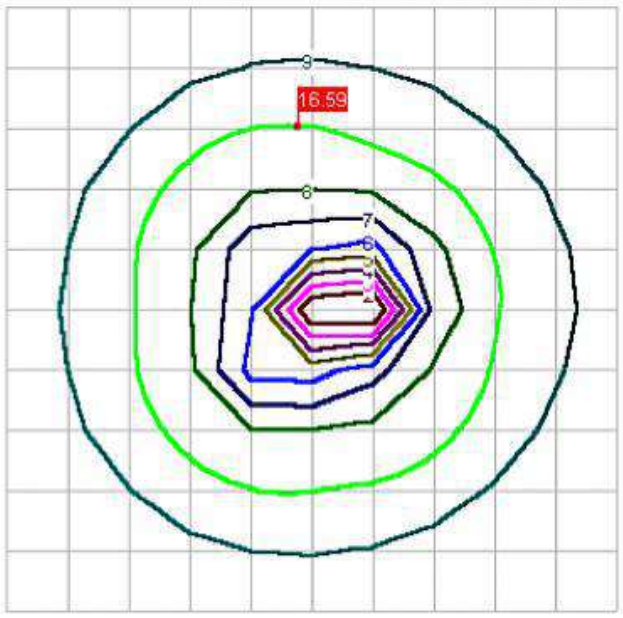

Fig. 3. Fire gas (NO) dissipation within the SZ limits (500 m) during MSW dump burning

(scale: 1 cell - 165 m; the inner black oval is the contour of the landfill)

*****

Also, it is necessary to be careful about the activities of waste incinerators. All purification systems for such plants still don't ensure the safety. So, in Doral, Florida, the plant-incinerator garbage burns approximately 1.000.000 tons of MSW each year, which causes thousands of odor and respiratory issues, complaints [25].

3.4 Microbial activity in the unequipped MSW Landfill

3.4.1. Experimental studies of MAFAM (see paragraph 2.5)

We have also conducted experimental studies of this process for the so-called MAFAM bacteria group. In a "close laboratory dump" their population reached the peak after 30 days and then it decreasing over the following 120 days (see Fig. 4).

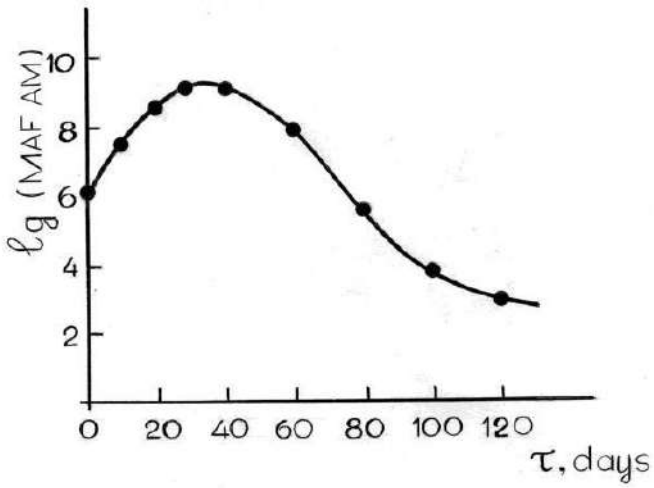

Fig. 4. Population increase graph of MAFAM microorganisms for "laboratory MSW micro-dump"

As it can be seen in Fig. 4, laboratory results show that the curves of breeding reach their maximum during the first quarter of the MSW biodegradation period.

3.4.2.Toxic and greenhouse gases emission

In order to check experimentally the correlation between the dynamics of microorganism colony development within a "closed laboratory MSW dump" and gas generation in this "close laboratory dump", we have implemented an additional analysis of gas samples within a glass vessel over a MSW layer - see Fig. 5.

Fig. 5 shows that the all toxic gases generated from disposed MSW have maximum at 1/4 – 1/3 of the incubation time. The measurements of the temperature of the "laboratory" dump have shown that during the process of biodegradation the temperature increases up to 50 - 60 °C. It allows us to state that the processes of gas emission from the body of landfills are determined mainly by bacterial activity, and this trend also coincides with the gas emission curves from real MSW landfills (Fig. 1).

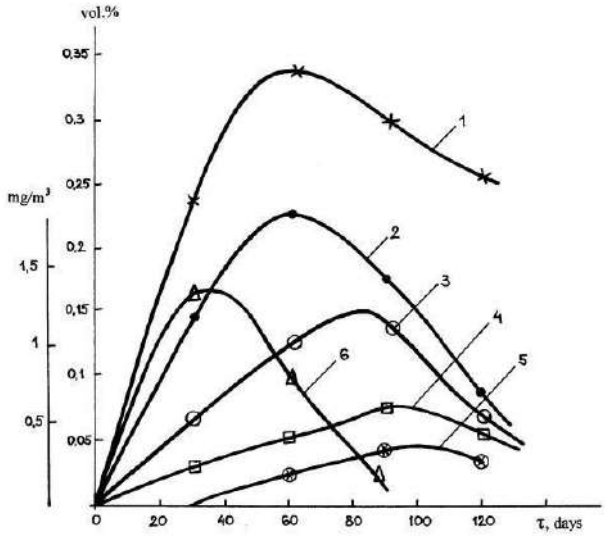

Fig. 5. Experimental dynamics of toxic and greenhouse gas emissions from a "laboratory MSW micro-dump"

1 - carbon dioxide (vol. %); 2 - methane (vol. %); 3 - ammonia (mg/m3); 4 - hydrogen sulfide (mg/m³); 5 - hydrogen chloride (mg/m³); 6 - formaldehyde (mg/m³)

3.5. Smoldering of unequipped MSW landfills and its extinguishing

Since MSW biodegradation reactions are exothermic, there is a potential for self-heating and self-ignition of dumps (which is often the case). From classical thermodynamics, it is known that the process of self-heating transforms into burning when the heat flow (+Q) from exothermic reactions of oxidation exceeds natural heat removal (-Q) from the reaction zone. The interrelation of [(+Q) > (-Q)] often takes place during natural MSW biodegradation processes, especially in summer time - in this extreme case, the temperature inside the landfill can sometimes reach 150-200 °C (see Fig. 6).

Fig. 6. Smoldering and self-ignition on the real unequipped landfill

We measured the concentrations of toxic gases at the border of the sanitary zone of a real non-equipped MSW landfill with visible intense smoldering and partial ignition (500 m from the edge of the landfill) - see Tab. 9.

Tab. 9. Concentration of toxic gases for the unequipped smoldering real landfill (on its border of the sanitary zone)

Toxic gas	Concentration, mg/m³	MPC*	Excess
CO	31.1	3.0	10.4 times
NO/NO_2	0.33	0.04	8.25 times
SO_2	0.8	0.05	16 times
CH_2O	0.02	0.003	6.7 times

**) MPC - maximum permitted concentration in air of settlements (average daily)*

As can be seen from Tab. 9, on the border of the sanitary zone of the landfill (this is 500 m from its edge!) there is a huge excess of permissible concentrations (from 5 to 15 times) of not just harmful, but extremely toxic gases. This means that a smoldering (especially burning) landfills is a high danger, and their smoldering (and even more active burning) should be extinguished as soon as possible. However, ordinary fire engines are unsuitable for these purposes: when the landfill is smoldering, due to the burning out of large volumes of solid waste, huge "hot pits" are formed, and fire engines with a driver can completely fall there.

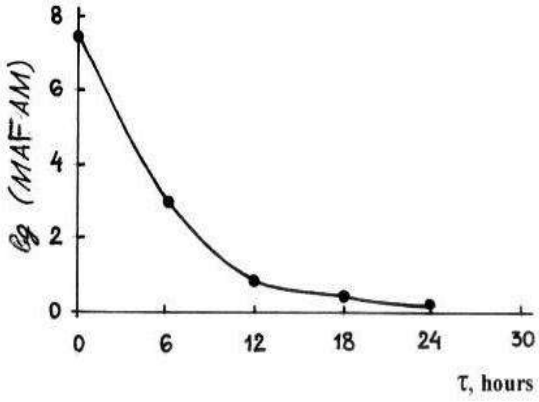

Fig. 7. Dynamics of decrease of MAFAM population after MSW treatment by a $Ca(OH)_2$ suspension

That's why we have searched for potential technologies to suppress the activity of bacteria inside the unequipped landfill. "Lime milk" (suspension of $Ca(OH)_2$) has been

selected as a simple, safe, and cheap reagent for that goal. We treated our "artificial laboratory MSW micro-dump" with a 10%-$Ca(OH)_2$ suspension at a ratio 0.1 volumes of suspension per 1 volume of MSW. As it follows from Fig. 7, after one day (24 hours) after the treatment, the quantity of MAFAM came close to zero (gas emission also stopped and the temperature inside the body of the "laboratory dump" approached room temperature.)

Extinguishing focuses of smoldering at real unequipped MSW landfills can be carried out by pumping special solutions or suspensions into its body - Fig. 8. In this case, first of all, the bacterial activity is suppressed, then the smoldering center is cooled. To extinguish smoldering focus in real MSW landfills, we can accept the ratio: 0.1 m^3 suspension per 1 m^3 of MSW in the center of the smoldering place.

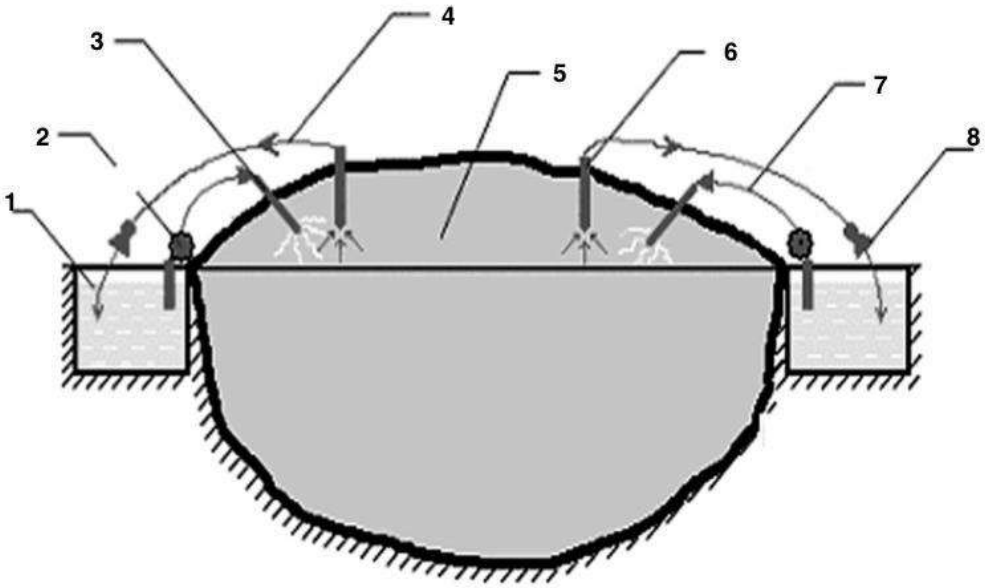

Fig. 8. Technological scheme for extinguishing an unequipped MSW landfill with lime suspension $Ca(OH)_2$:

***1** - mixing tank (cistern) for the preparation and storage of lime slurry [$Ca(OH)_2$]; **2** - powerful pump, supplying (under pressure) lime suspension from the tank to the injector (up to 60 m^3/h); **3** - injector (pipe section 4-6 m long and 0.1 m in diameter with perforation and a pointed end) for injecting the suspension directly into the smoldering center inside the landfill; **4** - pipeline for the removal of combustible and toxic gases from the landfill (they are pumped into the tank with $Ca(OH)_2$ for neutralization); **5** - MSW landfill body; 6 - powerful vacuum pump (gas capacity up to 1 m^3/s); **6** - perforated suction metal pipe (like injector, pos. 3) for venting gases from the landfill body; **7** - pipeline for supplying the suspension to the injector; **8** - powerful vacuum pump (gas capacity up to 1 m^3/s) for suction of hot and toxic gases from the landfill near the smoldering center (they are pumped into a tank with $Ca(OH)_2$ for neutralization).*

IV. CONCLUSIONS AND RECOMMENDATIONS

1. Researched MSW landfills are unequipped landfills (see chapter "Introduction"), therefore, they pose a danger to the environment (air, ground, and underground water) and the health of the population of nearby settlements. The general impact of unequipped MSW landfills on public health is well illustrated by Fig. 9.

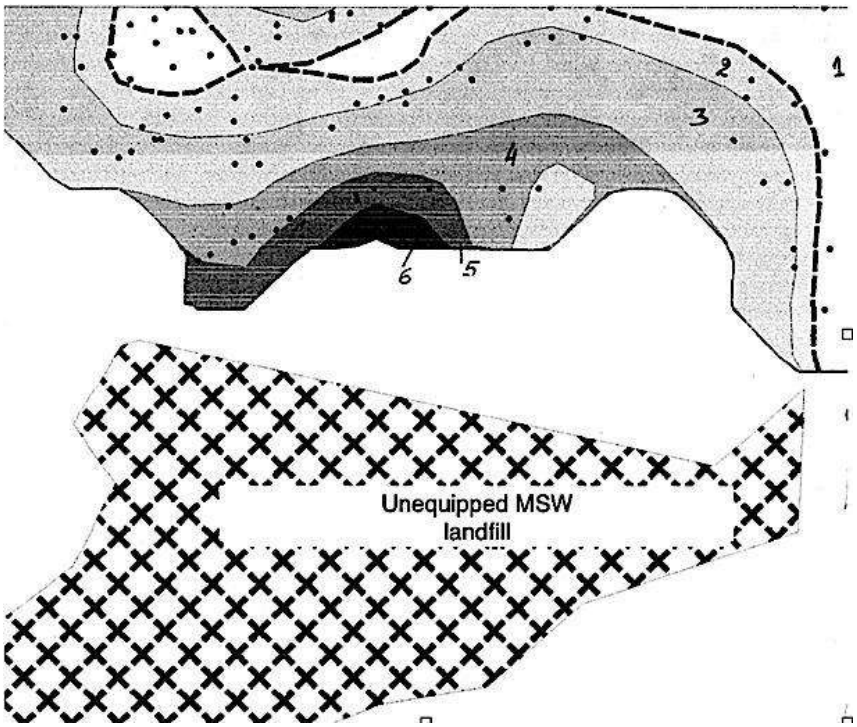

Fig. 9. Map of the average lifetime of people who live nearby unequipped MSW landfill No. 1 (it were unauthorized self-built dwellings; they were demolished in the 1970s) (scale: 1 cm – 0.5 km; data of Donetsk Medical Institute):
zone 1 - more than 66 years, 2 – 63 years, 3 – 59 years, 4 – 55 years, 5 – 51 years, 6 – less than 50 years;

2. The above results of a survey of four real MSW landfills, namely: the emission of "landfill gas" (which is a greenhouse gas), the emission of various poisonous gases, as well as highly toxic flue gases due to bacterial activity in landfill body.

3. The gas emission curves from a "close laboratory dump" of MSW during biodegradation (Fig. 4, 5) and also analogous experimental curves from real MSW dumps (Fig. 1) – all of them reach their maximum approximately one third of incubation period.

4. The distribution of emitted by an MSW landfill "heavy" (toxic) metals in air, water and soil has been studied.

5. It is showing a significant role in unequipped MSW landfill biodegradation is played by microorganisms, i.e. they are responsible both for environment pollution of greenhouse and toxic gases and for self-heating of some of landfill areas that often leads to smoldering and even burning.

6. On the border of the so-called sanitary zone of the smoldering landfill (500 m around), takes place a significant excess of MPS (maximum permissible concentration) within 5-15 times.

7. It is necessary to arrange a periodical treatment of an unequipped MSW landfill's "problem areas" with a 10%-suspension of a "lime milk" [$Ca(OH)_2$] to slow down microbiological activity and prevention of MSW landfill smoldering and self-ignition.

8. Thus, a detailed qualitative and quantitative analysis of the biodegradation of both real landfills and its laboratory model showed that an unequipped (or poorly equipped) MSW landfill, in fact, is a dangerous ungovernable biochemical reactor.

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Precision Farming: A Review of Methods, Technologies, and Future Prospects

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Abstract— *Precision farming, often referred to as precision agriculture, is a farming management concept that seeks to maximize agricultural operations via the use of technology in order to achieve greater levels of efficiency, production, and conservation of resources. The purpose of this survey study is to offer an overview of precision farming, including topics such as its fundamentals, methodologies, technology, and applications. It investigates the role that a variety of technologies, including global navigation satellite systems (GNSS), geographic information systems (GIS), remote sensing, and sensor technology, play in making it possible to implement precision agricultural methods. In addition, it addresses the advantages, disadvantages, and potential future applications of precision farming in the context of solving issues related to the sustainability of agriculture and the security of food supplies on a worldwide scale.*

Keywords—- ***GNSS, GIS, Remote sensing.***

I. INTRODUCTION

The notion of precision farming, which is often referred to as precision agriculture, is a farming management concept that makes use of cutting-edge technology and data-driven methodologies in order to improve agricultural operations on a geographical and temporal scale. Instead of applying agricultural inputs and techniques in a consistent manner over wide regions, the fundamental goal of precision farming is to adjust them to the precise requirements of individual plants or tiny zones within a field. Through the use of this strategy, farmers are able to optimize their production, reduce their input consumption, and mitigate their environmental consequences, which eventually results in agricultural systems that are more efficient and sustainable.

Precision farming is based on the premise of site-specific management, which acknowledges that the properties of the soil, terrain, climate, and other elements change within a field. This provides the foundation for the concept of precision farming. Using technology like as global positioning systems (GPS), geographic information systems (GIS), and remote sensing, farmers are able to properly map and analyze these changes, which enables them to make educated choices on the distribution of resources such as seeds, water, pesticides, and fertilizers. By ensuring that crops get the appropriate quantity of nutrients and water exactly when and where they are required, this focused application of inputs maximizes the potential for growth and yield as well as the potential for growth.

The concept of precision farming is founded on the principle of site-specific management, which recognizes that soil characteristics, topography, climate, and other factors vary within a field. By accurately mapping and analyzing these variations using technologies such as GPS, GIS, and remote sensing, farmers can make informed decisions about the allocation of resources such as fertilizers, pesticides, water, and seeds. This targeted application of inputs ensures that crops receive the right amount of nutrients and water precisely when and where they are needed, optimizing growth and yield potential.

Key components of precision farming include:

1) Data Collection: Precision farming relies on the collection of accurate and timely data about soil properties, crop health, weather conditions, and other relevant factors. This data can be obtained through various means, including satellite imagery, drones, ground-based sensors, and manual sampling.
2) Data Analysis: Once collected, data is analyzed to identify patterns, trends, and spatial variability within the field. Advanced analytics techniques, such as spatial interpolation, statistical modeling, and machine learning, are often employed to derive actionable insights from the data.
3) Decision Support Systems: Based on the analysis of data, decision support systems provide farmers with recommendations and guidance for optimizing farm management practices. These systems may include software tools, mobile applications, and online platforms that integrate data from multiple sources and provide real-time monitoring and decision-making support.
4) Precision Application Technologies: Precision farming relies on technologies that enable the precise application of inputs, such as variable rate application equipment, GPS-guided machinery, and automated irrigation systems. These technologies allow farmers to adjust input rates and application timing based on site-specific conditions, maximizing efficiency and minimizing waste. Precision farming is a comprehensive concept that incorporates various technologies and scientific knowledge, such as computer science, electronics, and geoprocessing. These technologies and knowledge are applied in agriculture to optimize production by accounting for variability and uncertainties within agricultural systems. Precision farming involves the use of sensors, information systems, enhanced machinery, and informed management to adapt production inputs site-specifically within a field and individually foreach animal.
5) This approach allows for better resource utilization, reduces the need for fertilizers, and improves water efficiency. By utilizing technology such as satellite positioning, internet-of-things, and data analytics, precision farming enables farmers to monitor and manage the quantity and quality of agricultural produce, while also maintaining the quality of the environment and improving the sustainability of the food supply. Precision farming is a comprehensive approach that utilizes various technologies and scientific knowledge to optimize agricultural production by considering variability and uncertainties within agricultural systems. Precision farming is a comprehensive approach that utilizes various technologies and scientific knowledge to optimize agricultural production by considering variability and uncertainties within agricultural systems. These technologies and knowledge are applied to adapt production inputs site-specifically within a field and individually for each animal, allowing for better use of resources while maintaining.

Evolution and significance in modern agriculture- Precision farming in contemporary agriculture represents a significant change from conventional agricultural techniques to data-driven and technology-enabled approaches, with profound implications for the industry. In the past, agriculture mostly used broad methods, applying fertilizers, insecticides, and water equally over extensive areas, without considering differences in soil qualities, terrain, or crop needs. Nevertheless, the introduction of precision farming has completely transformed this way of thinking by using sophisticated technology to customize agricultural methods according to the unique requirements of each field or even tiny areas within fields.

Precision farming originated in the late 20th century, with the early use of technology like Global Positioning Systems (GPS) and Geographic Information Systems (GIS) in agriculture. These technologies allowed farmers to precisely delineate field borders, evaluate soil properties, and evaluate the condition of crops from a spatial standpoint. With the advancement and affordability of processing power, precision farming solutions have broadened to include remote sensing technologies like satellite images and drones, along with sensor-based monitoring systems for collecting real-time data.

Precision farming is very important in contemporary agriculture because to its capacity to optimize resource utilization, improve crop productivity, and limit negative effects on the environment. Farmers may enhance efficiency and output while minimizing waste by accurately directing inputs such as fertilizers, herbicides, and water to specific regions of the field where they are most required. This focused strategy not only enhances economic benefits for farmers but also reduces environmental hazards linked to excessive pesticide use and fertilizer runoff.

Furthermore, precision farming enables farmers to make decisions based on data, enabling them to closely monitor the development of crops, identify pests and illnesses at an early stage, and take proactive measures to adapt to changing circumstances. Precision farming utilizes the integration of diverse data sources and sophisticated analytics to help farmers optimize planting schedules,

improve irrigation management, and anticipate crop yields with enhanced accuracy. This improved decision assistance enables farmers to reduce risks and maximize profits in a climate that is becoming more uncertain and varied.

Precision farming not only provides advantages at the farm level, but also plays a vital role in tackling wider issues that contemporary agriculture faces, including food security, sustainability, and climate change adaptation. Precision farming enhances resource efficiency and resilience, hence promoting the sustainable sustainability of agricultural systems and safeguarding food security for an expanding global population. Precision farming promotes the shift towards sustainable and eco-friendly agricultural techniques by decreasing greenhouse gas emissions, limiting soil erosion, and protecting water quality.

Principles of Precision Farming: The principles of precision farming are centered on the fundamental ideas of site-specific management, data-driven decision-making, and ongoing monitoring and modification. The deployment of precision farming methods and technology is guided by these principles, which allow farmers to maximize agricultural productivity while reducing inputs and environmental consequences. The following are the fundamental tenets of precision agriculture:

Site-Specific Management:

- Site-specific management recognizes that agricultural fields exhibit spatial variability in soil properties, topography, and other factors that influence crop growth and productivity.
- Instead of treating the entire field as uniform, precision farming identifies and manages small zones or management units within the field based on their unique characteristics.
- By tailoring management practices, such as planting, fertilization, and irrigation, to the specific needs of each zone, farmers can optimize resource use and maximize yield potential.
- Data-Driven Decision-Making: Precision farming relies on accurate and timely data to inform decision-making processes. This data may include information on soil properties, crop health, weather conditions, and historical yield data.
- Advanced technologies, such as GPS, GIS, remote sensing, and sensor networks, are used to collect, analyze, and interpret data from the field.
- By integrating data from multiple sources and applying advanced analytics techniques, farmers can gain insights into spatial variability and make informed decisions about crop management practices.

Continuous Monitoring and Adjustment:

- Precision farming involves ongoing monitoring of crop conditions, soil moisture levels, nutrient status, and other relevant parameters throughout the growing season. Monitoring may be conducted using remote sensing technologies, sensor networks, and on-the-ground observations. Based on real-time data and feedback, farmers can make timely adjustments to management practices, such as adjusting irrigation schedules, applying additional inputs to areas of deficiency, or implementing pest control measures. Continuous monitoring and adjustment enable farmers to respond quickly to changing conditions and optimize crop performance throughout the growing season.

II. TECHNOLOGIES ENABLING PRECISION FARMING

Global Navigation Satellite Systems (GNSS): Global Navigation Satellite Systems (GNSS) play a crucial role in enabling precision farming by providing accurate positioning and navigation information to agricultural machinery and devices. GNSS technologies, such as GPS (Global Positioning System), GLONASS (Global Navigation Satellite System), and more recently Galileo and BeiDou, offer farmers precise location data, which is essential for implementing site-specific management practices and precision agriculture techniques. Here's how GNSS technologies facilitate precision farming:

Accurate Positioning: GNSS receivers installed on agricultural equipment and devices receive signals from satellites orbiting the Earth, allowing them to determine their precise position in real-time. This accurate positioning is essential for guiding farm machinery during field operations, such as planting, spraying, harvesting, and soil sampling, ensuring that operations are carried out with high precision and efficiency.

Guidance Systems: GNSS-based guidance systems provide farmers with visual or audible cues to follow predefined paths or patterns in the field, such as straight-line rows or curved contours. By guiding agricultural machinery along precise paths, guidance systems help reduce overlaps and skips in input application, minimizing input wastage and optimizing resource use.

Auto-Steering: Auto-steering systems use GNSS data to automatically steer agricultural vehicles, such as tractors and sprayers, along predefined routes with high accuracy. These systems eliminate the need for manual steering by

the operator, reducing fatigue and allowing for more consistent and precise field operations.

Mapping and Georeferencing: GNSS technology enable the creation of accurate field maps and georeferenced data layers, which are essential for site-specific management and decision-making. Field boundaries, soil sampling points, crop health assessments, and yield maps can be precisely georeferenced using GNSS data, facilitating spatial analysis and management.

Integration with Precision Farming Technologies: GNSS receivers are integrated with other precision farming technologies, such as variable rate application (VRA) systems and data management software. By combining GNSS positioning data with information on soil properties, crop performance, and input requirements, farmers can implement targeted input application strategies tailored to the specific needs of different areas within a field.

Role of GPS, GLONASS, and other satellite Systems; - Global Navigation Satellite Systems (GNSS), including GPS (Global Positioning System), GLONASS (Global Navigation Satellite System), and other satellite systems like Galileo and BeiDou, play a critical role in precision farming by providing accurate positioning, navigation, and timing information to agricultural applications. Here's how each of these satellite systems contributes to precision farming:

1. **GPS (Global Positioning System):** GPS is a satellite-based navigation system developed and operated by the United States government. GPS provides global coverage and accurate positioning information to users equipped with GPS receivers. In precision farming, GPS technology enables farmers to precisely determine the location of agricultural machinery, equipment, and sensors in the field. GPS receivers are integrated into various precision farming applications, such as auto-steering systems, guidance systems, and yield monitors, allowing farmers to conduct field operations with high accuracy and efficiency.
2. **GLONASS (Global Navigation Satellite System):** GLONASS is a satellite-based navigation system developed by Russia, which provides similar positioning and navigation capabilities to GPS. GLONASS complements GPS by offering additional satellite coverage and redundancy, particularly at high latitudes and in urban environments where GPS signals may be obstructed or degraded. In precision farming, GLONASS receivers can be used alongside GPS receivers to improve positioning accuracy and reliability, especially in challenging environments.
3. **Galileo:** Galileo is the European Union's global satellite navigation system, designed to provide independent, reliable positioning and timing services to users worldwide. Galileo aims to offer higher positioning accuracy, integrity, and availability compared to existing GNSS systems like GPS and GLONASS. In precision farming, Galileo satellites can augment GPS and GLONASS signals, enhancing the overall accuracy and robustness of positioning information for agricultural applications.
4. **BeiDou:** BeiDou is China's satellite navigation system, which provides regional and global positioning services similar to GPS and GLONASS. BeiDou offers improved accuracy, coverage, and signal availability in the Asia-Pacific region, where it is being increasingly used for precision farming and other applications. In precision farming, BeiDou receivers can provide additional satellite signals to complement GPS, GLONASS, and Galileo, further enhancing positioning accuracy and reliability in diverse environments.

Precision agriculture applications of GNSS technology- Global Navigation Satellite Systems (GNSS) technology, such as GPS (Global Positioning System), GLONASS (Global Navigation Satellite System), Galileo, and BeiDou, offers a wide range of applications in precision agriculture. These applications leverage GNSS capabilities to enhance efficiency, accuracy, and productivity in various farming operations. Here are some precision agriculture applications of GNSS technology:

1. **Field Mapping and Boundary Definition:** GNSS technology enables farmers to accurately map field boundaries and delineate management zones within fields. GPS receivers integrated with mapping software allow farmers to create detailed field maps, which serve as a basis for precision agriculture practices such as variable rate application (VRA) and site-specific management.
2. **Auto-Steering and Guidance Systems:** Auto-steering systems use GNSS signals to automatically steer agricultural vehicles, such as tractors, combines, and sprayers, along predefined paths or guidance lines. By maintaining accurate positioning and guidance, auto-steering systems reduce operator fatigue, minimize overlaps and skips in field operations, and ensure precise implement placement.
3. **Variable Rate Application (VRA):** GNSS technology enables variable rate application (VRA) of inputs such as fertilizers, pesticides, and irrigation water based on spatial variability within fields. By integrating GPS data with soil maps, yield maps, and

other spatial information, farmers can apply inputs at varying rates according to the specific needs of different areas within a field, optimizing resource use and crop yields.

4. **Yield Monitoring and Mapping:** GNSS-equipped yield monitors installed on harvesters collect real-time data on crop yields as they are harvested. GPS positioning data is integrated with yield data to create detailed yield maps, which provide insights into spatial variability in crop performance and guide future management decisions.
5. **Soil Sampling and Precision Soil Management:** GNSS technology facilitates precise soil sampling by accurately recording sample locations in the field. GPS-guided soil sampling allows farmers to collect soil samples at predetermined locations, ensuring representative sampling and enabling site-specific soil nutrient management strategies.
6. **Irrigation Management:** GNSS technology supports precision irrigation by providing accurate positioning for irrigation equipment and sensors. GPS-guided irrigation systems can adjust water application rates based on spatial variability in soil moisture levels and crop water requirements, optimizing water use efficiency and minimizing water waste.
7. **Remote Sensing Integration:** GNSS data can be integrated with remote sensing technologies, such as satellite imagery and drones, to monitor crop health and growth patterns. By combining GNSS positioning with remote sensing data, farmers can identify areas of stress, disease, or nutrient deficiency in crops and target management interventions accordingly.

Geographic Information Systems (GIS):- Geographic Information Systems (GIS) are powerful tools that play a crucial role in precision farming by enabling farmers to manage, analyze, and visualize spatial data related to agricultural operations. GIS technology integrates geographic data, such as maps, satellite imagery, and field observations, with attribute data, such as soil characteristics, crop types, and yield data, to provide valuable insights for decision-making. Here's how GIS is utilized in precision farming:

1. Field Mapping and Management: GIS allows farmers to create detailed maps of their fields, including boundaries, management zones, and infrastructure such as irrigation systems and drainage ditches By digitizing field boundaries and other features, farmers can accurately delineate management zones based on factors such as soil type, topography, and historical yield data.
2. Soil Mapping and Analysis: GIS enables farmers to create soil maps that depict spatial variability in soil properties, such as texture, pH, organic matter content, and nutrient levels. By overlaying soil maps with other data layers, such as yield maps and crop health imagery, farmers can identify areas of nutrient deficiency or soil compaction and implement targeted management practices, such as variable rate fertilization or soil remediation.
3. Yield Monitoring and Analysis: GIS is used to analyze and visualize yield data collected from precision agriculture equipment, such as yield monitors on harvesters. By creating yield maps that display spatial variability in crop yields across the field, farmers can identify patterns, trends, and areas of underperformance or overperformance, informing future management decisions and crop planning.
4. Precision Application of Inputs: GIS-based prescription mapping allows farmers to create application maps for variable rate application (VRA) of inputs, such as fertilizers, pesticides, and irrigation water. By integrating GIS data with agronomic models and soil test results, farmers can generate prescription maps that specify optimal input rates for different areas within the field, optimizing resource use and maximizing crop yields.
5. Remote Sensing Integration: GIS facilitates the integration of remote sensing data, such as satellite imagery and aerial photographs, into precision farming workflows. By overlaying remote sensing data with GIS layers, farmers can monitor crop health, detect pests and diseases, assess vegetation vigor, and track changes in land cover and land use over time.
6. Decision Support Systems: GIS-based decision support systems provide farmers with tools for spatial analysis, scenario modeling, and risk assessment. By combining GIS capabilities with advanced analytics techniques, such as spatial interpolation, suitability analysis, and multi-criteria decision-making, farmers can make informed decisions about crop management practices, land use planning, and resource allocation.

Remote Sensing: Remote sensing, using satellite and aerial imaging, is essential in precision farming since it provides vital data for monitoring crops, estimating yields, detecting diseases, and analyzing soil. Remote sensing is used in precision farming to get accurate results.

Crop monitoring: - It is the use of satellite and aerial imaging to observe and assess the progress, advancement, and condition of crops over the whole period of growth. Satellites and drones acquire high-resolution images that offers farmers precise information on crop conditions, such as the extent of canopy cover, the amount of biomass accumulated, and the various phases of plant development. Through the examination of changes in the patterns of light reflected by crops over a period of time, farmers are able to evaluate the overall condition of the crops, identify elements that may be causing stress (such as lack of nutrients, insufficient water, or infestations of pests), and pinpoint regions where there is a risk of reduced crop output. Crop yields are estimated using remote sensing data, which involves assessing crop biomass, vegetation indices, and other indications of agricultural production. Farmers may use remote sensing data, in conjunction with ground-based measurements and historical yield data, to calibrate and establish models that can accurately forecast crop yields at various development stages and across diverse fields. Remote sensing-based yield prediction enables farmers to evaluate crop performance, make prompt choices about harvest timing, and strategize for grain storage and sale.

Disease Detection: Crop diseases and pest infestations are detected and monitored using satellite and aerial images. Remote sensing data may be used to detect spatial patterns of disease outbreaks, enabling farmers to identify regions with high disease prevalence and prioritize management actions. By combining remote sensing data with disease models and on-site observations, farmers may use specific control measures, such as pesticide usage or crop rotation, to reduce disease transmission and limit decreases in agricultural output.

Soil analysis: - It is involves the use of remote sensing methods, such as hyperspectral imaging and soil moisture mapping, to evaluate soil qualities and conditions. Satellite and aerial images may be used to accurately monitor soil moisture levels, soil texture, organic matter content, and soil salinity over extensive agricultural regions. Through the examination of regional heterogeneity in soil parameters, farmers may enhance irrigation scheduling, customize nutrient management tactics, and pinpoint regions susceptible to soil deterioration or erosion.

Sensor Technology: Sensor technology plays a vital role in precision farming by enabling real-time data collection and monitoring of various environmental and crop-related parameters. Here's how sensor technology contributes to precision farming:

1. Deployment of Sensors for Real-Time Data Collection: Sensor networks are deployed across agricultural fields to collect real-time data on soil, weather, crop, and environmental conditions. These sensors are strategically placed throughout the field to capture spatial variability in parameters such as soil moisture, temperature, nutrient levels, and crop health. Sensors may be installed in the soil, on weather stations, on drones, or mounted on agricultural machinery to continuously monitor field conditions during the growing season.
2. Monitoring Soil Moisture: Soil moisture sensors are used to measure the moisture content of the soil at different depths and locations within the field. These sensors provide real-time information on soil moisture levels, allowing farmers to assess soil water availability and make informed decisions about irrigation scheduling. By monitoring soil moisture continuously, farmers can optimize irrigation timing and volume, reduce water use, and minimize the risk of overwatering or underwatering crops.
3. Monitoring Soil Temperature:

Soil temperature sensors measure the temperature of the soil at various depths to assess soil thermal properties and conditions. These sensors help farmers monitor soil temperature fluctuations throughout the day and across different seasons, which is critical for crop growth and development. By tracking soil temperature, farmers can optimize planting schedules, manage crop emergence and growth stages, and mitigate the risk of frost damage or heat stress.

4. Monitoring Nutrient Levels: -Nutrient sensors are used to monitor soil nutrient levels, such as nitrogen (N), phosphorus (P), potassium (K), and other essential nutrients. These sensors provide real-time data on soil nutrient concentrations, allowing farmers to assess nutrient availability and make informed decisions about fertilizer applications. By monitoring nutrient levels continuously, farmers can implement precision nutrient management strategies, adjust fertilizer rates, and reduce nutrient losses through leaching or runoff.
5. Monitoring Other Environmental Parameters: Sensor technology is also used to monitor other environmental parameters, such as air temperature, humidity, wind speed, solar radiation, and atmospheric pressure. These sensors provide valuable data for assessing microclimate conditions, predicting weather patterns, and optimizing agronomic practices such as pest management, crop spraying, and harvesting operations.

Precision Farming Applications: Precision farming encompasses a variety of applications aimed at optimizing

agricultural practices, improving efficiency, and enhancing sustainability. Here are some key precision farming applications:

1. Variable Rate Technology (VRT): Variable Rate Technology (VRT) is a precision farming practice that involves the adaptive application of agricultural inputs, such as fertilizers, pesticides, and irrigation water, based on spatial variability within fields. Instead of applying inputs uniformly across the entire field, VRT allows farmers to tailor input application rates to match the specific needs of different areas or zones within the field. Here's how VRT works and its benefits:

Spatial Variability Assessment: VRT begins with the assessment of spatial variability within the field, which may include variations in soil properties, topography, crop health, and yield potential. Farmers use tools such as soil maps, yield maps, satellite imagery, and soil sensors to identify spatial patterns and zones of high and low productivity within the field.

Prescription Map Generation: Based on the spatial variability assessment, farmers generate prescription maps that specify optimal input rates for different zones or management units within the field.

Prescription maps are created using specialized software that integrates data from various sources, such as GIS, GPS, remote sensing, and soil sampling, to generate recommendations for input application rates.

1. Adaptive Input Application: Using VRT-enabled equipment, such as variable rate fertilizer spreaders, sprayers, or irrigation systems, farmers apply inputs at variable rates according to the prescription maps. VRT equipment is equipped with GPS guidance systems and controllers that adjust input application rates in real-time as the equipment moves through the field. Input rates may vary based on factors such as soil fertility, crop nutrient requirements, historical yield data, and environmental conditions.

Benefits of Variable Rate Technology (VRT):

1. Optimized Resource Use: VRT allows farmers to apply inputs more precisely, matching input rates to the specific needs of different areas within the field. By applying inputs where they are most needed and reducing inputs in areas with lower requirements, farmers can optimize resource use efficiency and minimize input wastage.
2. Increased Yield Potential: Tailoring input application rates to match crop requirements can help maximize yield potential by addressing nutrient deficiencies, optimizing soil fertility, and mitigating yield-limiting factors within the field.
3. Cost Savings: VRT helps farmers reduce input costs by avoiding over-application of inputs in areas with sufficient nutrient levels and minimizing input expenditures in low-yield or unproductive areas. By optimizing input use efficiency, farmers can achieve cost savings while maintaining or improving crop yields and profitability.
4. Environmental Benefits: VRT promotes environmental sustainability by minimizing nutrient runoff, reducing soil erosion, and mitigating the risk of groundwater contamination associated with excessive fertilizer and pesticide use. By targeting inputs to areas of need, VRT helps minimize environmental impacts and improve overall sustainability of agricultural practices. Variable rate technology involves the application of inputs, such as fertilizers, pesticides, and irrigation water, at variable rates based on spatial variability within fields. By integrating data from soil maps, yield maps, and other sources, farmers can create prescription maps that specify optimal input rates for different areas of the field. VRT allows farmers to tailor input applications to match the specific needs of crops, optimize resource use, and maximize yield potential while minimizing environmental impacts.
5. Precision Irrigation: Precision irrigation involves the precise application of water to crops based on real-time monitoring of soil moisture levels and crop water requirements. Sensor technology and automated irrigation systems enable farmers to apply water only when and where it is needed, reducing water waste and improving water use efficiency. Precision irrigation helps optimize crop growth, minimize water stress, and mitigate the risk of over-irrigation, soil erosion, and nutrient leaching.
6. Crop Monitoring and Management: Crop monitoring and management applications utilize remote sensing, satellite imagery, and sensor technology to monitor crop health, growth, and development. Farmers can use aerial imagery, drones, and satellite data to assess crop conditions, detect pest infestations, diseases, and nutrient deficiencies, and identify areas of potential yield variability within fields. By monitoring crops continuously and responding to changes in real-time, farmers can implement timely management interventions, such as pest control measures, nutrient applications, and irrigation adjustments, to optimize crop performance and minimize yield losses.

7. Data Analytics and Decision Support Systems: Data analytics and decision support systems integrate data from multiple sources, such as GPS, GIS, remote sensing, and sensor networks, to provide farmers with insights and recommendations for informed decision-making. These systems analyze historical data, predict future trends, and generate actionable recommendations for optimizing agronomic practices, input applications, and resource allocation. By leveraging advanced analytics techniques, such as machine learning and artificial intelligence, farmers can improve yield prediction accuracy, optimize resource use efficiency, and enhance overall farm management.
8. Precision Livestock Farming: Precision farming techniques are also applied to livestock management, including animal health monitoring, feed management, and environmental monitoring. Sensor technology, GPS tracking, and data analytics enable farmers to monitor animal behavior, health status, and performance metrics in real-time. Precision livestock farming helps optimize feed efficiency, reduce disease risks, improve animal welfare, and enhance overall productivity and profitability in animal agriculture.

Precision Irrigation: Precision irrigation is a key component of precision farming that focuses on efficient water management through the use of sensor-based irrigation systems. These systems leverage technology to optimize water application, minimize water waste, and reduce the environmental impact of irrigation practices. Here's how precision irrigation works and its benefits:

1. Sensor-Based Irrigation Systems: Precision irrigation systems utilize sensors to monitor soil moisture levels, weather conditions, crop water requirements, and other relevant parameters in real-time. Soil moisture sensors are installed at various depths within the root zone to continuously monitor soil moisture content. Weather stations or sensors measure environmental factors such as temperature, humidity, wind speed, and solar radiation.
2. Adaptive Water Application: Based on data collected by sensors, precision irrigation systems adjust water application rates and timing to match crop water requirements and soil moisture levels. Automated controllers and actuators regulate irrigation equipment, such as drip irrigation systems, sprinklers, or center pivots, to deliver precise amounts of water only when and where it is needed. By dynamically responding to changing conditions, precision irrigation systems ensure that crops receive the right amount of water at the right time, optimizing water use efficiency and crop productivity.
3. Reduction of Water Waste: Precision irrigation minimizes water waste by avoiding over-irrigation and runoff, which can occur with conventional irrigation methods. By applying water directly to the root zone and avoiding surface runoff, precision irrigation systems reduce water losses due to evaporation, deep percolation, and surface runoff. Soil moisture sensors prevent irrigation when soil moisture levels are adequate, avoiding unnecessary water application and conserving water resources.
4. Environmental Impact Reduction: Precision irrigation helps reduce the environmental impact of agriculture by minimizing water use, nutrient leaching, and soil erosion. By conserving water resources and reducing nutrient runoff, precision irrigation systems protect water quality and aquatic ecosystems. Reduced energy consumption associated with pumping and distributing water also contributes to lower greenhouse gas emissions and environmental sustainability.

III. BENEFITS OF PRECISION IRRIGATION

1. Improved Water Use Efficiency: Precision irrigation optimizes water use efficiency by delivering water directly to the root zone of crops, minimizing losses due to evaporation, runoff, and deep percolation. By matching water application rates to crop water requirements, precision irrigation systems ensure that water is used more efficiently, maximizing crop yields per unit of water applied.
2. Enhanced Crop Performance: By providing crops with optimal moisture levels, precision irrigation systems promote healthy root development, balanced growth, and improved crop vigor. Consistent soil moisture levels help reduce water stress, increase nutrient uptake, and enhance crop resilience to environmental stresses such as drought and heat.
3. Cost Savings: Precision irrigation systems help farmers reduce water, energy, and labor costs associated with irrigation. By optimizing water use efficiency and minimizing input wastage, precision irrigation systems can lead to significant cost savings over time, improving the profitability of agricultural operations.
4. Environmental Sustainability: Precision irrigation contributes to environmental sustainability by conserving water resources, minimizing water pollution, and reducing greenhouse gas emissions. By adopting precision irrigation practices, farmers can minimize their environmental footprint and contribute

to the long-term sustainability of agricultural production systems.

Precision irrigation involves the use of sensor-based irrigation systems to achieve efficient water management, minimize water waste, and reduce the environmental impact of irrigation practices. Here's how precision irrigation accomplishes these goals:

Efficient Water Management: Sensor-based irrigation systems utilize various sensors to monitor soil moisture levels, weather conditions, crop water requirements, and other relevant parameters in real-time. Soil moisture sensors are installed at different depths within the root zone to continuously monitor soil moisture content. Weather stations or sensors measure environmental factors such as temperature, humidity, wind speed, and solar radiation. Based on data collected by sensors, precision irrigation systems dynamically adjust water application rates and timing to match crop water requirements and soil moisture levels. Automated controllers and actuators regulate irrigation equipment, such as drip irrigation systems, sprinklers, or center pivots, to deliver precise amounts of water only when and where it is needed. By optimizing water application rates and timing, precision irrigation systems ensure that crops receive the right amount of water at the right time, maximizing water use efficiency and crop productivity.

Reduction of Water Waste: Precision irrigation minimizes water waste by avoiding over-irrigation and runoff, which are common with conventional irrigation methods. By applying water directly to the root zone and avoiding surface runoff, precision irrigation systems reduce water losses due to evaporation, deep percolation, and surface runoff. Soil moisture sensors prevent irrigation when soil moisture levels are adequate, avoiding unnecessary water application and conserving water resources. Additionally, precision irrigation systems can incorporate technologies such as drip irrigation, which delivers water directly to the root zone of plants, further minimizing water waste.

Environmental Impact Reduction: Precision irrigation systems help reduce the environmental impact of agriculture by conserving water resources, minimizing water pollution, and reducing energy consumption. By optimizing water use efficiency and minimizing nutrient runoff, precision irrigation systems protect water quality and aquatic ecosystems. Reduced energy consumption associated with pumping and distributing water also contributes to lower greenhouse gas emissions and environmental sustainability. Moreover, precision irrigation systems can help mitigate the impacts of climate change by promoting resilience to drought and other extreme weather events through efficient water management practices.

Crop Monitoring and Management: Crop monitoring and management are crucial components of precision farming, and remote sensing plays a significant role in assessing crop health and detecting pests, diseases, and nutrient deficiencies. Here's how remote sensing is utilized for crop monitoring and management in precision farming:

1. Remote Sensing for Crop Health Assessment: Remote sensing technologies, such as satellite imagery, aerial photography, and drones, are used to assess crop health and vigor. These remote sensing platforms capture multispectral or hyperspectral images of agricultural fields, which can reveal subtle variations in crop reflectance associated with different health conditions. By analyzing spectral signatures and vegetation indices derived from remote sensing data, farmers can assess crop health, monitor growth dynamics, and identify areas of stress or decline within fields. Crop health assessment using remote sensing helps farmers make informed decisions about management interventions, such as irrigation scheduling, nutrient application, and pest control.

2. Early Detection of Pests, Diseases, and Nutrient Deficiencies: Remote sensing enables the early detection of pests, diseases, and nutrient deficiencies by identifying spatial patterns and anomalies in crop reflectance. Changes in crop reflectance patterns, such as discoloration, chlorosis, or leaf wilting, may indicate the presence of pests, diseases, or nutrient imbalances. Remote sensing data can be processed using machine learning algorithms and image analysis techniques to detect and classify crop stress factors based on spectral signatures. By detecting pests, diseases, and nutrient deficiencies early, farmers can implement timely management strategies, such as targeted pesticide applications, disease control measures, and foliar nutrient sprays, to mitigate yield losses and minimize economic impacts.

IV. BENEFITS OF REMOTE SENSING FOR CROP MONITORING AND MANAGEMENT

1. Timely Decision-Making: Remote sensing provides timely and spatially explicit information on crop health and stress factors, allowing farmers to make proactive management decisions. Early detection of pests, diseases, and nutrient deficiencies enables farmers to intervene before crop damage becomes severe, maximizing the effectiveness of control measures and minimizing yield losses.

2. Improved Resource Allocation: Remote sensing helps optimize resource allocation by targeting management interventions to areas of need within fields. By identifying spatial variability in crop health and stress, farmers can tailor input applications, such as pesticides, fertilizers, and water, to match the specific requirements of different areas within the field, optimizing resource use efficiency.
3. Enhanced Sustainability: Remote sensing supports sustainable agriculture by promoting integrated pest management (IPM) practices, reducing reliance on chemical inputs, and minimizing environmental impacts. By facilitating precision management practices, such as targeted pesticide applications and site-specific nutrient management, remote sensing contributes to improved environmental sustainability and reduced pesticide residues in food and water.

Data Analytics and Decision Support Systems: Data analytics and decision support systems are essential components of precision farming, enabling farmers to integrate data from multiple sources and make informed decisions to optimize agricultural practices. These systems leverage advanced analytics techniques, such as predictive modeling, to provide insights into crop yield estimation and risk assessment. Here's how data analytics and decision support systems are utilized in precision farming:

1. Integration of Data from Multiple Sources: Precision farming involves the collection and integration of data from various sources, including GPS, GIS, remote sensing, weather stations, soil sensors, yield monitors, and farm management software. Data from these sources provide valuable information on soil properties, weather conditions, crop health, yield variability, and management practices. Data analytics platforms and decision support systems aggregate, process, and analyze data from multiple sources to generate actionable insights for farmers.
2. Informed Decision-Making: Data analytics and decision support systems enable farmers to make informed decisions by providing timely and accurate information on crop conditions, input requirements, and management strategies. These systems analyze historical data, monitor real-time field conditions, and generate recommendations for optimizing agronomic practices, input applications, and resource allocation. By integrating data from diverse sources and applying advanced analytics techniques, farmers can identify patterns, trends, and correlations that inform decision-making and improve farm management practices.
3. Predictive Modeling for Crop Yield Estimation: Predictive modeling techniques are used to estimate crop yields based on historical data, environmental factors, and agronomic variables. Machine learning algorithms, statistical models, and crop simulation models are employed to predict crop yields at different growth stages and across different field zones. Predictive modeling considers factors such as soil fertility, weather conditions, crop genetics, and management practices to generate accurate yield estimates and forecasts. By predicting crop yields in advance, farmers can anticipate production levels, plan harvest schedules, and make informed marketing decisions to maximize profitability.
4. Risk Assessment: Data analytics and decision support systems assess risks associated with agronomic practices, environmental conditions, and market dynamics. Risk assessment models analyze factors such as weather variability, pest and disease pressure, input costs, and market volatility to identify potential risks and vulnerabilities. By quantifying risks and uncertainties, farmers can develop risk management strategies, implement contingency plans, and mitigate the impacts of adverse events on crop production and profitability.

V. BENEFITS OF DATA ANALYTICS AND DECISION SUPPORT SYSTEMS

1. Improved Efficiency: Data analytics and decision support systems streamline farm management processes, optimize resource allocation, and enhance operational efficiency. By automating data analysis tasks and providing actionable insights, these systems enable farmers to make faster, more informed decisions, improving productivity and reducing input costs.

2. Enhanced Productivity: Data-driven decision-making helps farmers optimize agronomic practices, input applications, and crop management strategies to maximize yield potential and enhance overall productivity. By leveraging predictive modeling and risk assessment tools, farmers can identify opportunities for improvement, implement best practices, and achieve higher yields with fewer inputs.

3. Better Risk Management: Data analytics and decision support systems help farmers assess and mitigate risks associated with crop production, market fluctuations, and environmental factors. By identifying potential risks and vulnerabilities, farmers can develop risk management strategies, diversify operations, and improve resilience to adverse events, enhancing long-term sustainability and profitability.

4. Sustainable Agriculture: Data analytics and decision support systems support sustainable agriculture by promoting precision management practices, optimizing resource use efficiency, and minimizing environmental impacts. By optimizing input applications and reducing input wastage, farmers can conserve natural resources, minimize environmental pollution, and promote ecosystem health and biodiversity.

Precision farming offers a range of benefits that can revolutionize agricultural practices, but it also faces significant challenges that need to be addressed for widespread adoption.

Benefits:

1. Increased Productivity and Resource Efficiency: Precision farming optimizes resource use by applying inputs, such as water, fertilizers, and pesticides, precisely where and when they are needed. By tailoring management practices to match specific field conditions, precision farming maximizes crop yields while minimizing input wastage.
2. Reduced Environmental Impact: Precision farming practices help mitigate environmental degradation by minimizing nutrient runoff, soil erosion, and chemical leaching. By promoting sustainable agricultural practices, precision farming contributes to soil health, water quality, and biodiversity conservation.
3. Enhanced Profitability and Sustainability: Precision farming improves farm profitability by reducing input costs, increasing yields, and optimizing operational efficiency. By adopting precision farming practices, farmers can achieve higher returns on investment while promoting long-term sustainability and resilience in agriculture.

Challenges:

1. Initial Investment Costs: The adoption of precision farming technologies often requires significant upfront investments in equipment, sensors, software, and training. High initial costs may pose barriers to adoption, particularly for small and medium-sized farmers with limited financial resources.
2. Data Privacy and Security Concerns: Precision farming involves the collection, storage, and analysis of sensitive data related to farm operations and management. Data privacy and security concerns, including unauthorized access, data breaches, and misuse of information, need to be addressed to maintain trust and confidence in precision farming technologies.
3. Accessibility and Adoption Barriers in Developing Regions: Limited access to technology, infrastructure, and technical support services may hinder the adoption of precision farming practices in developing regions. Challenges such as inadequate internet connectivity, lack of skilled manpower, and insufficient funding for technology investments need to be overcome to promote inclusive adoption of precision farming.

VI. FUTURE PROSPECTS

The future prospects of precision farming are promising, driven by advancements in technology, data analytics, and the potential to address global food security challenges. Here are some key areas of development and opportunities for precision farming:

1. Advancements in Technology and Data Analytics: Continued advancements in sensor technology, remote sensing, and data analytics will enable more precise and efficient monitoring and management of agricultural systems. Emerging technologies such as Internet of Things (IoT), unmanned aerial vehicles (UAVs), and blockchain will further enhance data collection, connectivity, and traceability in precision farming. Integration of advanced sensors, robotics, and automation will enable autonomous farm operations and real-time decision-making, improving productivity and sustainability.
2. Integration of Artificial Intelligence and Machine Learning: Artificial intelligence (AI) and machine learning (ML) algorithms offer new opportunities for predictive modeling, pattern recognition, and decision support in precision farming. AI-driven systems can analyze large datasets, identify trends, and generate actionable insights for optimizing agronomic practices, input applications, and resource allocation. ML algorithms can learn from historical data, adapt to changing conditions, and optimize management strategies, leading to more efficient and adaptive precision farming systems.
3. Potential for Addressing Global Food Security Challenges: Precision farming has the potential to contribute significantly to global food security by increasing agricultural productivity, improving resource use efficiency, and reducing food losses. By optimizing crop yields and minimizing input wastage, precision farming practices can help meet the growing demand for food in a sustainable and environmentally responsible manner. Precision farming technologies can also enhance resilience to climate change, mitigate the impacts of extreme weather events, and improve the adaptive capacity of agricultural systems.

4. Sustainable Intensification and Resilience: Precision farming enables sustainable intensification of agricultural production, allowing farmers to produce more food on existing land while minimizing environmental impacts. By adopting precision farming practices, farmers can enhance the resilience of agricultural systems to climate variability, water scarcity, and other challenges, ensuring food security for future generations.
5. Adoption of Digital Agriculture Platforms: Digital agriculture platforms, powered by cloud computing, big data analytics, and mobile technologies, will facilitate the adoption and scaling of precision farming practices. These platforms provide farmers with access to real-time data, decision support tools, and agronomic services, empowering them to make informed decisions and optimize farm operations. Digital agriculture platforms also foster collaboration, knowledge sharing, and innovation across the agricultural value chain, driving continuous improvement and transformation in agriculture.

VII. CONCLUSION

In conclusion, precision farming represents a transformative approach to agriculture, characterized by the targeted application of inputs, data-driven decision-making, and the integration of advanced technologies. This review has highlighted the various methods and technologies employed in precision farming, including Global Navigation Satellite Systems (GNSS), Geographic Information Systems (GIS), remote sensing, sensor technology, and data analytics. By leveraging these tools, precision farming enables farmers to optimize resource use, enhance productivity, and reduce environmental impacts. Looking ahead, the future prospects of precision farming are promising, with advancements in technology, integration of artificial intelligence and machine learning, and the potential to address global food security challenges. As precision farming continues to evolve, it holds the key to unlocking sustainable and resilient agricultural systems that can meet the growing demand for food while preserving natural resources for future generations.

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Effect of Methanol Blends on performance of Two Stroke petrol Engine at varying load conditions

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Abstract— *Experiments were conducted to evaluate the performance evaluation of two stroke single cylinder, spark ignition (SI) engine, with methanol blended gasoline (40% gasoline, 20% methanol, & 10 % by volume). Performance parameters (brake horse power, specific energy consumption, thermal efficiency,) were determined at varying load condition it is observed that The experiments shows that 10% methanol blend with gasoline gives the best performance for the SI engine. The power output of the engine is almost near about the petrol on blend M10. It is recorded that the higher power output is 2. 85 KW with M10 blends at load 8 Kg as compare to petrol and the lowest power output is from the engine is 0. 82 KW with blend M40 at load 2 Kg. The specific fuel consumption decrease as load increase for petrol and methanol blends. The SFC of methanol (M10) is higher at low load condition and low at high load conditions as compare to other blends.*

Keywords— ***Gasoline, Methanol, specific fuel consumption. Brake horse power, Brake thermal efficiency, Engine.***

I. INTRODUCTION

In recent years, the automotive industry has been confronted with the urgent need to mitigate environmental pollution and reduce dependence on fossil fuels. This imperative has spurred research into alternative fuels and technologies that offer cleaner and more sustainable solutions. Among these alternatives, methanol has emerged as a promising candidate due to its renewable nature, lower carbon footprint, and potential to enhance engine performance when blended with conventional petrol.

A petrol engine (Known as a gasoline engine in America English) is an internal combustion engine with spark-ignition, designed to run on petrol (gasoline) and similar volatile fuels. Alcohols have been suggested as an engine fuel almost since automobile was invented. The alcohol used to change/modify the attitude toward the present fuel, i.e., gasoline and search for new alternatives. A major contributor to the greenhouse effect is the transport sector due to the heavy and increasing traffic level. In spite of ongoing activity to promote efficiency, the sector is still generating significant, especially in developing countries fairly drastic political decisions may have be taken to address this problem in the future. Furthermore, the dwindling supply of petroleum fuels will sooner or later become a limiting factor. An important step in efforts to solve the problem is to replace fossil source energy with Bio energy. In the transport sector this means either source energy with bio energy. In the transport sector this means either introducing bio fuels and using adapted vehicles, or

blending bio fuels with petroleum – based fuels for use with present vehicle fleets.

The performance of an engine, characterized by parameters such as power output, fuel efficiency, and emissions, is influenced by numerous factors, including fuel composition, operating conditions, and engine design. Understanding how methanol blends affect these performance metrics under varying load conditions is crucial for optimizing engine operation and developing strategies for cleaner and more efficient combustion processes.

II. MATERILS AND METHODS

The tests were conducted on a two-stroke, single-cylinder, and spark-ignition gasoline engine at varying load, in AKS University Satna, Madhya Pradesh.

2.1 Engine setup

The engine is water-cooled, self governed and use forced lubrication system. To assess the performance, one should obtain the input (energy input through the fuel I. e. Petrol) and output (the energy available at the output shaft) relationship .To measure using the fuel controlling arrangement and stopwatch. The output measured using an absorption type dynamometer. The brake drum of the dynamometer is broken using a wire rope-wound around the drum-with one end connected to a spring balance between and the other end to a hanger and dead weights. The friction between the dram and rope can be varied by changing the net tension (difference between the weight and the spring balance reading), by tightening the spring balance end as well as adding more dead weight. The heat energy due to friction is to be remove by cooling the drum-I.e. By circulating cooling water through the groove inside the drum. In short, the load is varied rope through the fixture arrangement.

Fig 2.1. Experimental engine setup

Table 2.1 Specification of engine

PARAMETER	DETAILS
Engine company and model type	Make-bajaj Model-chetak Type-single cylinder,2 stroke petrol,
Cooling system	Air cooled
Cylinder Number	Single cylinder
No. of gear	4

RPM	5000rpm
Stroke	57mm
Bore	57mm
Capacity	144.45CC
Compression Ratio®	4:5:1
Max. Torque	1.1 kgfm @ 3500rpm
Weight of engine	103kg
Top Speed	85kmph
Rope Dia.	1.7 cm
Brake drum Dia.	35 cm
Fuel Tank capacity	6.50liters

2.2 Fuel Input measuring arrangement

These arrangements consist of a fuel tank of suitable capacity mounted on a stand. The fuel goes to the engine through a 50 ml burette. The burette facilitates the measurement of fuel consumption for a define period with the help of stop watch. The fuel I. e. petrol and ethanol, was measured using a calibrated burette (of capacity 50c. c) and a stopwatch.

Fig .3.2 Fuel input measuring arrangement

2.3 Parameter evaluated

The aim of study the performance evaluation of the engine with different blends of ethanol with Petrol-Methanol and plotting the characteristics such as

2.3.1 Specific fuel consumption (SFC)

2.3.2 Brake Horse Power (BHP)

2.3.3 Brake thermal efficiency (BTE)

2.3.1 Specific fuel consumption (SFC)

Specific fuel consumption is defined as the fuel flow rate per unit power output. It is a measure of the efficiency of the engine in using the fuel supplied to produce work. It is desirable to obtain a lower value of SFC meaning that the engine uses less fuel to produce the same amount of work. This is one of the most important parameter to compare variable fuel. It is expressed in kg/kW Hour

$$\mathbf{SFC} = \frac{\mathbf{Mf}}{\mathbf{BHP}}$$

Where,

Mf = mass of fuel in kg/KW/hour

BHP = Brake horse power in kW

2.3.2 Brake Horse Power (BHP)

Brake horse power is one of the most important measurements is the test schedule of an engine. The net available at the shaft is known as brake power. It is define as rate of doing work is equal to the product of force liner velocity of the product of torque and angular velocity. Thus, the measurement of power involve the measurement of force(or torque) as well as speed. Un electrical loading dynamometer used for measuring brake power of engine. It is measured in kW.

$$\mathbf{BHP} = \frac{2 \times \pi \times N \times Te}{60 \times 1000}$$

N= Speed of the engine in rpm

Te= torque in Nm = w × R-(9. 81× Net mass applied in kg) x radius in m

2.3.3 Brake Thermal Efficiency (BTE)

A measure or overall efficiency of the engine is given by the brake thermal efficiency. BTE is the of energy in the brake power to the fuel energy

$$\mathbf{BTE} = \frac{BHP \times 3600}{Mf \times CV}$$

Where,

B. H. P = Brake horse power in kW.

Mf = Mass of fuel in kg/Hr.

CV = Calorific value in kJ/kg.

2.4 Engine Speed (rpm)

Speed is a rate variable defined as the time rate of motion. It may be linear, I. e., along the axis of moment or angular I. e., around the axis. The angular speed is measured by tachometer. Tachometer is an instrument used to measure angular velocity of the shaft either by registering the number of rotation during the period of contain or by indicating directly the number of rotation per minute. It indicated the value of rotary speed display a reading of an average speed. An Emerson make digital panel tachometer was used for measurement of engine rpm. It has a measuring range of 1 to 2000 rpm.

Fig. 2.3 Measurement of engine rpm

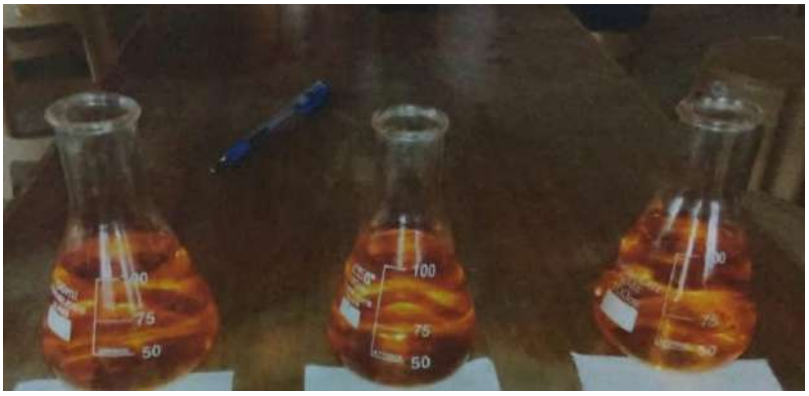

Fig 2.4 Petrol blends with different proportion of methanol

III. RESULT AND DISCUSSION

The experiment were conducted using petrol and methanol blends M10, M 20, M 40 and performance of the engine was evaluated using several parameter such as brake power, specific fuel consumption and thermal efficiency.

3. Performance evaluation of gasoline engine using petrol and methanol blends at varying load

3.1 Brake Power (KW) at varying load with different blends of petrol – methanol

The test was conducted for pure gasoline fuel which was blend base line fuel and then for different blends of gasoline – methanol such as M10, M20 and M40 samples. Fig 4.1 shows the variation in engine break power at varying load with petrol and methanol blends M10, M20 and M40. The power output of the engine is almost near about the petrol on blend M10. It is recorded that the higher power output is 2.85 KW with the blend M 10 at load 8 kg as compare to petrol and the lowest power output is from engine is 0.82 KW with blend M40 at load 2 kg. it was concluded that the blends the blend with 10 % have no more effect on power output. It also shows that the percentage of blends increase with petrol the brake power decrease if increase in load. Table 4.1 shows the M10 blends give higher brake power at all loads as compare to other blend M20, M40. It is recorded that M10 out power is 0.92 KW,1 .72Kw, 2.25 KW and 2.85 KW respectively at varying load 2 to 8 Kg.

Table 3.1 Break power at varying load with different blends of petrol-Methanol.

Load, kg	Petrol	M 10	M 20	M 40
	Break Power, KW			
2	0.98	0.92	0.89	0.82
4	1.82	1.72	1.59	1.43
6	2.3	2.25	2.1	1.91
8	2.96	2.85	2.4	2.25

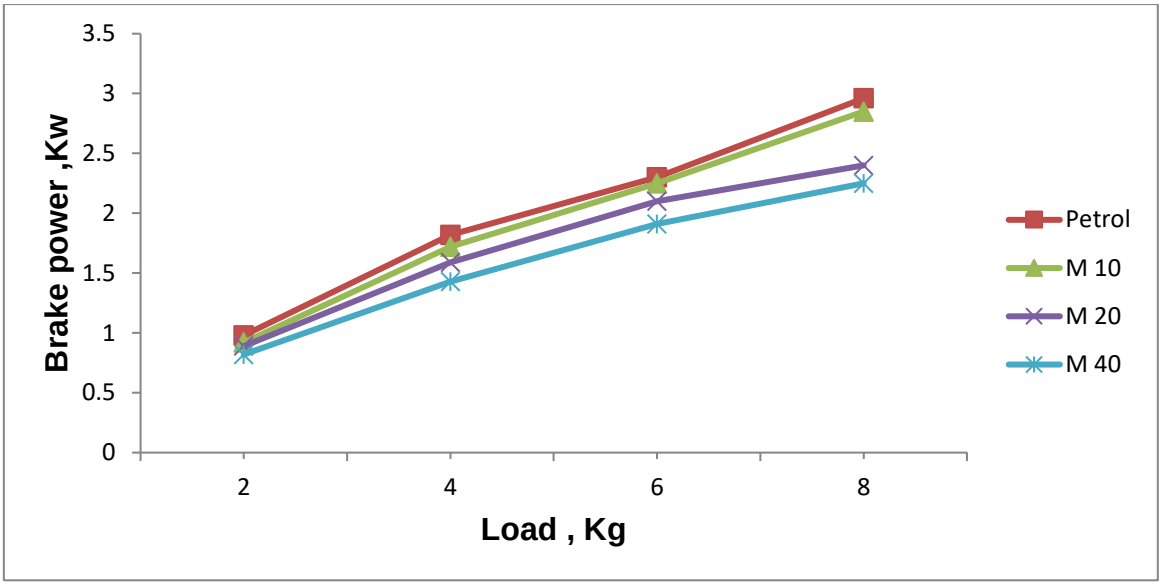

Fig .3.1 Break power at varying load with different blends of Petrol- Methanol

3.2 Specific Fuel consumption at varying load with different blends of Petrol methanol.

The test was conducted for pure gasoline fuel which was base line fuel and then for different blends of gasoline-Methanol such as M10, M20, and M40 samples. Fig 4. 2 and table 4. 2 represent the effect of methanol blend with petrol on specific fuel consumption at different load condition. It shows the specific fuel consumption decrease as load increase for petrol and methanol blends. The SFC of Methanol (M10) is higher at low load condition and low at high load conditions as compare to other blends. The specific fuel consumption of Methanol blend (M10) is nearly about petrol at lower load condition this is due to the inherent oxygen contents of Methanol. Fig 4.2 show the specific fuel consumption of methanol blend (M10) is higher at lower load 2 kg is 0. 74 kg/kW/h and the lower SFC of methanol blend (M10) is 0. 49 kg/kW/h.

Table 4. 2 Show the specific fuel consumption of ethanol (M 10) is lower at all load condition as compare to other blends. It is 0. 74 kg/kW/h, 0. 64 kg/kW/h, 0. 61 kg/kW/h and 0. 49 kg/kW/h respectively at load between 2 to 8 kg.

Table 3.2 Specific Fuel Consumption at varying load with different blends of petrol-Methanol.

Load , kg	**Petrol**	**M 10**	**M 20**	**M 40**
	Specific Fuel Consumption			
2	0.72	0.74	0.76	0.79
4	0.61	0.64	0.59	0.72
6	0.58	0.61	0.63	0.65
8	0.43	0.49	0.54	0.59

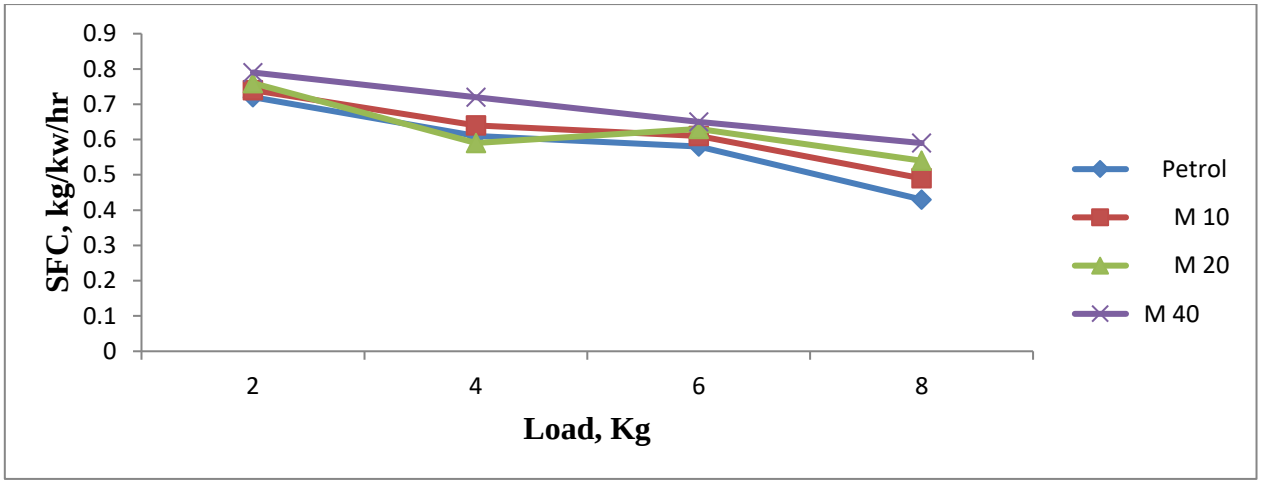

Fig. 3.2 Specific fuel Consumption at varying load different blends of Petrol-Methanol

3.3 Break thermal efficiency at varying with different blends of Petrol-Methanol

The test was conducted for pure gasoline fuel which was based line fuel and then for different blends of gasoline-methanol such as M 10, M 20, and M 40 samples .Fig 4.3 present the effect of using Methanol-petrol blends on break thermal efficiency. As shown in the figure, thermal efficiency increases as the percentage the effect of methanol increases, the maximum thermal efficiency is recorded with blend of 40% methanol in the petrol at all loads as compare to other blends the thermal efficiency increases because of better combustion due to higher octane no as compared to petrol. The maximum thermal efficiency is recorded 22.8% with the blends of M 40 at 8 kg load and the minimum thermal efficiency with recorded 10.7 with M10 at load 2 kg. Table 4.3 represent the thermal efficiency with different blends at all varying load the thermal efficiency is recorded maximum in M 40 it is 12% , 16.2% , 21.5% and 22.8 % at load between 2 to 8 kg .

Load , kg	Petrol	M 10	M 20	M 40
	Break Thermal Efficiency			
2	10.2	10.7	11.1	12
4	13.8	14.3	15	16.2
6	16.9	18.3	19.1	21.5
8	18.6	19	20.9	22.8

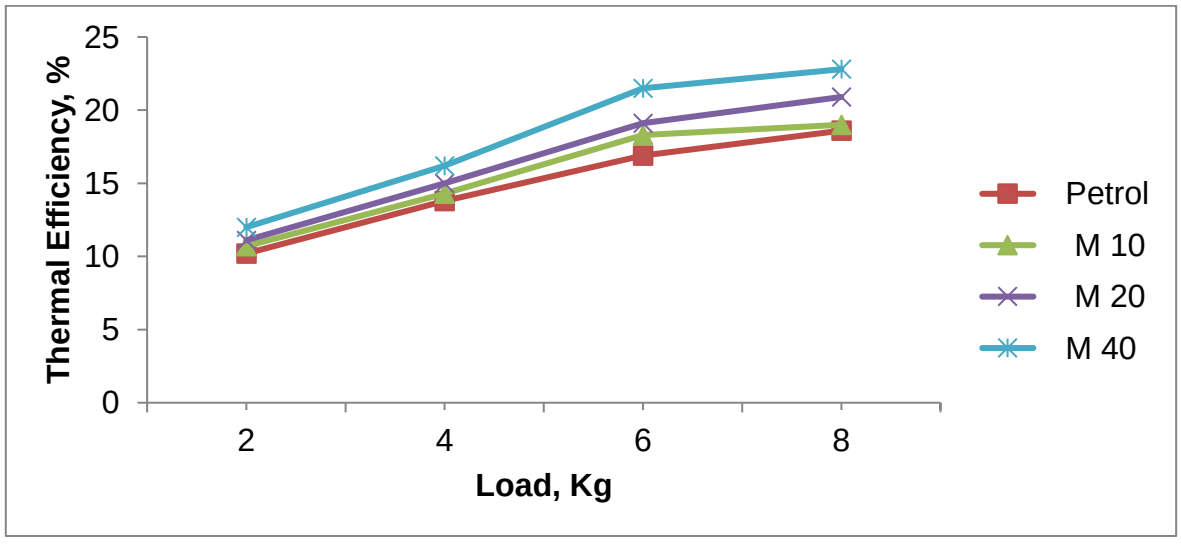

Fig 3.3 Break thermal efficiency at varying load with different blends of Petrol –Methanol

IV. CONCLUSION

The experimental performance on Sl engine with different ethanol blends with gasoline was investigated. The main outcomes of this analysis are as:

The experimental performance on Sl engine with different ethanol blends with gasoline was investigated. The main outcomes of this analysis are as:

- The experiments shows that 10% methanol blend with gasoline gives the best performance for the SI engine.
- Methanol blended with gasoline always improves the performance of SI engine and reduced the exhaust emissions also.
- It is concluded that the percentage of blends increase with petrol the brake horse power decrease at all varying load. The power output of the engine is almost near about the petrol on blend M10. It is recorded that the higher power output is 2. 85 KW with M10 blends at load 8 Kg as compare to petrol and the lowest power output is from the engine is 0.82 KW with blend M40 at load 2 Kg.
- The specific fuel consumption decrease as load increase for petrol and methanol blends. The SFC of methanol (M10) is higher at low load condition and low at high load conditions as compare to other blends.
- Thermal efficiency increases as the percentage of methanol increases. The maximum thermal efficiency is recorded with blend of 40% methanol in the petrol at all loads as compare to other blends which is observed that 22.8%.

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Extraction of Eugenol in Clove Extract and Evaluation of its Antioxidant Activity

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Abstract— *Higher total phenolic content was observed in steam-distilled clove extract compared to its oleoresin. Ensuring antioxidant activity of steam distilled and clove oleoresin using β-carotene–linoleic acid model system was found to be 85.51±0.19% and 77.88±0.15%, respectively, at a level of 200 ppm. The radical-scavenging activity of steam-distilled extract and oleoresin of clove were 88.93±0.23% and 80.84±0.36%, respectively, at the same level of 200ppm by using the DPPH method. Eugenol content in the steam-distilled extract of clove (0.518±0.005mg/ml) was significantly higher than that of clove oleoresin (0.433±0.007mg/ml). Recovery of eugenol content affected by the period of refluxing and clove extract refluxed for 4 hr found to be highest recovery in the steam distilled clove extract (0.763±0.007) than its clove oleoresin (0.635±0.020) with 13.904 min retention time using RP-HPLC. The steam-distilled clove extract was found to have the highest antioxidant activity than its oleoresin counterparts.*

Keywords— ***Antioxidant, β-carotene–linoleic acid, DPPH, eugenol, phenolic content.***

I. INTRODUCTION

India, Madagascar, Sri Lanka, Indonesia, and southern China grow clove (Syzygium aromaticum) (Bhuiyan et al., 2010). Distilling clove flowers, stems, or leaves yields clove oil (Anderson et al., 1997; Mylonasa et al., 2005). The USFDA lists clove oil as "GRAS" (Generally Regarded As Safe) at no more than 1500 ppm in all food categories (Kildeaa et al., 2004). The WHO Expert Committee on Food Additives recommends 2.5 mg/kg body weight of clove oil per day. Kildeaa et al., (2004) found it safe, efficacious, and affordable. Clove extract contains eugenol (4-allyl-2-methoxyphenol), which accounts for 70-90% of its weight (Keene et al., 1998), as well as eugenol acetate (17%) and β caryophylene (12%). Eugenol has various applications, including antioxidant and antibacterial properties (Rajakumar and Rao, 1993). It has more therapeutic features such as digestive stimulation, anti-inflammatory, antibacterial, anticarcinogenic, hypolipidemic, and antimutagenic actions (Aaby et al., 2004). In addition, it possesses anti-vomiting, analgesic, antispasmodic, anti-carminative, kidney-reinforcing, and antiseptic (Liu et al., 1997). It may prevent cancer due to its antioxidant characteristics, according to Lee and Shibamoto (2001). It contains calcium, iron, phosphorus, sodium, potassium, vitamin A, and vitamin C. For decades, clove oil has been used to treat toothaches, headaches, and joint discomfort (Shelef, 1983; Soto and Burhanuddin, 1995). It is used worldwide for food flavouring and dental local anaesthetic (Anderson et al., 1997).

Given the popularity of nutraceuticals and their nutritional benefits, clove extract's antioxidant properties should be assessed. The present work examined the antioxidant activity of steam-distilled clove extract, clove oleoresin, and their antioxidant component, eugenol. Three antioxidant experiments examined steam-distilled and

clove oleoresin antioxidant activity. We also examined the antioxidant activity of the major eugenol in steam-distilled clove extract and clove oleoresin using RP-HPLC.

II. MATERIALS AND METHODS

2.1 Clove Extract:

Clove oleoresin was obtained from Synthite Industry Ltd. in Kerala, India, while the steam-distilled extract was provided by Katyani Exports in Delhi, India.

2.2 Determination of antioxidant capacity: To assess antioxidant potential, total phenolic content, β carotene linoleic acid model system, and radical scavenging assays were used (Prior et al., 2005).

2.3 Total phenolic content

Folin-Ciocalteu's technique was used to determine the total phenolic content of clove oleoresin and steam distilled clove extract (Kahkonen et al., 1999). In a test tube, 400 µl of adequately diluted sample/gallic acid standard was placed. It was combined using a vortex mixer after adding 2000 µl of diluted Folin-Ciocalteu's reagent. After 3 minutes, 1600 µl of sodium carbonate solution was added and incubated at room temperature for 30 minutes in the dark. Instead of sample, 400 µl of distilled water was used for blank preparation. Using a Spectrophotometer (Specord 700), the absorbance of the samples was measured against a blank at 765nm. The concentration of gallic acid (400 µl of 10-100 g/ml) was collected in place of the sample for standard curve preparation and quantified with regard to the standard curve. The outcomes were expressed as gallic acid equivalents (GAE), milligrams per gm of spice extract.

2.4 β-carotene–linoleic acid model system

The antioxidant activity of solvent extracts was evaluated using the procedure described by Marco (1968), with slight modifications. β-carotene (0.2 mg) in chloroform (0.5 ml) was mixed with 20 mg linoleic acid and 200 mg Tween 40. Nitrogen gas was used to evaporate the chloroform at 40°C. The resultant solution was immediately diluted with 10 ml of double distilled water, and the emulsion was thoroughly stirred for one minute using a magnetic stirrer. The emulsion was diluted further with 40 mL of distilled water. Aliquots (4 mL) of this reagent were placed into separate stopper test tubes holding 1 mL of the necessary amount of ethanol-based sample extracts. At a rate of 200 ppm, the steam-distilled and oleoresin of clove extract dissolved in ethanol. A control was created using 1 ml of ethanol and 4 ml of emulsion. Optical densities of all samples were tested immediately (t = 0), again after 15 minutes, then every 30 minutes for the next 3 hours (t = 180). In between observations, the tubes were submerged in a water bath set at 50°C. After that, it was independently added to a model system of β-carotene linoleic acid, and the activity was measured spectrophotometrically 700 (Specord) at 470 nm. The following expression was used to assess the extracts antioxidant activity (AA) in terms of the photo-oxidation of beta-carotene. AA is represented by the antioxidant activity, A_o is the initial absorbance of sample, A_t is the absorbance of sample after time t, $A0_o$ is the initial absorbance of control, and $A0_t$ is the absorbance of control after time t respectively.

$$AA = 100\left[1 - \frac{A0 - At}{A00 - A0t}\right]\%$$

2.5 Radical-scavenging activity by DPPH model system

According to the method described by Blois (1958), the extracts of steam-distilled and oleoresin clove were taken at a rate of 200 ppm, dissolved in ethanol, and tested for their capacity to scavenge free radicals in the DPPH system. Test tubes containing 200 ppm of sample extract diluted in ethanol (1 ml) were then filled with 4 ml of a 0.1 mM ethyl acetate solution of DPPH and shaken vigorously. Tubes were left at 27°C for 20 min. Without any additional extract, the control was made as described above, and ethanol was used to adjust the baseline. Optical densities (OD) of the samples were measured spectrophotometrically (Specord 700) at 517 nm. Radical-scavenging activity was expressed as % inhibition percentage.

$$\%\ \text{Radical scavenging activity} = \frac{\text{Control OD} - \text{Sample O}}{\text{Control OD}}$$

2.6 Extraction and quantification of eugenol from clove extract

The preparation of the sample is the first and most important step. It is necessary to extract the appropriate chemical components from the spice materials in order to proceed with further separation and characterization. 30 mg of clove extracts (steam distilled and oleoresin clove) was taken in a round-bottom flask, 30 ml of ethanol was added to the flask and allowed to reflux. Using a water reflux condenser, the sample was allowed to reflux for varying amounts of time ranging from 2 to 5 hours. Following the process of refluxing, the mixture was evaporated on a water bath until it reached the desired level of dryness. After re-dissolving the resulting solution in 30 mL of ethanol, it was filtered using a PTFE (Polytetrafluoroethylene) syringe filter with a 0.45-m pore size and then immediately injected into an HPLC system in a volume of 20 µL. Quantification was performed based on the standard curve of eugenol concentration in ethanol, which ranged from 0.25 to 1.5 mg/10 ml. At a wavelength

of 280 nm, the analysis was carried out using a C18 column (5 µm, 4.5×250mm, 100 A^0). The mobile phase consisted of methanol and water at a volumetric ratio of 60:40. The temperature of the column was maintained at 30^0C and the flow rate was held constant at 0.8ml/min.

III. RESULTS AND DISCUSSION

3.1 Total Phenolic Content by Folin-Ciocalteu method

The Folin–Ciocalteu method (Kahkone et al., 1999) was used to measure the total amount of phenolic substance. A standard curve of gallic acid (ranging from 10 to 100 µg/ml) was prepared to evaluate the overall phenolic content of oleoresin and steam-distilled clove extracts and results were expressed as mg gallic acid equivalents (GAE) per gram of spice extract. Table 1 displayed the results that oleoresin had a GAE of 177.039±0.35mg and the steam-distilled clove extract had a GAE of 256.506±0.45mg/gm respectively. The reason for this difference was probably because different extracts contained different antioxidant chemicals that bind to water and fat (flavonoids, terpenoids, carotenoids, phytoestrogens). Zhou et al. (2011) looked at the hyrophillic and lipophillic antioxidant activity of loquat fruits and found that the overall antioxidant activity and phenolic content were linked to hydrophilic antioxidant compounds in a good way. Vicas et al. (2009) found similar results by using FRAP (ferric tripyridyltriazine complex) of mistletoe (Visum album) had about 100 times less lipophillic antioxidant activity (LAA) than hydrophilic antioxidant activity (HAA). Also, HAA was linked to higher levels of total phenolics in the leaves (R2=0.9363) and stems (R2=7337) of mistletoe (Visum album). In 2005, Shan et al. said that clove bud's main phenolic substances are phenolic acids (gallic acid), flavonolglucosides, tannin, and phenolic volatile oils (eugenol and acetyl eugenol).The phenolic chemical content of clove extract that was made by steam distillation was higher. It also depends on the method used to separate the oil and how well the different parts of the oil dissolve (Reverchon, 1997; Mostafa et al., 2004). A study by Ozka et al. (2012) found that a steam-distilled extract of the Rosa damascena flower had a high phenolic content.

Table 1. Total phenolic content of steam distilled and clove oleoresin

Samples	Total phenolic content (mg of GAE/gm)
Clove Oleoresin	177.039±0.35[b]
Steam distilled	256.506±0.45[a]

Data are presented as means ± SEM (n=3).

a-bMeans with different lowercase superscripts letters are significantly different (P < 0.05) from each other.

3.2 Antioxidant activity by β-carotene–linoleic acid model system

Table 2 shows the antioxidant activity of clove extract (steam distilled and oleoresin) at 200 ppm using β-carotene-linoleic acid linked oxidation model system (Marco, 1968). At a concentration of 200 ppm, the antioxidant activity of steam distilled clove extract (84.950±0.23%) was substantially (P>0.05) higher than that of oleoresin (77.886±0.31%). This difference can be attributed to the difference in total phenolic content of steam distilled clove extract and clove oleoresin, which is dependent on extraction method and solubility of oil components. This is consistent with the findings of Mostafa et al., (2004), who found that super critical fluid (SFE) extraction increases the yield of phenolic compounds due to an increase in the solubility of oil components.

Table 2. Antioxidant activity of steam distilled and clove oleoresin

Samples	Antioxidant activities (%) at 200ppm
Clove oleoresin	77.886±0.15[b]
Steam distilled clove extract	85.510±0.19[a]

Data are presented as means ± SEM (n=3).

a-bMeans with different lowercase superscripts letters are significantly different (P < 0.05) from each other.

3.3 Radical-scavenging activity by DPPH assay

The radical-scavenging activity of clove extracts (steam distilled and oleoresin) was tested in the DPPH system at 200 ppm and the findings are shown in Table 3. Steam distilled clove extract and its oleoresin were shown to have radical-scavenging activities of 88.935±0.23% and 80.841±0.36%, respectively. The radical-scavenging potential of steam distilled clove extract was found to be considerably (P>0.05) higher than that of its oleoresin counterpart. This disparity in radical-scavenging (antioxidant) activity could be attributed to differing hydrophilic and lipophillic antioxidant chemicals. Another reason could be that the total phenolic content of steam distilled clove extract and clove oleoresin differs. The results of the radical-scavenging (antioxidant) assay were similar with the data obtained from the total phenolic

content and the β-carotene-linoleic acid model system. Our findings suggested that there was a positive relationship between phenolic content and antioxidant activity, since the higher activity of the steam distilled clove extract may be related to its higher phenolic content.

Table 3: Radical-scavenging activities of clove extracts (steam distilled and oleoresin) as expressed in % inhibition

Samples	% Inhibition
Clove oleoresin	80.841±0.36[b]
Steam distilled clove extract	88.935±0.23[a]

Data are presented as means ± SEM (n=3).

a-bMeans with different lowercase superscripts letters are significantly different (P < 0.05) from each other.

3.4 Quantitative analysis of eugenol content in clove extracts (oleoresin and steam distilled) by RP-HPLC.

Eugenol (4-allyl-2-methoxyphenol), a well-known clove phenolic component, has numerous therapeutic effects, including antioxidant action. Eugenol was measured using a modified HPLC method established by Yun et al., (2010). The extraction process was optimized for quantitative analysis to improve the sensitivity and selectivity for separation of eugenol content.

3.4.1 Optimization of eugenol extraction using two different solvent

The extraction technique of eugenol content was standardized, and the efficiency of several extraction solvents from clove extracts (steam distilled and oleoresin) were assessed in order to get chromatograms with superior resolution of neighboring peaks in a short period. The eugenol content of steam distilled clove extract and clove oleoresin was extracted using two different solvents, methanol (absolute) and ethanol (95%). The results showed that ethanol extraction yielded more eugenol than methanol extraction. As a result, ethanol was chosen as the extraction solvent for determining eugenol. Also, for industrial uses, ethanol is probably preferable to methanol since the resulting solvent residues are less hazardous. The results are shown in Fig 1.

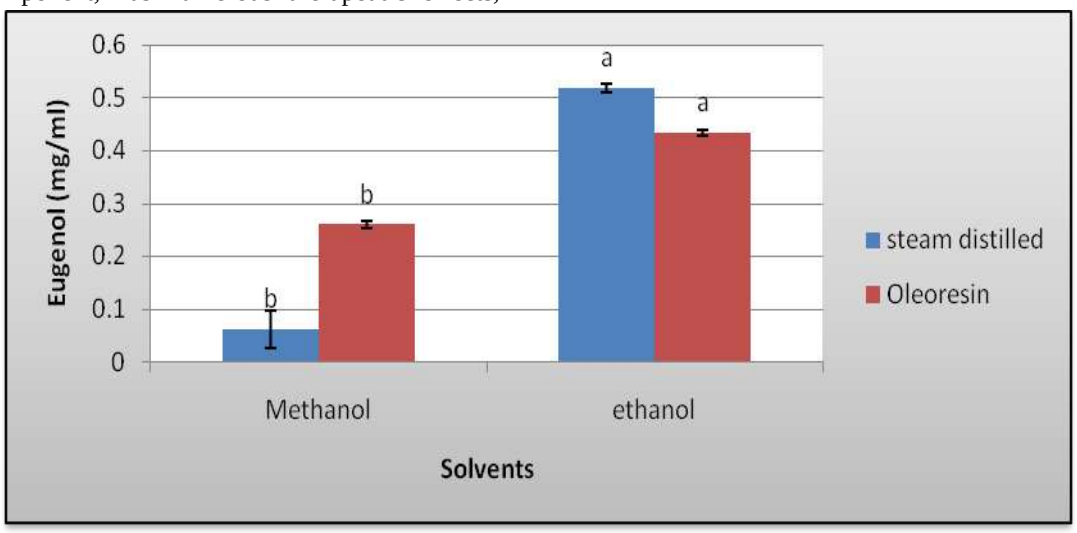
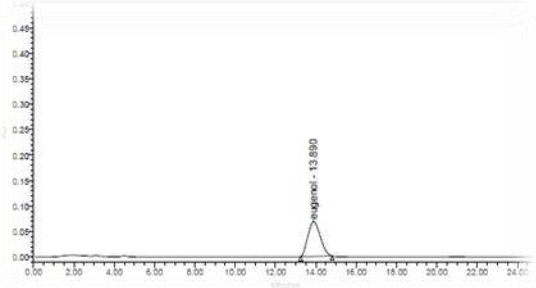
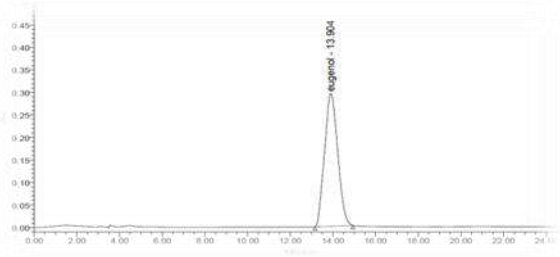


Fig.1: Two different solvent used to estimate eugenol content from clove extracts (oleoresin and steam distilled) as expressed in (mg/ml)

The mean changes between the samples were analyzed by one-way ANOVA.a-b Means with different lowercase superscripts letters are significantly different (P < 0.05) from each other. Error bars show the variations of three determinations in terms of standard error of mean.



Eugenol obtained by using Methanol



Eugenol obtained by using ethanol

3.4.2 Optimization of eugenol extraction using two different solvent different time interval of refluxing period

To establish the ideal extraction time for eugenol content, several refluxing times at different intervals such as 2 hr, 4 hr, and 5 hr were tried in terms of eugenol content extraction. The steam distilled clove extract and oleoresin were refluxed with each solvent for 2 to 5 hours using a water reflux condenser. Following the refluxing, the samples were evaporated on a water bath until dry. The resulting solution was redissolved with the solvent and filtered through a 0.45m PTFE syringe filter before being injected directly into the HPLC apparatus. The analysis was carried out at a flow rate of 0.8ml/min at a column temperature of 30°C. The eugenol concentration of oleoresin and steam distilled clove extract was determined using a eugenol standard curve (range from 0.25 to 1.5 ppm). Eugenol had a retention time of 13.904 ± 0.5 minutes. It was discovered that raising the refluxing time from 2 hours to 4 hours increased the eugenol content, whereas increasing the refluxing time to 5 hours lowered the eugenol content. As shown in fig 2, the quantitative HPLC results demonstrate that the 4 hr extraction time has the highest value of eugenol concentration in the steam distilled clove extract (0.763±0.007) than its clove oleoresin (0.635±0.020). Aside from the solvent and method of extraction, environmental factors and different species may also account for the variance in extract activity. Fig 2 shows that the eugenol concentration of steam distilled clove extract was found to be higher than that of its oleoresin counterparts. Selected solvents, such as 95% ethanol, were employed because they were stable and compatible with reversed-phase HPLC separation of eugenol content.

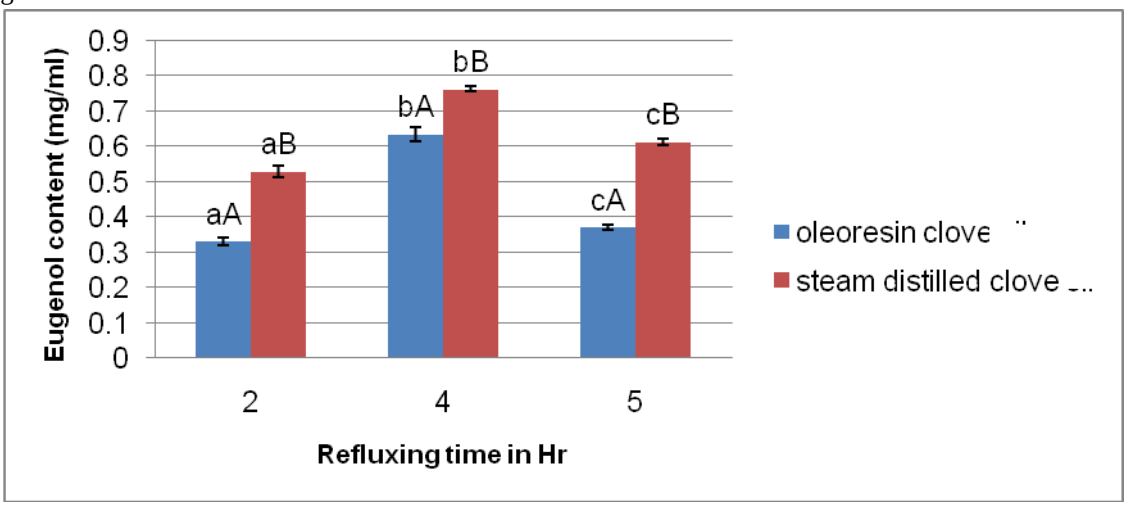


Fig.2. Eugenol content obtained at different time interval of refluxing (mg/ml) of clove extracts (steam distilled and oleoresin)

The mean changes between the samples were analyzed by one-way ANOVA. a-b Means with different lowercase superscripts letters are significantly different ($P < 0.05$) from each other. A-B Means with different uppercase superscripts letters are significantly different ($P < 0.05$) from each other. Error bars show the variations of three determinations in terms of standard error of mean

IV. CONCLUSION

Steam distilled clove extract has a much higher phenolic concentration than clove oleoresin. There was also a favourable association between total phenolic content, antioxidant potential, and free radical-scavenging activity for clove oleoresin and steam distilled clove extract. The measurement of eugenol content in steam distilled clove extract was found to be substantially greater than in oleoresin analogues.

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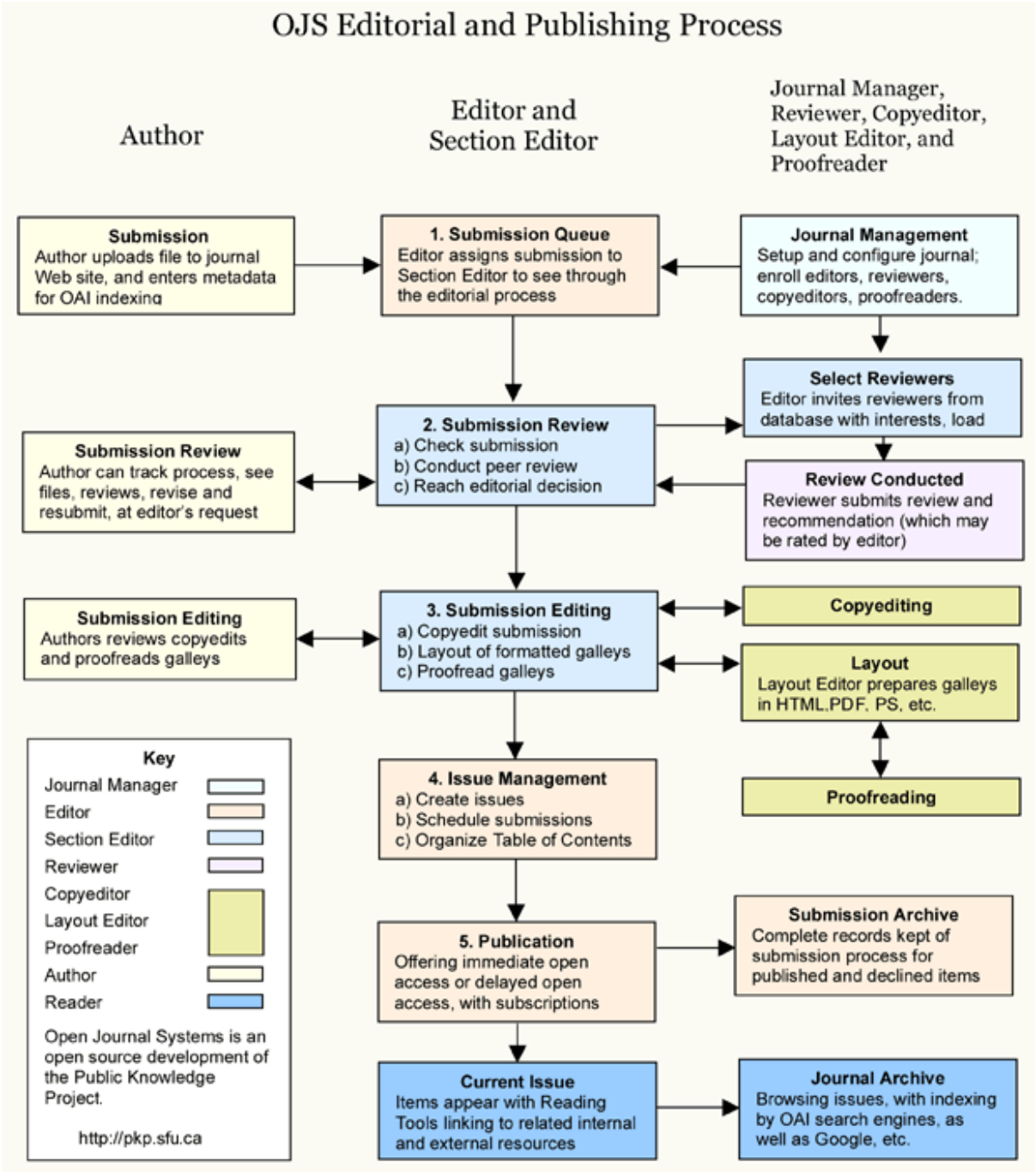

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