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Impact of Elevated CO_2 and Temperature on Growth, Physiology and Yield of Black Gram (*Vigna mungo* L. Hepper) Genotypes

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***Abstract*— *Climate change significantly impacts crop production and productivity, particularly in leguminous crops like black gram, which are primarily cultivated under rainfed conditions. The present study evaluated the response of four black gram genotypes (Vigna mungo L. Hepper) to elevated CO_2 (eCO_2) and elevated temperature (eT) using the Carbon dioxide and Temperature Gradient Chamber (CTGC) facility at ICAR-CRIDA. The experimental conditions included ambient temperature (aT), eCO_2 (550 ± 50 ppm), and three gradients of elevated temperature (eT1 = aT+1.5°C, eT2 = aT+3.0°C, eT3 = aT+4.5°C) individually and in combination with eCO_2. Results revealed that eCO_2 significantly improved biomass accumulation, photosynthetic rate, and yield traits of all the black gram genotypes, while temperature at higher gradients negatively impacted plant growth and yield. The combined treatment of eCO_2 + eT had a mitigating effect, particularly at eT1+eCO_2, where plants exhibited improved photosynthetic rate, water-use efficiency, and biomass accumulation. However, this amelioration effect declined at eT2+eCO_2 and became negligible at eT3+eCO_2. The negative effects of elevated temperature counteracted the advantage of elevated CO_2. However, at eT3, the negative effects of temperature stress outweighed the benefits of eCO_2, leading to reduced yield. Among the genotypes, PLU-826 exhibited the highest photosynthetic rate (Anet) under eCO_2, while PSRJ-95016 showed improved performance under eT1+eCO_2. Yield parameters such as pod number and seed weight significantly declined under eT3, highlighting the importance of selecting climate-resilient genotypes to sustain black gram productivity under changing environmental conditions.*

Keywords*— Black gram (Vigna mungo L. Hepper), climate change, elevated CO_2, genotypes, temperature, yield.

I. INTRODUCTION

Climate change, characterized by rising atmospheric CO_2 levels and increasing global temperatures, presents a substantial threat to agricultural productivity, especially for rainfed crops like black gram (*Vigna mungo* L. Hepper). As a C_3 legume, black gram is generally responsive to elevated CO_2 (eCO_2), with reported improvements in photosynthetic capacity, biomass accumulation, and water-use efficiency [1-2]. However, these benefits are often constrained by the concurrent rise in temperature, which can accelerate phenological development, reduce grain filling, and increase oxidative stress and respiration losses [3-4].

While the individual effects of eCO_2 and elevated temperature (eT) have been well characterized in cereal crops like rice and wheat [5], the combined impact of these climate factors on leguminous crops remains underexplored. Recent studies suggest that legume responses to eCO_2 and eT are highly genotype-specific, necessitating the identification of cultivars with resilience to climate extremes [6-8]. Black gram, in particular, lacks robust experimental data under controlled conditions simulating future climate scenarios.

Given that atmospheric CO_2 concentrations are expected to reach ~550 ppm by 2050 [9], it becomes critical to understand the interactive effects of eCO_2 and eT on the physiological and yield performance of black gram genotypes. Moreover, studies in crops like soybean and mungbean have demonstrated significant genotype-by-environment interactions under eCO_2 and heat stress [10-11], highlighting the need to identify stable and high-performing black gram genotypes under these evolving conditions.

The present study was aimed to assess the growth, morpho-physiological, yield and yield components response of four black gram genotypes under eCO_2, eT, individually and in combination in CTGC facility. These findings contribute in identifying better performing genotype/s of this important rainfed short duration leguminous crop in predicted future climatic conditions.

II. METHODOLOGY

2.1 Plant material and experimental conditions

Four black gram genotypes- IPU-06-02, PLU-826, PSRJ-95016 and IPU-94-1 were received from ICAR-IIPR, Kanpur and which are popular released varieties in India. The growth and yield responses of black gram genotypes were assessed at elevated CO_2 (eCO_2) of 550ppm and at three gradients of elevated temperature both individually and in combination with eCO_2 in Carbon dioxide and Temperature Gradient Chamber (CTGC) facility during summer season at Central Research Institute for Dryland Agriculture (ICAR-CRIDA, 17°27'N latitude, 78°35'E longitude, and approximately 515 meters above sea level) Hyderabad, Telangana, India.

The CTGC facility consists of eight chambers with 30 meters length, 6 meters width and 4 meters height at the centre [12]. These 8 chambers categorised as- two chambers were maintained at ambient temperature (aT); two chambers are with elevated CO_2 ($eCO_2 - 550 \pm 50$ppm); two chambers are with elevated temperature (eT) with three gradients (eT1- aT+1.5°C; eT2- aT+3.0°C; eT3- aT+4.5°C); two chambers are with elevated temperature at three gradients over aT with the combination of eCO_2 (eT1+eCO_2; eT2+eCO_2; eT3+eCO_2). This facility is equipped with advanced SCADA (Supervisory Control and Data Acquisition) software linked with PLC, which facilitates monitoring and controlling set environmental parameters like temperature, humidity and CO_2.

The land within the chambers were ploughed thoroughly and black gram genotypes were sown with 30 X 10 cm spacing. The recommended doses of fertilizers (N @ 20 kg ha^{-1} and P @ 40 kg ha^{-1} and K @ 20 kg ha^{-1}) were applied and maintained uniform irrigation at regular intervals along with standard plant protection measures to control pests and diseases throughout the study period.

2.2 Weather conditions

The weather parameters during crop growth period were presented in Fig 1. The maximum air temperature during crop growth period ranged from 30°C to 39.8°C with an average of 36.46°C, while minimum temperature ranged from 23.2°C to 29.2°C with an average value of 26°C.

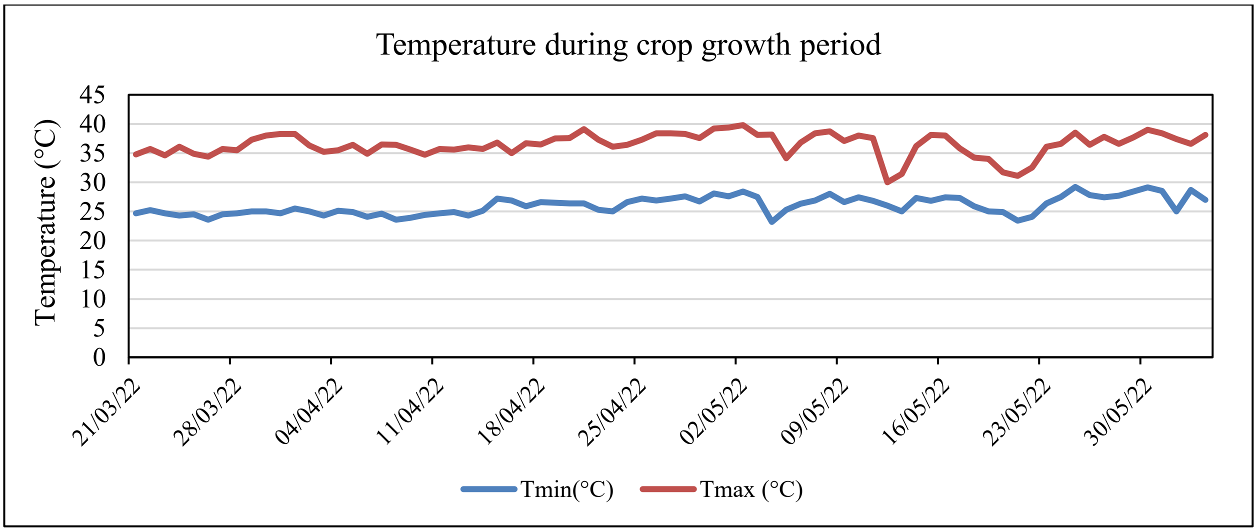

Fig.1. Minimum and maximum temperatures during crop growth period

The data on morphological, physiological, biomass and yield traits of all the genotypes under different treatments were recorded in three replications.

2.3 Physiological parameters

Physiological observations were recorded in all the treatments during pre-flowering stage on the fully expanded leaf of each genotype in three replicates. The net photosynthetic rate (A_{net}), stomatal conductance (g_s) and transpiration rate (Tr) were recorded between 10:00 and 12:00 h using portable photosynthesis system (LI-6400, LI-COR, Nebraska, USA) with irradiance set at 1200 μmol $m^{-2}s^{-1}$ under different treatments by setting the respective CO_2 level and temperature of CTGC. The water use efficiency (WUEi) was calculated as the ratio of Anet and Tr using the formula WUE= Anet/Tr.

2.4 Morphological parameters

Morphological observations were recorded at harvest by carefully uprooting the plants in three replications from each treatment for each genotype. Plants were divided into stems, leaves, roots and pods. The roots were cleaned gently with water. Root and shoot lengths were measured in cm with the measuring scale, and root volume was recorded as displacement of water and expressed in ml.

2.5 Biomass and yield parameter

The harvested plant parts were subjected to drying in a hot air oven set at 60°C and allowed to dry until the plant samples reached constant weight for determination of dry weights. The dry weights of the leaf, stem and root was measured using scientific weighing balance and expressed as gram per plant. The yield characteristics, including pod number/pl, pod weight (g/pl), and 100 seed weight (g), total biomass (g/pl), vegetative biomass (g/pl), and the HI (harvest index) were determined from the collected data sets. Harvest index was calculated as HI = (Seed yield/Total biomass) ×100 and expressed in percentage.

2.6 Statistical analysis

The analysis of variance (ANOVA) of replicated data was performed using STAR software ver. 0.1 to assess the significant differences among the genotypes, treatments, and their interaction.

III. RESULTS AND DISCUSSION

3.1 Physiological traits

The ANOVA results (Table 1) revealed that physiological parameters of Anet, gs, Tr and WUE were highly significant ($p< 0.01$) for Genotypes, Treatments and their interaction. The results demonstrated significant ($p< 0.01$) effects of elevated CO_2 (eCO_2) and elevated temperature (eT) on physiological parameters across black gram genotypes. The net photosynthetic rate (Anet) increased under eCO_2, with overall mean enhancement of 43.44% which ranged from 40.17% (IPU-94-1) to 50.11% (PSRJ-95016) as compared with ambient conditions, indicating improved carbon assimilation. These findings align with recent studies in black gram showing a 22.3% increase in photosynthesis and a 41.3% increase in yield under elevated CO_2 combined with high day temperature (+3 °C), indicating improved CO_2 assimilation and reproductive performance [4] (Guna et al., 2023).

The Anet of all black gram genotypes was maintained up to eT2 even when the temperatures are above optimum for the crop selected, however at eT3 it was impacted negatively, which supports the work of [13] Hatfield et al. (2011), this drop-in photosynthesis at eT3 is consistent with findings in kidney bean, where elevated temperature beyond 34 °C reduced reproductive success despite enhanced CO_2 [14]. Among the four genotypes, PSRJ-95016 recorded negative response for Anet at all three elevated temperature gradients (eT1 to eT3) over aT with highest reduction at eT3 (**-7.71%**).

However, these adverse effects of eT were alleviated in presence of eCO_2 (eT+ eCO_2) across all genotypes with an average increase over aT ranged from 21.72% (eT3+ eCO_2) to 37.46% (eT1+ eCO_2). This reduction in temperature-induced stress due to CO_2 enrichment is consistent with findings by [2] Leakey et al. (2009), who observed similar alleviatory effects of elevated CO_2 in various crops. Although overall increased Anet was recorded in individual eCO_2 compared to in combination with eT, PLU-826 exhibited genotypic variability by showing better performance than eCO_2 (42.23 μmol CO_2 m^{-2} s^{-1}) at eT1+ eCO_2 (45.40 μmol CO_2 m^{-2} s^{-1}) and eT2+ eCO_2 (42.93 μmol CO_2 m^{-2} s^{-1}).

Stomatal conductance (gs) exhibited mixed response among treatments and genotypes. In contrast to Anet, higher gs was recorded with eT1 (35.05%) than eCO_2 (17.57%) but genotypic difference observed with PSRJ-95016 with higher gs value recorded at eCO_2 (28.31%). Even though all the genotypes showed low gs at eT2 and eT3 compared to eT1, but IPU-06-02 showed difference response to elevated temperature gradient with increasing gs values at eT3(42.41%) and eT2(45.56%) respectively. The highest gs was observed in PLU-826 at eT1 (2.55 cm s^{-1}), which represented a 56.44% increase over aT. However, extreme temperature stress (eT3) reduced gs across all genotypes, with PLU-826 experiencing the most pronounced decline (-16.97%). This reduction was compensated in presence of eCO_2 (eT2+eCO_2, eT2+eCO_2)

except in PSRJ-95016. These findings align with reports of [15] Sage & Kubien, (2007) indicating that moderate temperature elevations enhance gs, but prolonged exposure reduces stomatal function.

Transpiration rate (Tr) also exhibited a mixed response to treatments, with eCO_2 generally leading to reductions in with eT. The Tr was increased as the temperature gradient was increased with the highest Tr recorded in IPU-94-1 at eT3 (16.77 mmol H_2O m^{-2} s^{-1}), reflecting a 16.17% increase over aT. In contrast, PSRJ-95016 recorded the largest decline in Tr at eT3 (-12.72%), suggesting a strong stomatal limitation under heat stress. These results support findings from pprevious studies on groundnut [16], which reported reduced Tr under elevated CO_2. This is likely due to the stomatal closure mechanisms induced by CO_2 enrichment, which reduces water loss but also limits CO_2 uptake.

The response of Water-use efficiency (WUE) clearly indicating eT impacted more of Anet than Tr, leading to lower values of WUE in eT conditions. Interestingly, an average increase in WUE (47.87%) was recorded under eT1+eCO_2, compared to eCO_2 (44.60%) over aT across all genotypes. The exception was IPU-06-02, which exhibited a significantly higher increase in WUE under eCO_2 (68.17%) than eT1+eCO_2 (46.80%). Similarly, PSRJ-95016 showed a 49.72% increase in WUE under eCO_2, but only 43.04% with eT1+eCO_2. The alleviation effect of eCO_2 on temperature-induced stress was evident in WUE trends, as genotypes with better water-use strategies maintained photosynthetic efficiency even under heat stress. The water-saving effect under eCO_2 supports the CO_2 fertilization theory, where reduced stomatal aperture conserves transpiration without limiting photosynthesis [1].

The interactive effects of eCO_2 and eT suggest that elevated temperature gradients can be beneficial when coupled with CO_2 enrichment, but higher temperature stress offsets these advantages.

3.2 Morphological characteristics

ANOVA revealed that all the morphological parameters such as plant height, stem girth, branch number, root length, root volume, leaf number was highly significant ($P<0.01$) for genotypes (except for plant height), treatments, and interaction of genotypes × treatments (except with branch number) (Table 1).

The elevated temperature negatively impacted morphological traits, including plant height, stem girth, leaf number, and branch number, with noticeable variations among black gram genotypes except root parameters (Table 3). Whereas elevated CO_2, both individually and in combination with elevated temperature significantly and positively influenced these traits. The highest mean performance of plant height was recorded under eCO_2+eT1 over aT while lowest was under eT3 (Table 3). Among the genotypes, PSRJ-95016 recorded maximum reduction (-28.21%) of plant height under eT3.

Table 1. ANOVA of all parameters of black gram genotypes at ambient, elevated CO_2, three gradients of elevated temperature and its combination with elevated CO_2 conditions

ANOVA	A_{net}	gs	Tr	WUE	PH	LN	BN	SG	RL	RV	LDW	SDW	RDW	PWT	TBM	PN	SN	SWT	VBM	HI (%)
G	37.7	0.3	15.3**	0.3**	3.12	364.1**	12.5**	0.9**	7.2**	1.4	60.7**	191	2.1	83.5	892.0**	2568	39803**	74.4**	1014	123
T	353.2	0.3	2.0**	2.2**	2360	626.3**	9.1**	1.4**	21.4	1.2	36.3	96.0	0.4	231.8**	868.5**	2794	27506**	196.7**	307 **	150
G x T	10.1	0.1	2.6**	0.1**	81.2	46.0**	0.3	0.2**	3.8**	0.5	10.8**	27.3	0.4	9.9	81.4	28.0	1403.6**	9.1	80.8	17.6

Error	3.34	0.00	0.6	0.03	11.9	6.2	0.2	0.0	1.2	0.0	1.5	2.9	0.0	3.1	8.2	12.4	85.9	3.0	5.6	6.9
CV (%)	5.1	2.8	5.3	7.4	3.9	7.3	8.1	4.0	7.3	6.5	10.9	10.5	9.7	6.9	5.3	4.3	2.1	81.0	10.3	9.7

(*Significant at 0.05%, **Significant at 0.01%); G-genotypes; T-treatments; GxT- genotypes x treatments; CV- coefficient of variation; A_{net}- photosynthetic rate; gs- stomatal conductance; Tr- transpiration rate; WUE- water use efficiency; PH- plant height; LN- leaf number; BN- branch number; SG- stem girth, RL- root length; RV- root volume; LDW-leaf dry weight; SDW-stem dry weight; RDW- root dry weight; PWT-Pod weight; TBM- total biomass; PN-pod number; SN- seed number; SWT-seed weight; VBM- vegetative biomass; HI- harvest index

Table 2. Physiological parameters of black gram genotypes at ambient, elevated CO_2, three gradients of elevated temperature and its combination with elevated CO_2 conditions

Photosynthetic rate (µmol CO_2/m^2/s)								
	aT	**eCO_2**	**eT1**	**eT2**	**eT3**	**eT1 +eCO_2**	**eT2 +eCO_2**	**eT3 +eCO_2**
IPU-06-02	31.50	44.50	31.13	32.10	31.47	42.47	39.87	37.83
PLU-826	29.70	42.23	35.07	30.53	31.43	45.40	42.93	38.23
PSRJ-95016	30.27	45.43	28.87	29.30	27.93	39.40	36.47	35.90
IPU-94-1	31.37	43.97	33.13	36.20	31.17	41.40	38.63	37.47
Mean	**30.71**	**44.03**	**32.05**	**32.03**	**30.50**	**42.17**	**39.48**	**37.36**
Stomatal conductance (cm/s)								
IPU-06-02	1.16	1.39	1.61	1.69	1.66	1.60	1.79	1.30
PLU-826	1.63	1.95	2.55	1.48	1.35	1.95	1.64	1.96
PSRJ-95016	1.48	1.90	1.76	1.55	1.82	1.86	1.79	1.55
IPU-94-1	1.49	1.54	1.89	1.73	1.70	1.80	1.84	1.73
Mean	**1.44**	**1.70**	**1.95**	**1.61**	**1.63**	**1.81**	**1.77**	**1.64**
Transpiration rate (mmol/m^2/s)								
IPU-06-02	14.13	11.83	13.70	14.10	15.10	12.93	14.00	14.93
PLU-826	15.10	16.37	15.63	15.37	14.83	14.43	16.30	15.33
PSRJ-95016	14.93	15.07	13.93	13.80	13.03	13.60	13.70	13.30
IPU-94-1	14.43	15.70	15.30	15.90	16.77	13.47	14.17	14.70
Mean	**14.65**	**14.74**	**14.64**	**14.79**	**14.93**	**13.61**	**14.54**	**14.57**
Water Use Efficiency (µmol CO_2/mmol H_2O)								
IPU-06-02	2.24	3.76	2.27	2.28	2.08	3.28	2.85	2.53
PLU-826	1.97	2.58	2.25	1.99	2.12	3.15	2.64	2.50
PSRJ-95016	2.03	3.03	2.07	2.12	2.15	2.90	2.67	2.71
IPU-94-1	2.17	2.81	2.18	2.29	1.86	3.07	2.73	2.56
Mean	**2.10**	**3.05**	**2.19**	**2.17**	**2.05**	**3.10**	**2.72**	**2.58**

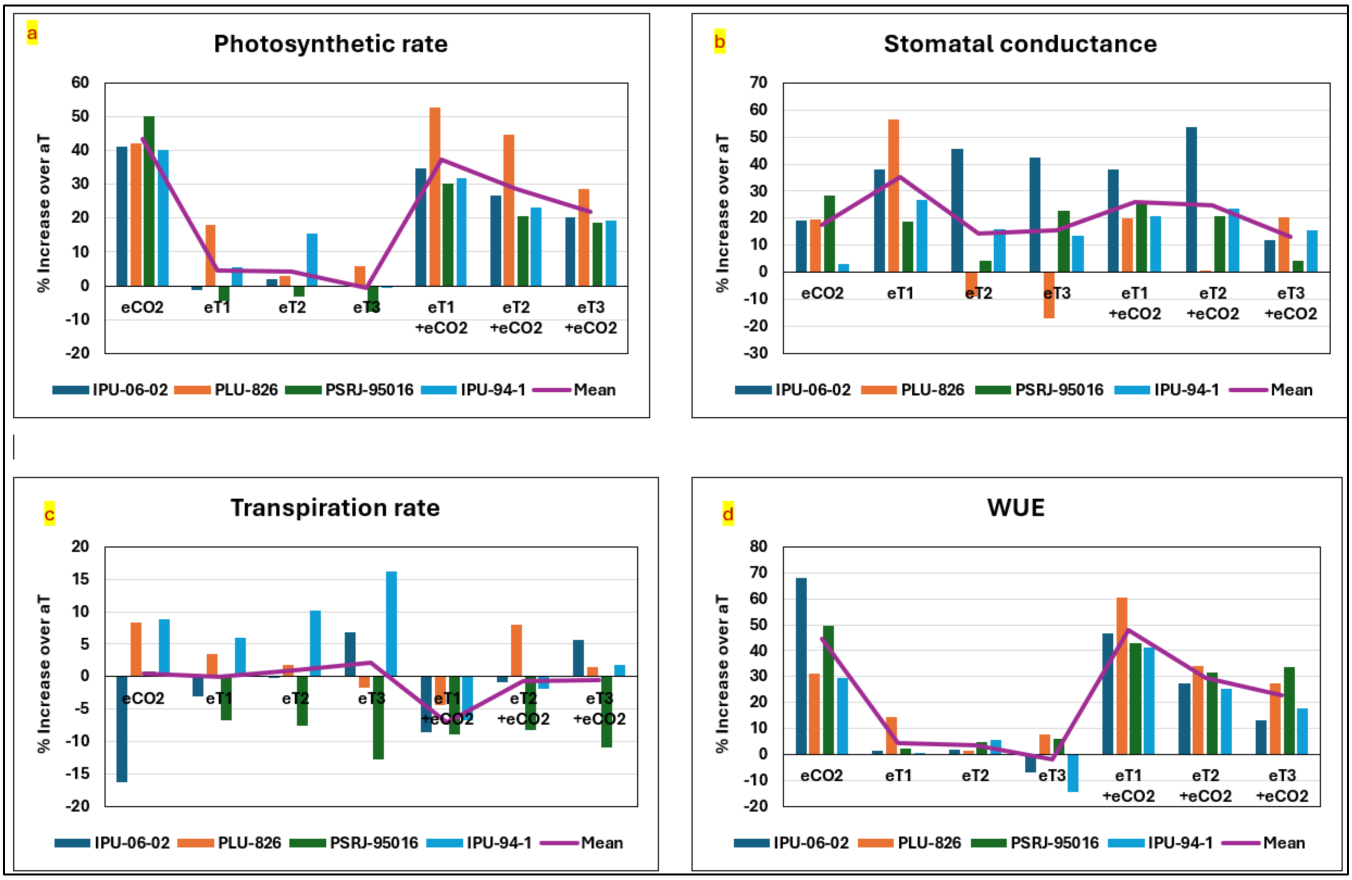

Fig.2. Physiological traits response of black gram at individual and in combination of elevated CO_2 and temperature over ambient condition

According to [17] Vanaja et al. (2024), elevated CO_2 enhanced plant height by increasing biomass allocation towards stem and petiole development, contributing to structural growth with improved carbon assimilation. Similar improvements in plant height and leaf number under eCO_2 have been observed in SPAR chamber experiments with black gram, showing 29-45% early vegetative growth increases [4].

Stem girth followed a similar trend, the stem girth decreased as the temperature gradient increases, which was ameliorated in the presence eCO_2, with a significant increase over aT. The highest stem girth was recorded under eCO_2+eT1 and lowest under eT3. There is a significance genotypic variation was observed among genotypes with highest amelioration of eCO_2 was observed with IPU-94-1 (eCO_2+eT1) and lowest with PSRJ-95016 (eCO_2+eT3).

A significant decrease in leaf number was observed across all black gram genotypes under elevated temperature, with the overall reduction ranging from eT1 (-2.84%) to eT3 (-25.58) (Table 3). However, this reduction was mitigated in the presence of eCO_2 (eCO_2+eT), particularly under eCO_2+eT1 conditions, where the leaf number surpassed that under individual eCO_2. While highest leaf number was recorded with PLU-826 under eCO_2 alone, the greatest improvement (over aT) under combined treatment (eCO_2+eT1) was observed/noted in IPU-06-02, highlighting the genotypic variation in response to these conditions.

Branch number followed a similar trend, showing a negative impact under elevated temperature at eT2 and eT3 across all genotypes, except for PSRJ-95016, which was adversely affected at all three temp gradients/levels. The decline in branch number, however, was lessened under eCO_2 across genotypes. The most notable improvement was observed in IPU-94-1 under eCO_2+eT1, with a 37.50% increase over aT. In contrast, PSRJ-95016 exhibited the least recovery, recording a -10.53% decrease under eCO_2+eT3.

Table 3. Mean performance of morphological traits of four black gram genotypes at aT, eCO_2, eT1, eT2, eT3, eT1 +eCO_2, eT2 +eCO_2 and eT3 +eCO_2

Genotype								
Plant height (cm)	**aT**	**eCO_2**	**eT1**	**eT2**	**eT3**	**eT1 +eCO_2**	**eT2 +eCO_2**	**eT3 +eCO_2**
IPU-06-02	81.00	90.33	81.67	78.00	66.00	111.00	102.67	97.33
PLU-826	83.00	107.00	80.67	77.67	66.33	100.33	98.00	90.00
PSRJ-95016	91.00	107.67	84.67	71.33	65.33	109.00	100.33	80.00
IPU-94-1	82.67	103.67	76.67	74.67	70.00	106.33	101.33	93.33
Mean	**84.42**	**102.17**	**80.92**	**75.42**	**66.92**	**106.67**	**100.58**	**90.17**
Stem girth (mm)								
IPU-06-02	6.03	6.53	5.97	5.67	5.47	6.53	6.17	6.07
PLU-826	5.60	6.60	6.33	6.50	5.77	6.93	6.67	6.57
PSRJ-95016	5.50	6.27	5.73	5.63	6.03	6.53	6.27	5.57
IPU-94-1	5.67	6.17	5.50	6.10	5.60	6.50	6.37	6.27
Mean	**5.70**	**6.39**	**5.88**	**5.98**	**5.72**	**6.63**	**6.37**	**6.12**
Leaf number/plant								
IPU-06-02	27.33	33.00	25.33	18.00	16.67	48.67	35.67	26.67
PLU-826	34.00	50.00	32.67	29.67	24.00	48.33	45.33	35.67
PSRJ-95016	34.00	41.33	31.00	29.00	24.33	37.00	35.67	30.67
IPU-94-1	30.67	39.33	33.33	33.33	29.00	44.00	43.33	39.33
Mean	**31.50**	**40.92**	**30.58**	**27.50**	**23.50**	**44.50**	**40.00**	**33.08**
Branch number								
IPU-06-02	6.67	8.33	6.67	6.33	5.67	8.67	7.67	7.33
PLU-826	5.33	6.67	5.33	5.00	4.33	7.00	5.33	5.00
PSRJ-95016	6.33	7.00	5.33	5.33	4.67	6.67	6.67	5.67
IPU-94-1	5.33	7.00	5.33	5.00	5.00	7.33	6.33	5.67
Mean	**5.92**	**7.25**	**5.67**	**5.42**	**4.92**	**7.42**	**6.50**	**5.92**
Root length (cm)								
IPU-06-02	13.67	16.33	14.00	14.67	15.67	13.67	15.00	15.13
PLU-826	13.67	16.67	15.05	15.33	15.83	16.00	13.33	13.33
PSRJ-95016	12.67	16.00	13.50	18.67	18.67	16.00	15.33	15.33
IPU-94-1	13.33	16.67	13.33	17.00	16.73	13.67	12.33	13.00
Mean	**13.33**	**16.42**	**13.97**	**16.42**	**16.73**	**14.83**	**14.00**	**14.20**
Root volume (ml)								
IPU-06-02	2.67	3.63	3.02	3.42	3.17	3.00	2.67	2.70
PLU-826	2.77	3.70	2.90	3.10	3.27	4.00	4.00	4.00
PSRJ-95016	2.30	3.67	2.50	3.00	3.40	3.27	2.80	2.53
IPU-94-1	2.63	3.00	3.27	3.00	3.13	3.97	3.93	3.50
Mean	**2.59**	**3.50**	**2.92**	**3.13**	**3.24**	**3.56**	**3.35**	**3.18**

3.3 Biomass and yield traits

ANOVA revealed highly significant ($P<0.01$) variability in genotypes, treatments, and their interaction for biomass and yield traits (Table 1). Elevated CO_2 (eCO_2) positively influenced biomass accumulation, while elevated temperature (eT) had a negative impact. The degree of response varied among genotypes for root and stem characteristics.

Shoot dry weight (SDW) and leaf dry weight (LDW) showed notable improvements under eCO_2+eT1 compared to individual eCO_2 treatments, relative to ambient conditions (aT). Root dry weight (RDW) was slightly higher under individual eCO_2. Vegetative biomass (VBM) was highest under eCO_2+eT1, while the percentage increase in reproductive biomass (pod weight and seed weight) was greater under individual eCO_2 across most genotypes.

This differential response in biomass allocation suggests that eCO_2 may enhance carbon partitioning towards reproductive structures, potentially improving yield potential. Similar trends were reported in black gram where eCO_2 combined with moderate heat stress increased photosynthesis, pod number (28%), and grain yield by ~41% [4]. Broader evidence also supports this trend. A meta-analysis showed that elevated CO_2 increased reproductive allocation in crops [18]. Similarly, in quinoa, seed dry mass increased by 12–44% while total biomass rose only ~10%, indicating preferential allocation to reproduction [19]. These findings reinforce our observation that eCO_2 can enhance reproductive investment, improving yield potential in black gram. Genotypic variation was observed, with IPU-94-1 showing a significant reduction in VBM under eCO_2, contrary to other genotypes. However, reproductive biomass increased under eCO_2 in IPU-94-1, indicating a possible shift in resource allocation favoring yield components. This variation in response highlights the importance of genotype selection for adapting to future climate scenarios.

Table 4. Mean performance of biomass of four black gram genotypes at aT, eCO_2, eT1, eT2, eT3, eT1 +eCO_2, eT2 +eCO_2 and eT3 +eCO_2

Stem Dry Weight (g/pl)								
	aT	**eCO_2**	**eT1**	**eT2**	**eT3**	**eT1 + eCO_2**	**eT2 + eCO_2**	**eT3 + eCO_2**
IPU-06-02	13.16	15.67	16.42	13.15	10.26	17.63	16.05	15.92
PLU-826	13.77	24.84	19.35	15.97	12.23	26.06	22.03	20.47
PSRJ-95016	10.65	15.84	11.84	13.68	9.06	19.12	13.80	12.15
IPU-94-1	18.52	15.25	14.75	19.21	21.20	26.50	15.98	13.22
Mean	**14.02**	**17.90**	**15.59**	**15.50**	**13.19**	**22.32**	**16.96**	**15.44**
Leaf Dry Weight (g/pl)								
IPU-06-02	8.45	14.01	11.40	10.83	8.20	11.63	11.39	11.25
PLU-826	9.56	14.14	11.72	13.19	8.08	14.19	12.43	10.28
PSRJ-95016	7.79	11.85	9.53	8.80	6.29	12.57	7.74	8.95
IPU-94-1	15.14	12.59	11.50	14.09	14.01	18.36	10.04	8.18
Mean	**10.23**	**13.15**	**11.04**	**11.73**	**9.15**	**14.19**	**10.40**	**9.67**
Root Dry Weight (g/pl)								
IPU-06-02	0.80	0.73	0.57	1.25	0.90	0.87	0.63	0.52
PLU-826	1.42	1.81	1.44	0.80	1.28	1.95	1.52	1.94
PSRJ-95016	0.84	1.85	1.65	1.44	1.55	1.38	0.85	1.02
IPU-94-1	1.56	1.33	1.35	1.24	1.89	1.42	1.12	0.91
Mean	**1.16**	**1.43**	**1.25**	**1.18**	**1.40**	**1.40**	**1.03**	**1.10**

Vegetative Biomass (g/pl)								
IPU-06-02	28.45	36.70	33.10	31.40	23.94	38.63	34.19	33.15
PLU-826	30.04	45.96	38.13	35.24	26.94	47.66	41.29	38.35
PSRJ-95016	24.71	35.83	28.69	29.31	22.48	39.38	28.43	28.45
IPU-94-1	40.71	35.99	34.12	41.08	42.52	52.98	33.71	28.75
Mean	**30.98**	**38.62**	**33.51**	**34.26**	**28.97**	**44.66**	**34.41**	**32.18**
Total Biomass (g/pl)								
IPU-06-02	46.09	64.07	50.97	47.40	36.37	62.12	54.07	49.69
PLU-826	48.72	71.12	55.68	50.13	42.27	75.49	62.61	59.36
PSRJ-95016	40.65	60.59	44.93	44.27	35.13	60.38	45.56	43.33
IPU-94-1	63.58	66.11	53.62	59.93	60.47	76.36	51.81	48.15
Mean	**49.76**	**65.47**	**51.30**	**50.43**	**43.56**	**68.59**	**53.51**	**50.13**

Yield parameters were significantly influenced by eCO_2 and temperature gradients, with notable variations across genotypes. A significant reduction in mean performance was observed under eT as temperature gradient increased. These parameters improved under eCO_2 alone and in combination (eCO_2+eT). Seed number and seed weight were notably higher under individual eCO_2, while husk weight was significantly higher in combination (eCO_2+eT), suggesting improved seed filling across black gram genotypes under individual eCO_2. Though not directly assessed in this study, elevated CO_2 combined with heat stress has been shown to reduce grain micronutrient concentrations—particularly iron and zinc—by approximately 9–18% in legumes [20]. These reductions are often associated with nutrient dilution effects under eCO_2. On average, pod number increased under eCO_2 (32.55%) and eCO_2+eT1 (31.13%) compared to aT. PSRJ-95016 showed the highest increase (33.58%), demonstrating a strong response to CO_2 enrichment. Pod weight was highest in PSRJ-95016 under eCO_2 (45.25% increase over aT), while IPU-06-02 recorded the steepest decline under eT3 (-28.24%), indicating its vulnerability to temperature stress.

Seed number and seed weight showed substantial increases under eCO_2, with PSRJ-95016 exhibiting the highest improvement among all genotypes over aT. However, PSRJ-95016 also showed reductions under all eT gradients, indicating sensitivity to temperature stress. Similar trends have been documented in mungbean and kidney bean, where extreme temperature nullified the benefits of elevated CO_2 by impairing reproductive development [10, 15]. IPU-06-02 recorded the highest significant reduction under eT3.

Harvest index (HI) and 100-seed weight were negatively impacted by eT but ameliorated by eCO_2. Interestingly, IPU-94-1 recorded the lowest VBM under eCO_2 but the highest HI, with the greatest amelioration under eCO_2+eT3. For 100-seed weight, PLU-826 showed the highest value under eCO_2+eT1, demonstrating the greatest amelioration among all genotypes.

These findings highlight the significant role of eCO_2 in improving yield traits, while eT leads to reductions. The results confirm that genotypic variation plays a crucial role in adapting to changing environmental conditions and emphasize the need for selecting stress-resilient cultivars to sustain productivity in future climates. The observed increases in biomass and yield components under eCO_2 align with previous studies on legumes and other C3 crops. The enhanced carbon assimilation and improved water-use efficiency under eCO_2 likely contribute to the observed improvements in growth and yield parameters. However, the negative impacts of elevated temperature, particularly at higher gradients, underscore the complex interactions between CO_2 and temperature in determining crop productivity.

The genotypic variations observed in this study emphasize the importance of breeding and selecting cultivars that can capitalize on the benefits of eCO_2 while maintaining resilience to temperature stress. These patterns align with recent reviews highlighting that crop genotypes with greater reproductive allocation and efficient resource use perform better under elevated CO_2 and warming scenarios [21]. Future research should focus on understanding the physiological and molecular mechanisms underlying these genotypic differences to inform breeding strategies for climate-resilient black gram varieties.

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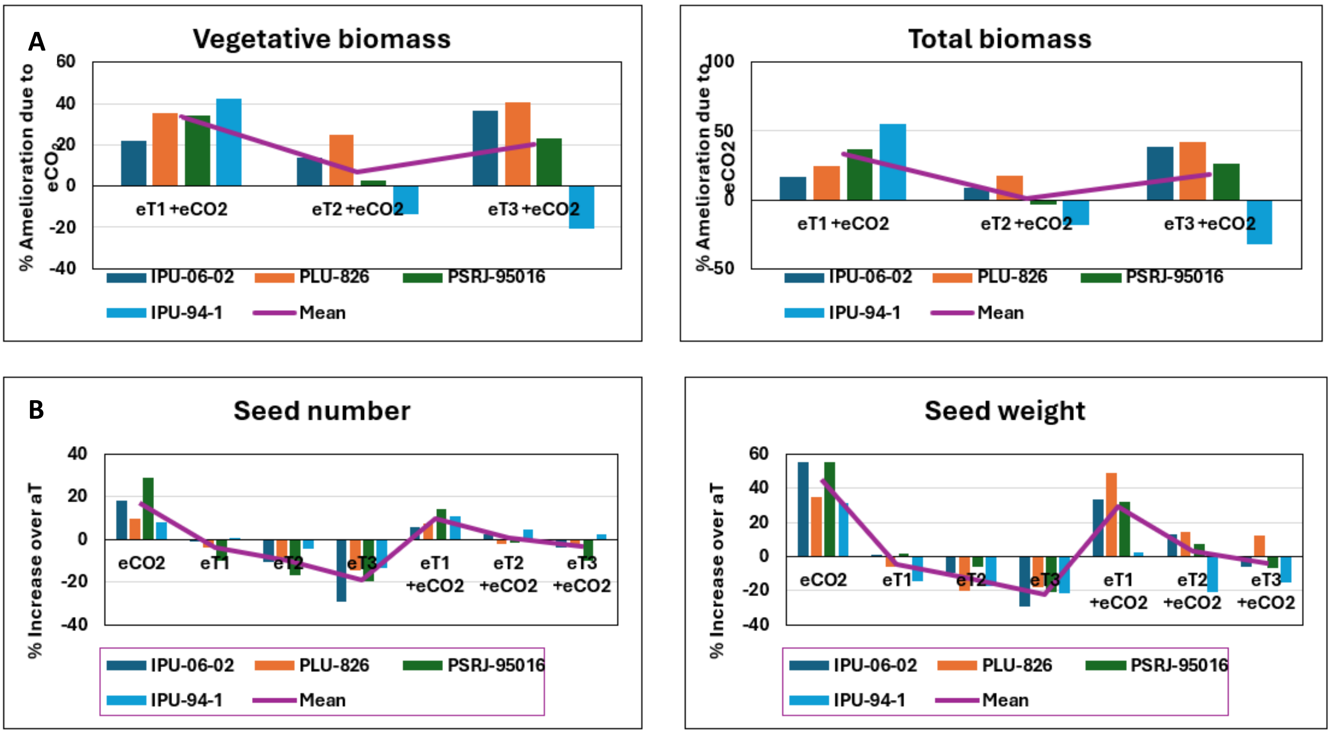

Fig.3. A. Amelioration of high temperature impact by eCO_2 on biomass; B. Impact of eCO_2 and elevated temperature in over yield traits of four black gram genotypes

Table 5. Mean performance of yield traits of four black gram genotypes at aT, eCO_2, eT1, eT2, eT3, eT1 +eCO_2, eT2 +eCO_2 and eT3 +eCO_2

Cluster no./pl								
	aT	**eCO_2**	**eT1**	**eT2**	**eT3**	**eT1 +eCO_2**	**eT2+eCO_2**	**eT3 +eCO_2**
IPU-06-02	18.00	24.67	20.00	16.67	14.67	31.00	29.33	23.67
PLU-826	29.33	41.67	28.00	21.67	17.33	50.67	38.00	30.33
PSRJ-95016	26.00	42.00	23.67	20.33	18.33	36.67	30.67	30.00
IPU-94-1	25.67	31.00	21.33	20.33	18.67	36.33	30.33	22.00
Mean	**24.75**	**34.83**	**23.25**	**19.75**	**17.25**	**38.67**	**32.08**	**26.50**
Pod number/pl								
IPU-06-02	74.33	96.67	71.33	65.67	53.33	95.33	88.00	72.67
PLU-826	85.00	107.33	78.00	77.33	63.67	106.67	96.00	80.00
PSRJ-95016	65.33	91.67	64.00	62.67	51.00	93.00	75.00	60.33
IPU-94-1	90.33	120.67	81.33	79.67	78.33	116.00	97.67	86.67
Mean	**78.75**	**104.08**	**73.67**	**71.33**	**61.58**	**102.75**	**89.17**	**74.92**
Pod wt. (g/pl)								
IPU-06-02	23.69	33.67	22.58	22.17	17.00	32.00	26.00	22.00
PLU-826	23.98	30.33	23.17	20.18	20.67	33.29	26.63	26.68
PSRJ-95016	21.38	31.05	21.91	20.35	18.23	27.32	23.16	21.20

IPU-94-1	28.36	36.93	26.03	25.39	23.36	30.08	24.67	25.84
Mean	**24.35**	**32.99**	**23.42**	**22.02**	**19.82**	**30.67**	**25.12**	**23.93**
Husk wt. (g/pl)								
IPU-06-02	6.05	6.30	4.71	6.17	4.57	8.51	6.12	5.46
PLU-826	5.29	5.16	5.62	5.28	5.35	5.45	5.31	5.66
PSRJ-95016	5.44	6.29	5.67	5.39	5.58	6.31	6.03	6.33
IPU-94-1	5.49	6.81	6.52	6.54	5.41	6.70	6.58	6.44
Mean	**5.57**	**6.14**	**5.63**	**5.85**	**5.23**	**6.74**	**6.01**	**5.97**
Seed no.								
IPU-06-02	427.33	504.33	422.33	381.33	303.00	453.00	437.33	411.00
PLU-826	476.67	522.67	459.33	436.00	408.00	513.00	466.00	464.00
PSRJ-95016	408.00	526.33	367.67	340.33	327.67	465.67	400.67	368.00
IPU-94-1	481.00	521.00	482.33	461.33	416.33	532.33	503.33	493.67
Mean	**448.25**	**518.58**	**432.92**	**404.75**	**363.75**	**491.00**	**451.83**	**434.17**
Seed wt. (g/pl)								
IPU-06-02	17.64	27.37	17.87	16.00	12.43	23.49	19.88	16.54
PLU-826	18.69	25.16	17.55	14.90	15.33	27.84	21.32	21.01
PSRJ-95016	15.94	24.76	16.23	14.96	12.65	21.00	17.13	14.88
IPU-94-1	22.87	30.12	19.51	18.85	17.95	23.38	18.10	19.40
Mean	**18.79**	**26.85**	**17.79**	**16.18**	**14.59**	**23.93**	**19.11**	**17.96**
HI (%)								
IPU-06-02	38.24	42.70	35.05	33.79	34.14	37.79	36.75	33.35
PLU-826	38.35	35.42	31.51	29.79	36.23	36.87	34.00	35.44
PSRJ-95016	39.19	40.80	36.03	33.74	36.00	34.80	37.63	34.29
IPU-94-1	36.09	45.48	36.41	31.48	29.75	30.62	34.94	40.30
Mean	**37.97**	**41.10**	**34.75**	**32.20**	**34.03**	**35.02**	**35.83**	**35.85**
100 Seed wt. (g)								
IPU-06-02	4.13	5.43	4.23	4.20	4.10	5.19	4.54	4.03
PLU-826	3.92	4.81	3.82	3.42	3.75	5.42	4.58	4.53
PSRJ-95016	3.90	4.70	4.42	4.39	3.86	4.51	4.28	4.04
IPU-94-1	4.75	5.78	4.05	4.09	4.31	4.39	3.60	3.93
Mean	**4.17**	**5.18**	**4.13**	**4.03**	**4.01**	**4.88**	**4.25**	**4.13**

*HI = Harvest Index.

IV. CONCLUSION

Elevated CO_2 (eCO_2) significantly influenced black gram genotypes, enhancing photosynthetic efficiency, WUE, and biomass accumulation. PLU-826 and IPU-06-02 exhibited greater adaptability under eCO_2+eT1, with higher photosynthetic rate, WUE, and pod number, whereas PSRJ-95016 and IPU-94-1 were more sensitive to eT3, with reduced biomass and yield. Morphological traits, including plant height and leaf number, improved under eCO_2, particularly in IPU-06-02 and PLU-826. Biomass accumulation increased significantly under eCO_2, with PSRJ-95016 and IPU-06-02 exhibiting enhanced carbon assimilation. Yield traits, including pod number, seed weight, and harvest index (HI), responded positively to eCO_2, with IPU-06-02 showing the highest seed weight under eCO_2 and IPU-94-1 experiencing the greatest reduction under eT3. The interaction of eCO_2 and moderate temperature elevation (eT1) improved yield, while extreme heat stress (eT3) reduced these benefits.

These results emphasize the role of eCO_2 in alleviating temperature stress, improving productivity, and highlighting genotypic differences. Selecting climate-resilient black gram cultivars is crucial for future adaptation.

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