International Journal of Environment, Agriculture and Biotechnology
Vol-10, Issue-2; Mar-Apr, 2025
Peer-Reviewed International Journal
Journal Home Page Available: https://ijeab.com/
Journal DOI: 10.22161/ijeab

Evaluation of the potential of medicinal compounds of 10 Vietnamese rice varieties

Nguyen Thi Lang, Bui Chi Hieu, Bien Anh Khoa, Nguyen Trong Phuoc, Bui Chi Buu

High Agricultural Technology Research Institute for Mekong Delta
Web: hatri.org

Received: 03 Feb 2025; Received in revised form: 04 Mar 2025; Accepted: 11 Mar 2025; Available online: 17 Mar 2025

Abstract— *This study to quantify flavonoids and polyphenols between 10 different rice varieties from white and brown rice. The phenolic content (TPC) of 10 rice varieties in the Mekong Delta :TPC of brown rice and white rice of different rice varieties differed significantly ($p < 0.05$). The TPC measured in brown rice was significantly higher (118.98-206.06%) than in white rice. Brown rice TPC levels were highest in NepThan (771.12 mg/100 g), while the lowest levels were found in OM5451 (100.12GAE mg/100 g). The highest and lowest levels of GAE were found in HATRI 11 (215.06 mg/100 g) and OM5451 (133.08 mg/100 g). The difference in the total phenolic content between varieties can be attributed to differences in genotype. It is noteworthy that HATRI 11's TPC is the highest among white rice, but it is only 119.47% higher than the lowest white rice varieties. The flavonoid content (TFC) of 10 TFC rice varieties of brown rice and white rice of different rice varieties differed significantly. The TFC of brown rice is in the range of 142.26–919.1RE mg/100 g, while white rice is in the range of 68.72–645.29RE mg/100 g: brown rice has a total flavonoid content 10%-20% higher than white rice. The total anthocyanin content in free form and the bonds vary between different genotypes of pigmented rice bran. The content of free and binding anthocyanins in the rice fraction of ten different genotypes of pigmented rice ranged from 2.18 to 256.11 and 5.25 to 38.51 mg of Cy3-GE/100 g DM, respectively. The highest concentration of anthocyanins was detected in free form. Nep Than showed the highest anthocyanin content (234.62 mg Cy3-GE/100 g DM), followed by HATRI 11 (73.88 mg Cy3-GE/100 g DM) and white rice (50.42 mg Cy3-GE/100 g DM). Polyphenols and flavonoids and the mechanism of rice growth and development from the limited description of previous works. Our studies have enriched the active compounds of rice and laid a solid foundation to improve the active compounds for the type of rice served as functional foods for Vietnam.*

Keywords— ***anthocyanin content, brown rice, flavonoid content, Polyphenols, white rice***

I. INTRODUCTION

Rice is a staple food for more than half of the global population. The majority of rice consumers are observed to suffer from problems such as malnutrition, Fe and Zn deficiency, and health problems related to oxidative stress such as stroke, psoriasis, type II diabetes, heart disease, obesity, cancer, dermatitis, and rheumatoid arthritis (Shridhar, and ctv.,2015). Antioxidants protect cells against free radicals, which can cause disease in humans. Since much of the global population depends on rice, grain enrichment with Fe, Zn, and antioxidant compounds are priority areas of rice research (Kuma et al.,2020; Zhu et al.,2018). Consuming antioxidant-rich rice is a better and cheaper option to combat stress-related disorders and gain other health benefits (Zhu et al.,2018). Enhancing the nutritional value of the antioxidant compounds in rice, is the best and cheapest way to achieve health.

There is growing interest in identifying new natural sources of potential compounds in food and medicinal chemistry. Rice contains special bioactive compounds, such as ferulic acid and hydroxycinnamic acid, which have attracted increasing attention from scientists and consumers alike (Alvese et al.,2016). These phenolic compounds have antioxidant and anti-inflammatory properties. They are

correlated with a reduced risk of various chronic diseases, including heart disease, and a reduction in type 2 diabetes symptoms (Liu et al.,2015). These highly bioactive compounds are distributed mainly in the bran layer (Verardo et al.,2016). Therefore, the uneven distribution of bioactive compounds can affect functional characteristics in rice (Shen et al.,2009).

Polyphenols are are good in phytochemicals , which have wide uses. Phenolic compounds are common secondary metabolites in rice plant growth, and they are useful in pollination, seed diffusion, and disease and pest prevention (Cheynier, 2012). Plant phenols include monophenols, diphenyl phenols and polyphenols. Plant polyphenols are beneficial to human health, accounting for a relatively high percentage in phenolic substances. Plant polyphenols are phenylpropanoid derivatives, including flavonoids, phenolic acids, stilbenes and curcumin (Quideau et al., 2011). These compounds display many biological activities such as antioxidant, antibacterial, anti-inflammatory, anti-tumor and antiviral effects (Maleki et al., 2019), possessing great application potential in the medicine, foods, cosmetics and chemicals (Yahfoufi et al., 2018; Fraga et al., 2019).

Binding phenolic compounds, such as ferulic, coumaric and caffeic acids, can be hydrolyzed in the large intestine by intestinal enzymes, freeing them from binding macromolecules (Pang et al .,2018; Ge et al.,2021). Phenolic compounds are synthesized in plant cells, and are often referred to as functional components as hydrogen atoms on aromatic rings with hydroxyl (Alu'datt et al., 2017). Their antioxidant capacity is important in minimizing the negative effects of oxidative stress, which has been linked to the pathogenesis of many diseases (Ma et al., 2019). These substances can generally be divided into two main groups, flavonoids (flavanols, flavonols, anthocyanins) and non-flavonoids (phenolic acids, stilbenes, tannins and their derivatives) (Zhang and Tsao, 2016; Alu'datt et al., 2017). On this point, the nutritional and bioactive values of phenolic compounds have been confirmed from several crops, medicinal plants and rice plants (Neri-Numa et al., 2020; de Araújo et al, 2021).

Antioxidants are present in plants both in the form of enzymes and non-enzymes. Enzymatic antioxidants are catalase, peroxidase, superoxide dismutase, glutathione and other proteins and non-enzymatic antioxidants including phenolic protective compounds (vitamin E, flavonoids, phenolic acids and others); Nitrogenous compounds (alkaloids, amino acids and amines), carotenoids and chlorophyll derivatives (Govindaraj et al.,2017). Enzymatic antioxidants protect plant cells from damage caused by reactive oxygen species and act as a defense system to maintain cellular structural and functional integrity by inhibiting oxidative degradation to macromolecules such as lipids, proteins and nucleic acids (Rossatto et al.,2017). Therefore, improving these characteristics in rice will lead to the development of better quality rice. Non-enzymatic antioxidants such as phenolic acids, flavonoids, anthocyanins and proanthocyanidins, tocopherols and tocotrienols (vitamin E), andγ-oryzanol have been reported...

The antioxidant activity of rice plants is promoted by various phytochemicals in experiments (Gong,et al.,2017). Polyphenols are the main antioxidants in rice, while other bioactive compounds, such as phytosterols, also have antioxidant properties (Ragaee et la.,2013). These bioactive antioxidants act as a preventive and protective mechanism against chronic diseases caused by oxidative damage caused by excessive free radical production in living organisms [Podio et al.,2017]. There are many methods for measuring the antioxidant activity of these substances in vitro, such as DPPH, ABTS, PSC, and ORAC, based on different antioxidant mechanisms (Desta et al.,2022).

Understanding the genetic basis of these complex antioxidant traits and identifying key QTLs is essential to improving these phytochemicals through molecular breeding to improve the growing nutritional issues of rice-fed populations and seed quality. The identification of QTLs/genes for higher carotenoid content and the development of functional markers is slow in rice because reports of carotenoids are not available in rice (Zhai et al.,2016). Widespread genetic variation for carotenoid content exists in rice. White rice accumulates very small amounts of carotenoids (Ashraf et al.,2017).

The pigments that provide color, anthocyanidins and proanthocyanidins, are present in the pericardium and aleurone of rice grains. Eleven QTLs such as qTAC1.1 and qTAC5.1 controlling anthocyanin content, qSOD1.1, qSOD5.1 and qSOD10.1 for superoxide dismutase (SOD), qTFC6.1, qTFC11.1 and qTFC12.1 for total flavonoid content (TFC), qOZ8.1 and qOZ11.1 for γ-oryzanol (OZ) and qAC11.1 for ABTS activity were discovered as novel locus. The chromosome position on 11 at 45.3 cM modulates GO, TFC, and anthocyanin content (TAC), and on chromosome 12 at 101.8 cM controls TAC and ABTS activity, respectively, were found to be antioxidant hotspots. (Bastia et al.,2022)

This study to quantify flavonoids and polyphenols between 10 different rice varieties from white and brown rice, describes gene expression profiles associated with biosynthesis. Polyphenols and flavonoids and the mechanism of rice growth and development from the limited description of previous works. Our studies have

enriched the active compounds of rice and laid a solid foundation to improve the active compounds for the type of rice served as functional foods for Vietnam.

II. METHODS AND MATERIALS

Ten rice varieties grown at HATRI Mekong Delta High-tech Agricultural Research Institute (table 1). After harvesting, seeds are dried to 13 ± 1% moisture at temperatures below 40°C. All rice samples are peeled and polished by rice peeling machine and rice milling machine, set to 8% milling level, to obtain ground rice bran. To separate the grains from the rice bran, they are sieved through a sieve of 180 μm (80 mesh).

Table 1 . 10 lines varities at High Agricultural Technology Research Institute for Mekong delta(HATRI)

Lines	Crossing	Traits	Bwown rice	White rice
HATRI11	jinmibyeo/Dular//5*jinmibyeo	Red color , Tolerance for drought	84,71	6,28
HATRI 2 (TPG1)	Jinmibyeo/SP6	Tolerance for drought	82,45	76,57
HATRI 200	Kuming/SP6	Aroma , Japonica , salinity tolerance	85,42	76,98
HATRI 10	OM7347/KhaoDawmali 105	Aroma , drought tolerance	83,63	75,47
OM5451		Popular at MEKong	81,15	74,62
HATRI722	Jasmine 85/	Aroma	84,42	73,65
OM4900	C53/Jasmine 85	Submergence, salinity and drought	86,42	74,68
HUYẾT RỒNG	landrace	Can Tho Landace	88,96	7,52
NẾP THAN	Landrace	An Giang Landrace	88,56	0
IR64	IRRI	Good genes	82,42	76,25

Extraction of free phenolics and favonoids

Extraction of free phenolics and favonoids 10 rice (0.5 g) were treated with 50 mL of acidifed methanol solution (95% methanol: 1 M HCl 85:15, v/v). Te mixture was homogenised using homogenizer for 5 min in an ice bath. Solutions were centrifuged at 2500g for 10 min and supernatants were removed. Te fltered supernatants were concentrated by evaporation at 45 °C using hot plate. Te concentrated fltrate was then diluted with 10 mL of acidifed methanol and stored until analysis. Extraction of bound phenolics and favonoids Te residue obtained from the free phenolics extraction was hydrolyzed with NaOH (40 mL, 2 M) at room temperature for 1 h with continuous shaking. Hexanes (10 mL) were used to extract lipids. Te hydrolysate was then neutralised with 10 mL of 2 M HCL. Solution was transferred to separation funnel and was then extracted fve times with ethyl acetate. Te ethyl acetate layer (supernatants) were pooled and evaporated using hot plate (at 45 °C). Residue was dissolved in distilled water (10 mL) and then stored until analysis.

Extraction of bound phenolics and favonoids

The residue obtained from the free phenolics extraction was hydrolyzed with NaOH (40 mL, 2 M) at room temperature for 1 h with continuous shaking. Hexanes (10 mL) were used to extract lipids. The hydrolysate was then neutralised with 10 mL of 2 M HCL. Solution was transferred to separation funnel and was then extracted fve times with ethyl acetate. The ethyl acetate layer (supernatants) were pooled and evaporated using hot plate (at 45 °C). Residue was dissolved in distilled water (10 mL) and then stored until analysis.

Determination of Total Phenolic

Total favonoid content Extracts (1 mL) were mixed with NaNO2 solution (4 mL, 1:5, w/v) and incubated at room temperature for 6 min. 0.3 mL of AlCl3 solution (1:10, w/v) was added, the reagents were mixed well, and the reaction was allowed to stand for another 6 min. Immediately after that, 1M NaOH solution (2.0 mL) was added to each extract and incubated for 10 min at room temperature. Te absorbance of the solutions was read at 510 nm using a spectrophotometer (UV2550, Shimadzu, Japan). Diferent concentrations of quercetin standard were used to prepare a calibration curve. Results were expressed as milligram quercetin equivalents (QE)/100 gDM . (Ghasemzadehet al.,2015).

Determination of Flavonoid Content

Currently, the determination of TFC was depended on the aluminium chloride colorimetric method described by(Qiu, et la 2010). Briefly, a 50 µL supernatant was mixed with 100 µL distilled water. Then, 5% $NaNO_2$ was added into the mixture and incubated for 5 min. Subsequently, 10% $AlCl_3$ $6H_2O$ solution was drawn and added to the mixture for incubation for 3 min. Finally, 60 µL 4%NaOH was added to the termination reaction. The samples were read at 510 nm. Absolute methanol was used as the control, while a standard rutin curve was used to calculate the content of TFC. Results were recorded as mg of RE/100 g DW.

***Determination of DPPH* (1,1-Diphenyl-2-picrylhydrazyl) assay** Radical Scavenging Activity

The method by (Ghasemzadeh và ctv.,2015), was used with slight modifications to assess DPPH. The mixtures were shaken vigorously, and the sample was taken then incubated for 30 min in the dark. Mixture was measured at 517 nm.

DPPH radical scavenging effect (%)=1−Asample−AbackgroundAcontrol×100%DPPH radical scavenging effect (%)=1−Asample−AbackgroundAcontrol×100%

(3)

where A_{sample}, $A_{control}$, and $A_{background}$ refer to sample (sample and DPPH), control (without sample), and background (without sample), respectively.

RNA Isolation and Sequencing

Total RNA was isolated from the grain rice using "NucleoSpin® RNA Plant" kit (Macherey-Nagel, Germany) following user's manual. RNA quality and quantity was determined using Nanodrop 1000 spectrophotometer (Thermo Fisher Scientific Inc, Wilmington, DE, USA) and Bioanalyzer RNA Nano chip (Agilent Technologies, Santa Clara, CA, USA). The RNA samples with 260/280 ratio of 1.8 to 2.1, 260/230 ratio of 2.0 to 2.3 and RNA integrity number (RIN) more than 7.0, were used for mRNA sequencing. The cDNA library was prepared using mRNA-Seq Sample Prep kit (Illumina Inc., San Diego, CA, USA) following manufacturer's instructions. Poly (A)-containing mRNA was isolated using magnetic beads with oligo (dT) and fragmented into short pieces. These short fragments were used as templates to synthesize first-strand cDNA using reverse transcriptase and random hexamer-primers. The second-strand cDNA was then synthesized using DNA polymerase, dNTPs and RNase H. After completing purification and end repair process, the cDNA fragments were ligated to sequencing adapters. The fragments were then purified and amplified by PCR to obtain the final library followed by purification. Paired-end sequencing was carried out on Illumina HiSeq 2500 platform and raw reads of 100nt were generated. Filtered reads were obtained after running the quality control (QC) using NGS-QC box (Kata et la .,2015)

Statistical Analyses

All measurements in this study were presented as means ± standard deviations. Each antioxidant activity assay was carried out three times from the same extracts in order to determine their reproducibility. Statistical differences and principal component analysis were analyzed with SPSS 25 (SPSS Inc., Chicago, IL, USA) (Li et al .2021). Canonical correspondence analysis and networks were conducted with Origin software.

III. RESULT AND DISCUSSION

3.1. The phenolic content (TPC) of 10 rice varieties in the Mekong Delta :TPC of brown rice and white rice of different rice varieties differed significantly ($p < 0.05$). The TPC measured in brown rice was significantly higher (118.98-206.06%) than in white rice. Brown rice TPC levels were highest in NepThan (771.12 mg/100 g), while the lowest levels were found in OM5451 (100.12GAE mg/100 g). The highest and lowest levels of GAE were found in HATRI 11 (215.06 mg/100 g) and OM5451 (133.08 mg/100 g). The difference in the total phenolic content between varieties can be attributed to differences in genotype. It is noteworthy that HATRI 11's TPC is the highest among white rice, but it is only 119.47% higher than the lowest white rice varieties.

This may indicate that milling during rice processing has different effects on the active compounds of different varieties. Meanwhile, the difference between the ratio of brown rice and white rice in different varieties can be attributed to inconsistent trends. The content of free phenolics and favonoids, binding and total phenolic acid content in brown and white rice portions of sixteen different genotypes of pigmented rice is shown in Table 1. The free phenolic content in brown rice portion varies from 153.30 to 771.15 mg GAE / 100 g DM. The binding phenolic content ranges from 102.05 to 443.55 mg GAE / 100 g. Total phenolic (4) Viability (%) = 100 – optical density of sample / optical density of control × 100 (5) Optical density of sample = absorption of cells treated with extraction – absorption of cells treated with an average DMSO content of 0.1% ranged from 269.85 to 1214.7 mg GAE/100 g DM. As shown in the Table 1

Brown rice contains the highest content of free phenolics, binding and total phenolics (771.15; 374.15 and 1,145.3 mg GAE/100 g DM), followed by white rice (521.36; 386.22 and 907.58 mg GAE/100 g DM, respectively) In one study by (Shen et al. 2009) the total free

favonoid content of white, red and black rice was compared and it was found that the average favonoid content in white rice was lower than in red and black rice. The current results suggest that phenolic and favonoid compounds in rice bran are mostly present in free form, and this is an important issue for future research. Forms of phenolics and favonoid bonds are covalently conjugated to the structure of cell walls via ester bonds (Ali et al.,2018).

In the colon, they are broken down by microfora and can release phenolics that are bound to carry out beneficial biological activities (Choi et al.,2010). The current results are consistent with previous reports, in which phenolics and favonoids in rice are mainly distributed in free form (Zhang et al.,2010, Ti et al.,2014).

Table 2. Identifed free, bound and total individual phenolics, favonoidsfrom 10 varities rice

line	Brown rice		Wilte rice		TPC	Brown rice		White rice		TFC
	TPC free (GAE mg/ 100g)	TPC bound (GAE mg/ 100g)	TPC free (GAE mg/ 100g)	TPC bound (GAE mg/ 100g)	Total brown rice (GAE mg/ 100g)	TFC free (mg QE/ 100g DM)	TFC bound (mgQE/ 100gDM)	TFC free (mg QE/ 100g DM)	TFC bound (mgQE/ 100 gDM)	Total TFC white rice(mg QE/ 100g DM)
HATRI 11	575,25b	348,12b	415,48b	222,78b	923,37b	491,56b	367,8a	260,48b	156,14b	859,3b6
HATRI 2 (TPG1)	268,74d	216,25c	125,17e	108,75c	485d	245,35d	166,15c	216,15b	107,14d	411,5d
HATRI 200	285,16d	124,27d	220,25d	112,42c	409,43d	245,85d	156,24c	201,44b	123,10c	402,0d9
HATRI 10	175,50e	132,23d	107,14e	85,52d	307,73e	188,16e	110,25c	98,57c	65,12	298,4e1
OM 5451	100,12f	85,74e	95,45f	56,15d	185,86g	106,74e	35,52d	155,47c	13,25f	142,2f6
HATRI 722	144,20e	118,25d	98,38f	55,47d	262,45f	135,15e	105,25c	85,41c	52,14e	204,4e
OM 4900	145,60e	132,23d	107,14e	85,52d	277,83f	133,25e	114,52	90,45c	59,78e	247,7e7
HUYẾT RỒNG	489,56c	274,33c	344,28c	256,14b	763,83c	342,51c	207,15b	215,12b	142,41b	549,6c6
NẾP THAN	771,15a	374,15a	521,36a	386,22a	1,145,3a	526,65a	392,45a	432,15a	213,14a	919,1a
IR 64	162,30e	122,45d	100,37e	92,56d	284,75f	128,17e	109,33c	90,53c	74,15e	237,5e

3.2. The flavonoid content (TFC) of 10 TFC rice varieties of brown rice and white rice of different rice varieties differed significantly (table 1; statistically significant < 0.05). The TFC of brown rice is in the range of 142.26–919.1RE mg/100 g, while white rice is in the range of 68.72–645.29RE mg/100 g: brown rice has a total flavonoid content 10%-20% higher than white rice. Among them, the difference between brown rice and white rice from HATRI 722 and OM5451 varieties is smaller. Different trends in TFC and TPC are variation in the ten varieties, possibly because the distribution positions of total phenols and flavonoids in rice are influenced by genotype. In 10 rice varieties, the free flavonoid content of brown rice and white rice differed significantly ($p < 0.05$). In brown rice, it is in the range of 146.98 – 193.65 RE mg/100 g, with OM5451 and Nep Than having higher contents, and the content of free flavonoids in white rice is between 55.47 – 432.15 RE mg/100 g respectively.

There were significant differences in binding flavonoid content in brown and white rice of 10 statistically significant rice varieties ($p < 0.05$). The flavonoid content of the free, binding HATRI 11 variety in brown rice and white rice is in the range of 491.56b –367.8RE mg/100 g and260.48 –156.14 RE mg/100 g, respectively. The binding flavonoid content in brown rice varies 0.59 – 1.50 times that of white rice for the same variety, while OM4900, IR64, HATRI722 and OM5451 all have lower binding flavonoid content in brown rice than in white rice. The binding content in brown rice is 0.48 - 1.28 times higher than in white rice. Notably, the binding flavonoid content in brown rice of OM 5451 is lower than the content in white rice

3.3. Anthocyanin content in 10 rice varieties :The content of free and associated anthocyanins in ten varieties with different genotypes of pigmented rice is presented in Table 2. The total anthocyanin content in free form and the bonds vary between different genotypes of pigmented rice bran. The content of free and binding

anthocyanins in the rice fraction of ten different genotypes of pigmented rice ranged from 2.18 to 256.11 and 5.25 to 38.51 mg of Cy3-GE/100 g DM, respectively. The highest concentration of anthocyanins was detected in free form. Nep Than showed the highest anthocyanin content (234.62 mg Cy3-GE/100 g DM), followed by HATRI 11 (73.88 mg Cy3-GE/100 g DM) and white rice (50.42 mg Cy3-GE/100 g DM).

Table 3. Identifed free, bound and total individual anthocyanin from 10 varities rice

lines	**Brown rice**		**TAC Total brown rice(**GAE mg /100 g)	**White rice**		**TAC (White rice)Total g (**GAE mg /100 g)
	TAC free(GAE mg /100g	TAC bound (GAE mg /100 g)		TAC free (GAE mg /100g)	TAC bound (GAE mg /100g)	
HATRI 11	68,33b	15,55b	73,88b	42,27b	8,15c	50,42b
HATRI 2 (TPG1)	38,77c	6,27e	45,04c	8,75h	3,14d	11,89d
HATRI 200	15,32e	6,18e	21,50d	10,48g	3,45d	13,93d
HATRI 10	15,8e	7,25d	23,05d	10,42g	0,55g	10,97d
OM5451	16,57	9,25c	25,82d	11,45g	0,25g	11,70d
HATRI722	20,15d	9,56c	29,71d	17,14e	2,85e	19,99d
OM4900	24,56d	10,12b	34,68c	18,25d	1,65f	19,90d
HUYẾT RỒNG	63,20b	16,47b	79,67b	20.34c	14,78b	36,12c
NẾP THAN	183,05a	24,16a	234,62a	58,56a	15,35a	73,34a
IR 64	20,69d	9,45c	30,14c	15,71f	0,8g	16,51d

3.4. The composition and content of phenolic acids vary significantly between varieties While the phenolic acid content in each brown rice is significantly higher than that of white rice ($p < 0.05$) (table 3). Glutinous brown rice contains relatively high levels of caffeic acid, sinapic acid (a hydroxycinnamic acid), ferulic acid and p-hydroxybenzoic acid, while the overall content of six phenolic acids of OM5451, HATRI10, IR64 is lower than that of other brown rice varieties. Meanwhile, HATRI 200 white rice has a relatively higher overall content of phenolic acids. The p-hydroxybenzoic acid content of brown rice in HATRI 200 is more than twice that of white rice. Phenolics and favonoid composition: Five phenolic compounds (protocatechuic acid, syringic acid, ferulic acid, cinnamic acid and p-coumaric acid) have been detected in white and brown rice of different varieties (Table 4). Protocatechuic acid, with content from 0.23 to 3.15 mg/100g DM on brown rice and highest content (in Glutinous Coal)

Ferulic acid is an important phenolic acid component in rice; The content of varied varieties was 18.85-29.71 mg/kg in brown rice and 9.54-18.66 mg/kg in white rice. The white rice content of these two acids is 2.0-2.4% and 1.4-2.2% lower than brown rice, respectively. Both syringic acid and p-coumaric acid show similar trends in different varieties, with little variation in p-coumaric acid content in different varieties. The cinnamic acid content in brown rice is in the range of 9.25-19.25 mg/kg, which is 1.25-2.0 times higher than the content of white rice. P-coumaric acid is lowest in brown rice with 10.47 mg/kg in OM5451 and with white rice as low as 8.27 mg/kg in IR64. Among the favonoid compounds identified in various rice varieties.

Favonoid compounds (quercetin, apigenin, catechins, luteolin and myrecitin) The content of quercetin in free and bound form ranged from 2.71 to 11.89 mg/100 g DM and from 0.16 to 3.66 mg/100 g DM, respectively. Apigenin in concentrations between 5.75 and 10.55 and 4.52 and 9.55 mg/100g DM, for brown rice and white rice respectively. Catechins also exist in brown rice and white rice with concentrations ranging from 10.11 to 20.14 and 7.25 and 18.23 mg/100 g DM, respectively (Table 5).

Table 4: Content of phenolic acids 10 varieties

lines	Brown rice	White rice	Brown rice	White rice	Brown rice	White rice	Brown rice	White rice	Brown rice	White rice
	protocatechuic acid (mg/ 100g DM	protocatechuic acid (mg/ 100g DM	axit syringic (mg/ 100g DM	axit syringic (mg/ 100g DM	axit ferulic (mg/ 100g DM	axit ferulic kết (mg/ 100g DM	cinnamic acid (mg/ 100g DM	cinnamic acid (mg/ 100g DM	p-coumaric acid (mg/100g DM)	p-coumaric acid (mg/ 100g DM)
HATRI 11	2,56c	1,95c	15,25a	6,45a	5,25c	3,85c	15,44b	0	24,15a	20,55a
HATRI 2(TPG1)	1,25d	0,88f	10,25b	3,15c	4,22d	2,56d	14.55b	12,33b	15,47c	14,25c
HATRI 200	2,15c	1,12d	10,56b	3,22c	3,25e	1,03f	11,25c	10,99c	14,23c	13,48d
HATRI10	1,08d	0,56g	9,35c	2,14d	3,10e	0,95g	10,58c	9,50d	13,25d	10,66e
OM5451	0,99e	0,23g	6,45e	1,25e	2,90f	0,35h	9,25d	5,11d	10,47e	8,62g
HATRI 722	1,44d	0,45g	7,56d	1,54e	2,65f	1,48f	9,50d	8,25d	11,52e	9,55f
OM4900	1,55d	0,62f	6,78e	1,89e	3,16e	1,75e	9,98d	8,75d	11,54e	8,73g
HUYẾT RỒNG	3,25b	2,66b	11,25b	4,15b	6,15b	5,99b	14,55b	12,78b	15,77c	13,28d
NẾP THAN	4,55a	3,15a	15,21a	6,24a	9,25a	8.10a	19,25a	14,15a	18,74b	17,25b
IR64	1,62d	0,55g	8,25c	1,99e	3,56e	1.02f	10,28c	8,28d	10,99f	8,27g

Table 5: Favonoid compounds (quercetin, apigenin, catechins, luteolin and myrecitin)in 10 varieties

lines	Brown rice	White rice	Brown rice	White rice	Brown rice	White rice	Brown rice	White rice
	quercetin (mg/100g DM)	quercetin (mg/100 gDM)	apigenin (mg/100 gDM)	apigenin (mg/100gDM)	catechin (mg/100 gDM)	catechin (mg/100 gDM)	luteolin (mg/100gDM)	luteolin (mg/100 gDM)
HATRI 11	6,18c	5,55c	8,14c	6,25c	15,66b	13,23b	7,72b	5,25c
HATRI 2 (TPG1)	4,15d	3,25d	7,88d	5,22d	10,12d	8,55d	6,14c	4,66d
HATRI 200	4,55d	3,47d	7,99d	6,10c	12,11c	10,22c	6,55c	4,29d
HATRI 10	3,66e	2,15e	6,22e	5,22d	11,25d	8,54d	5,57d	3,22e
OM5451	3,58e	1,22	5,75f	4,52e	10,11d	7,25e	5,01d	2,95f
HATRI 722	3,78e	2,55f	6,25e	4,58e	10,56d	8,54d	5,78d	4,15d
OM4900	3,47e	2,45f	6,78e	4,55e	10,98d	7,58e	5,67d	4,47d
HUYẾT RỒNG	10,52b	7,25b	9,25b	8,10b	15,90b	13,25b	7,98b	6,18b
NẾP THAN	12,55a	10,25a	10,55a	9,25a	20,14a	18,23a	9,21a	8,71a
IR64	3,45e	2,68f	6,45e	4,25e	10,99d	8,29d	5,66d	4,57d

Luteolin in brown rice, with concentrations ranging from 5.01 to 9.21 mg/100 g DM and 2.95 to 8.71 mg/100 g DM, respectively. Furthermore, catechins and myrecitin are the most abundant favonoid compounds in brown and red rice bran, while apigenin and quercetin are the most abundant favonoid compounds in black rice bran (Zhou et al.,2018) showing that brown rice contains high levels of ferulic acid and p-coumaric acid and gallic acid content, Low vanillic, caffeinic and syringic, consistent with current studies.

5/DPPH activity Free radicals are an intermediate metabolite of various biochemical reactions in human life activities. It has high chemical activity and is an effective

defense system of the human body. However, the excessive accumulation of free radicals that cannot be scavenged in time will attack life macromolecules and various organelles, and cause interhuman damage at the molecular, cellular and tissue level, which can further accelerate the human aging process and cause various chronic diseases (Akbari et al, 2022; Anand et al., 2022).

Various mechanisms, such as free radical scavenging, capacity reduction, metal ions, and lipid peroxidation inhibition, have been studied to explain how rice bran extract can be used as an antioxidant (Ghasemzadeh et al.,2015). DPPH radical scavenging tests are based on the transfer of electrons from the molecule of the donor radical to the corresponding radical. The DPPH method is the simplest method for measuring the ability of antioxidants to block free radicals. DPPH thoroughly scavenged the effects of all extracts in white rice and brown rice (free and bonded) increased with increasing concentration (Figure 2).

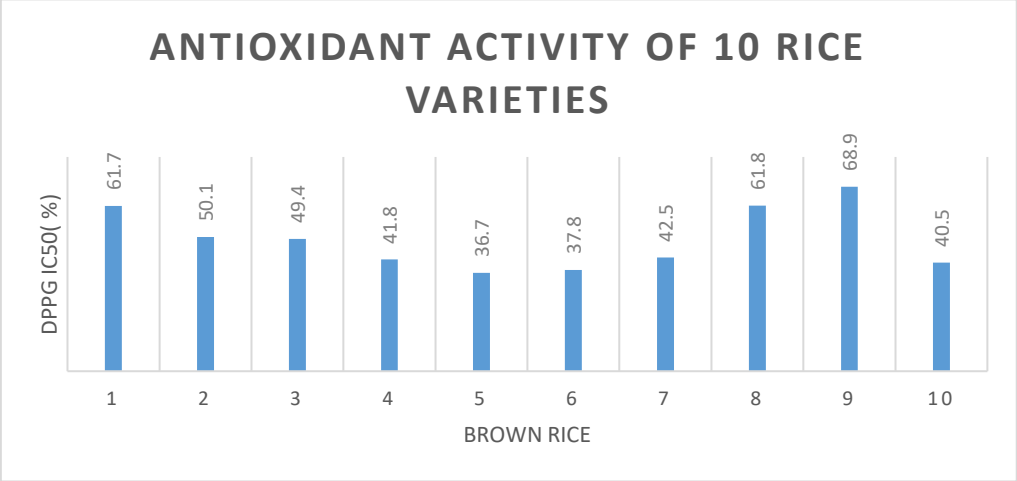
DPPH operations have been markedly affected. The rate of DPPH root scavenging varied significantly in brown rice (64.72–70.92%, $p < 0.05$). Brown rice of OM5451 has the lowest antioxidant capacity for removing DPPH radicals and is significantly different from other varieties ($p < 0.05$). The coefficient of variation between different breeds is 3.1%. Meanwhile, the scavenging rate of DPPH free radicals in white rice was on average 24.5% lower than brown rice at 37.8% (OM5451). Spirit and Dragon Blood had the highest scavenging rates (61.8,-68.9%), while HATRI 722 and OM 5451 had the lowest scavenging rates (36.7-37.8%). The DPPH antioxidant activities of different white rice varieties are significantly different ($p < 0.05$).

The 10-variety extract demonstrated the highest DPPH activity, followed by rice extraction. DPPH activity of different varieties ranges from 24.5 to 60.4%, for white rice and DPPH activity in free fractions and bonds respectively ranges from within brown rice 36.7 to 68.9% (figure 1a)

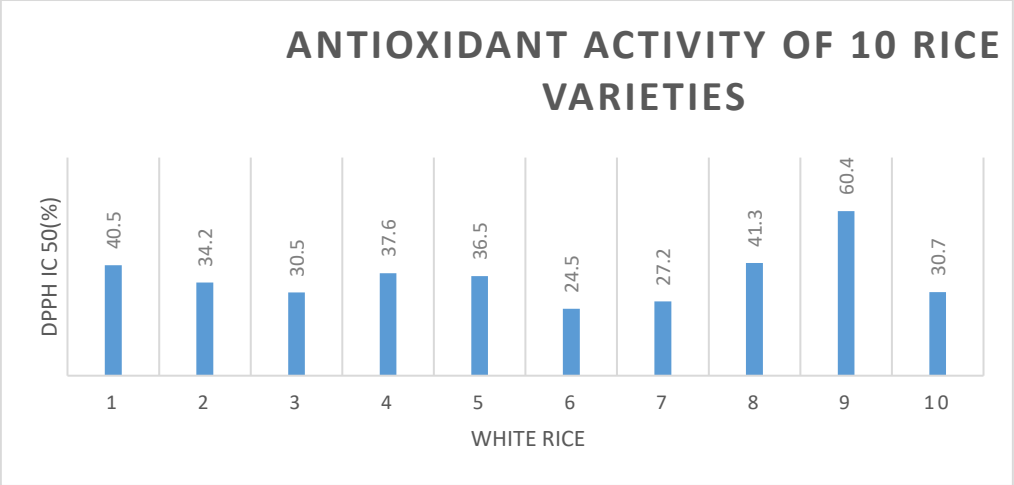
Figure 1: Antioxidant activity value of ten rice varieties (Figure 1A: Antioxidant activity of brown rice of 10 rice varieties. Figure 1B: Antioxidant activity of White rice of 10 rice varieties **3.6. RNA-Seq-based transcriptional analysis of rice sheds light on key factors related to the ability to carry phenolic genes, flavonoid content and anthocyanin content of rice varieties** In many rice varieties after genetic analysis there are many heterozygous factors and many hypotheses. Among them, the dominance hypothesis, the dominant hypothesis, and the epistasis hypothesis have been widely accepted and underlie heterozygous research (Shasidhar et al.,2020). The heterogeneity of plants is not fully and logically explained by any of these hypotheses or views, no matter how different they are. Thus, SSR molecular markers and quantitative trait locus mapping (QTL) are increasingly becoming a standard tool for testing the genetic basis in breeds due to Yu et la 2017

Gene total phenolics content(TPC) liên kết với RM24616 trên nhiễm sắc thể số 9 summarized QTL's effect on heterozygosity based on 35 studies and found that dominance and epistasis were equally proportional in these studies, suggesting that QTL mapping results varied between species and even within different groups of the same specimen. Therefore, SSR and QTL markings are not sufficient to comprehensively explain the heteromorphism. Genes associated with traits established with TPC on chromosome 9 are recorded on rice varieties Genes associated with traits established with TPC on chromosome 9 are recorded on rice varieties (Krishnendu et al.,2022) Total phenolics content (TPC) gene binds RM24616 on chromosome 9

A



B



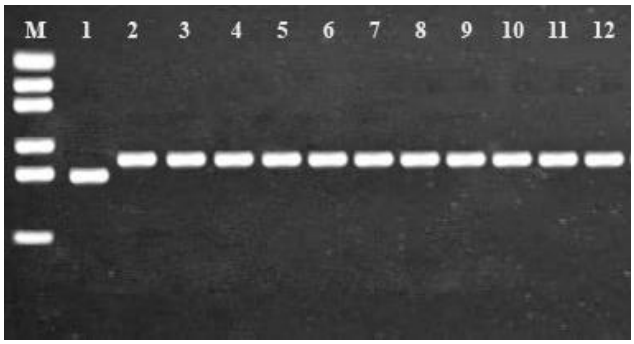


Fig.2: The PCR producted of RM24616 (chromsome 9)

Total flavonoid content (**TFC) with RM 17115 on chromosome 4**

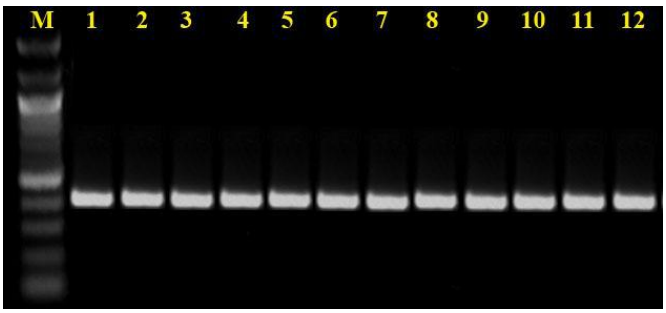


Fig.3: The PCR producted of RM 17115 on chromosome 4

Total anthocyanin conten(TAC) linked with RM285 omn chromosome 9 and RM28828 on chromosome 12 (figure 4)

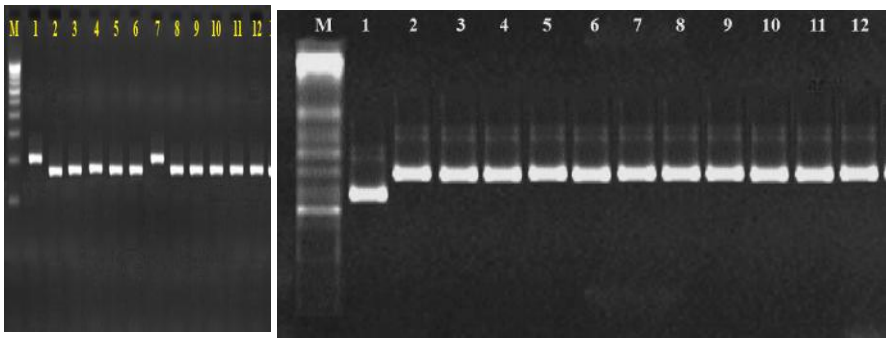


Fig.4 The PCR producted of RM 285 (chromsome 9) and RM28828 on Chromosome 12 on agarose gel.

Comprehensive RNA-Seq analysis of rice varieties and can identify many genes involved in biosynthesis The results of RNA-Seq analysis have been confirmed using qRT-PCR for several genes. Our results help explain the accumulation of secondary metabolites. The transcription data reported here will facilitate future studies of the molecular mechanisms of polysaccharide biosynthesis and provide new insights into rice plants Comparison of RNA-sequencing code sequences of 10 rice varieties: DNA sequences of 10 cladogram rice varieties (Figure 5), And they will be described the similarity between joinings, to show a large number of different nucleotides. The difference in the number of nucleotide sequences between varieties is shown in Figure 5

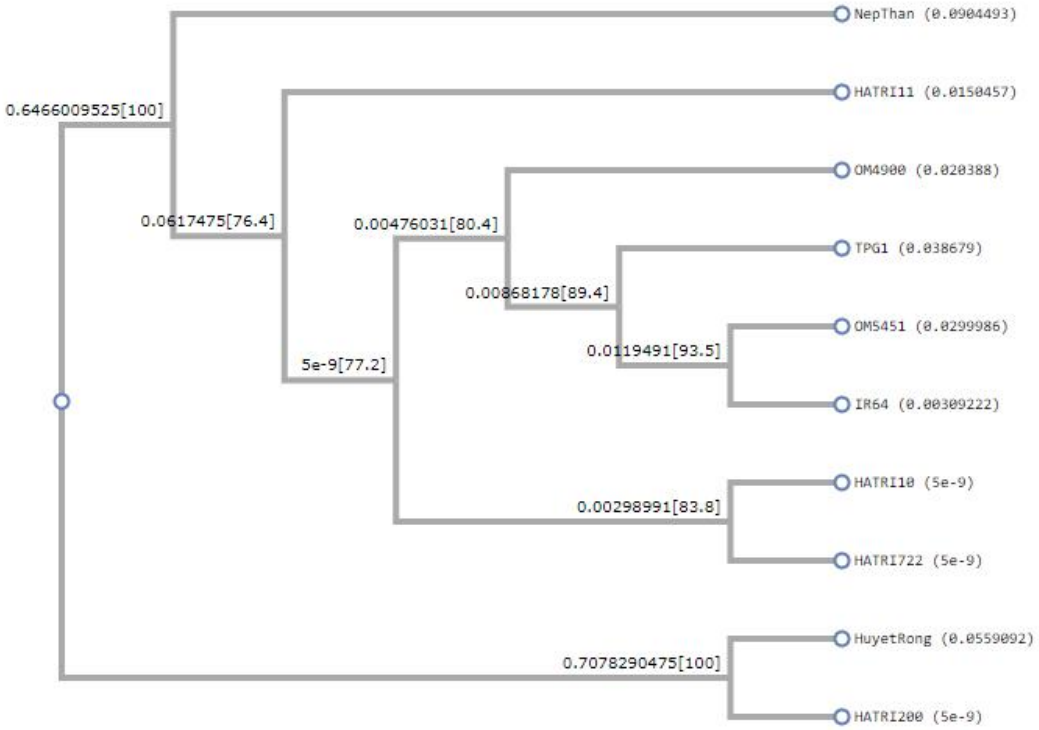


Fig.5: Phylogenetic plants based on TPC sequence using analysis of 10 different rice varieties

IV. CONCLUSIONS

Looking at a series of analytical results, the determination of polyphenols in rice extract aims to determine the quantitative profile of the quality of rice grains. Significant strides have been made in elucidating the chemical structure of these bioactive compounds but while mass spectrometry based techniques certainly represent a powerful tool for defining brown rice phenolic profiles, white rice. We strongly believe that the research efforts undertaken to date constitute an excellent starting point towards the development of analytical tools aimed at investigating the phenolic fraction of rice for demand in medicinal chemistry.

V. FUTURE PROSPECTS

The use of phenolic compounds and flavonoids is a potential candidate of bioactive agents in the field of pharmaceuticals and pharmaceuticals to enhance human health, prevent and cure various diseases. In order to explore and conduct alternatives using plant chemical compounds, medicinal plant surveys along with robust profile research need to be undertaken. The targeted compounds should be used in biomedical and pharmaceutical research ranging from in vitro, in vivo and step clinical trials to evaluating the safety, efficacy and also side effects of the candidate compounds tested.

1. Consequently, the flavonoid and phenolic compounds which are abundant found in a large number of rice and other plants may possible be an interesting choice of molecules for drug and medical product development.
2. In the same species of medicinal rice , the different cultivars may provide different amount of flavonoid and phenolic compounds as well as the biological activities. Thus, the cultivars of medicinal plant should be taken into account for

the future medical and pharmaceutical research studies.

3. The geographic areas of raw plant material should also be analyzed and compared in the future research. Since the environmental factors e.g., nutrients and mineral in soil are also effect on the quality and quantity of phytochemical compounds in some species of medicinal plant and rice.
4. The molecular mechanism and signaling pathway of many known flavonoid and phenolic compounds are need to be done in the future, so as to apply this knowledge to the drug development processes.
5. The need of purified compounds to confirm data obtained with the plant extracts.

REFERENCES

[1] Akbari, B., Baghaei-Yazdi, N., Bahmaie, M., Abhari, F. M. (2022). The role of plant-derived natural antioxidants in reduction of oxidative stress. *Biofactors* 48, 611–633.

[2] Ali Ghasemzadeh, Mohamad Taghi Karbalaii, Hawa Z. E. Jaafar & Asmah Rahmat 2018. Phytochemical constituents, antioxidant activity, and antiproliferative properties of black, red, and brown rice bran. *Chemistry Central Journal* volume 12, Article number: 17 (2018)

[3] Alu'datt, M. H., Rababah, T., Alhamad, M. N., Al-Mahasneh, M. A., Almajwal, A., Gammoh, S., et al. (2017). A review of phenolic compounds in oil-bearing plants: Distribution, identification and occurrence of phenolic compounds. *Food Chem.* 218, 99–106.

[4] Alves, G.H.; Ferreira, C.D.; Vivian, P.G.; Fernandes Monks, J.L.; Elias, M.C.; Vanier, N.L.; De Oliveira, M. The revisited levels of free and bound phenolics in rice: Effects of the extraction procedure. *Food Chem.* 2016, *208*, 116–123.

[5] Ashraf, H.; Murtaza, I.; Nazir, N.; Wani, A.B.; Naqash, S.; Husaini, A.M. Nutritional profiling of pigmented and scented rice genotypes of Kashmir Himalayas. J. Pharmacogn. Phytochem. 2017, 6, 910–916.

[6] Bastia, R.; Pandit, E.; Sanghamitra, P.; Barik, S.R.; Nayak, D.K.; Sahoo, A.; Moharana, A.; Meher, J.; Dash, P.K.; Raj, R.; et al. Association Mapping for Quantitative Trait Loci Controlling Superoxide Dismutase, Flavonoids, Anthocyanins, Carotenoids, γ-Oryzanol and Antioxidant Activity in Rice. Agronomy 2022, 12, 3036. https://doi.org/10.3390/ agronomy12123036

[7] Cheynier, V. (2012). Phenolic compounds: From plants to foods. *Phytochem. Rev.* 11, 153–177.

[8] Choi SP, Kim SP, Kang MY, Nam SH, Friedman M (2010) Protective effects of black rice bran against chemically-induced inflammation of mouse skin. J Agric Food Chem 58(18):10007–10015

[9] de Araújo, F. F., de Paulo Farias, D., Neri-Numa, I. A., Pastore, G. M. (2021). Polyphenols and their applications: An approach in food chemistry and innovation potential. *Food Chem.* 338, 127535.

[10] Desta, K.T.; Hur, O.S.; Lee, S.; Yoon, H.; Shin, M.J.; Yi, J.; Choi, Y.M. Origin and seed coat color differently affect the concentrations of metabolites and antioxidant activities in soybean (*Glycine max* (L.) Merrill) seeds. *Food Chem.* 2022, *381*, 132249.

[11] Fraga, C. G., Croft, K. D., Kennedy, D. O., Tomás-Barberán, F. A. (2019). The effects of polyphenols and other bioactives on human health. *Food Funct.* 10, 514–528.

[12] Ge, X.; Jing, L.; Zhao, K.; Su, C.; Zhang, B.; Zhang, Q.; Han, L.; Yu, X.; Li, W. The phenolic compounds profile, quantitative analysis and antioxidant activity of four naked barley grains with different color. *Food Chem.* 2021, *335*, 127655.

[13] Ghasemzadeh A, Jaafar HZ, Juraimi AS, Tayebi-Meigooni A (2015) Comparative evaluation of diferent extraction techniques and solvents for the assay of phytochemicals and antioxidant activity of Hashemi rice bran. Molecules 20(6):10822–10838

[14] Ghasemzadeh A, Jaafar HZ, Rahmat A (2015) Phytochemical constituents and biological activities of diferent extracts of Strobilanthes crispus (L.) Bremek leaves grown in diferent locations of Malaysia. BMC complement Altern Med 15(1):422

[15] Gong, E.S.; Luo, S.J.; Li, T.; Liu, C.M.; Zhang, G.W.; Chen, J.; Liu, R.H. Phytochemical profiles and antioxidant activity of brown rice varieties. *Food Chem.* 2017, *227*, 432–443.

[16] Govindaraj, M.; Masilamani, P.V.; Albert, A.; Bhaskaran, M. Role of antioxidant in seed quality—A review. Agric. Rev. 2017, 38, 180–190. [CrossRef]

[17] Katta, M.A.; Khan, A.W.; Doddamani, D.; Thudi, M.; Varshney, R.K. NGS-QCbox and raspberry for parallel, automated and rapid quality control analysis of large-scale next generation sequencing (Illumina) data. PLoS ONE 2015, 10, e0139868. [CrossRef]

[18] Krishnendu Chattopadhyay ,Torit Baran Bagchi , Priyadarshini Sanghamitra ,Sutapa Sarkar, Bishnu Charan Marndi ,Awadesh Kumar ,Moharana ,Shuvendu Shekhar Mohapatra ,Kumar Sahoo .2022 Mapping genetic determinants for grain physicochemical and nutritional traits in unpolished rice using genome-wide association analysis. Research Square DOI: https://doi.org/10.21203/rs.3.rs-1656320/v1

[19] Kumar, A.K.; Govindaraj, M.; Karthikeyan, A.; Shobhana, V.G.; Warkentin, T.D. Genomics-Integrated Breeding for Carotenoids and Folates in Staple Cereal Grains to Reduce Malnutrition. Front. Genet. 2020, 11, 414.

[20] Li, Y. L., Ning, Y., Xu, W. H., Zhou, G. Y. (2015). Dynamic study on two lignans contents in different parts of sinopodophyllum hexandrum. *Chin. J. Chin. Mater. Med.* 40, 1837–1841.

[21] Liu, L.; Guo, J.; Zhang, R.; Wei, Z.; Deng, Y.; Guo, J.; Zhang, M. Effect of degree of milling on phenolic profiles and cellular antioxidant activity of whole brown rice. *Food Chem.* 2015, *185*, 318–325.

[22] Li, W.; Wen, L.; Chen, Z.; Zhang, Z.; Pang, X.; Deng, Z.; Guo, Y. Study on metabolic variation in whole grains of four proso millet varieties reveals metabolites important for

antioxidant properties and quality traits. *Food Chem.* **2021**, *357*, 129791. [Google Scholar] [CrossRef]

[23] Ma, L. K., Tang, L. L., Yi, Q. (2019). Salvianolic acids: potential source of natural drugs for the treatment of fibrosis disease and cancer. *Front. Pharmacol.* 10.

[24] Neri-Numa, I. A., Arruda, H. S., Geraldi, M. V., Júnior, M. R. M., Pastore, G. M. (2020). Natural prebiotic carbohydrates, carotenoids and flavonoids as ingredients in food systems. *Curr. Opin. Food Sci.* 33, 98–107. doi: 10.1016/j.cofs.2020.03.004.

[25] Oki T, Masuda M, Kobayashi M, Nishiba Y, Furuta S, Suda I, Sato T (2002) Polymeric procyanidins as radical-scavenging components in red-hulled rice. J Agric Food Chem 50(26):7524–7529.

[26] Pang, Y.; Ahmed, S.; Xu, Y.; Beta, T.; Zhu, Z.; Shao, Y.; Bao, J. Bound phenolic compounds and antioxidant properties of whole grain and bran of white, red and black rice. *Food Chem.* 2018, *240*, 212–221.

[27] Podio, N.S.; Baroni, M.V.; Wunderlin, D.A. Relation between polyphenol profile and antioxidant capacity of different Argentinean wheat varieties. A Boosted Regression Trees study. Food Chem. 2017, 232, 79–88.

[28] Quideau, S., Deffieux, D., Douat-Casassus, C. (2011). Plant polyphenols: chemical properties, biological activities and synthesis. *Angew. Chem. Int. Ed.* 50, 586–621.

[29] Ragaee, S.; Seetharaman, K.; Abdel-Aal, E.S. The impact of milling and thermal processing on phenolic compounds in cereal grains. *Crit. Rev. Food Sci. Nutr.* 2014, *54*, 837–849.

[30] Rossatto, T.; de-Amaral, M.N.; Benitez, L.C.; Vighi, I.L.; Braga, E.; de Magalhães, J.; Maia, A.M.M.; da Silva, P.L. Gene expression and activity of antioxidant enzymes in rice plants, cv. BRS AG, under saline stress. Physiol. Mol. Biol. Plants 2017, 23, 865–875.

[31] Shasidhar, Y.; Variath, M.T.; Vishwakarma, M.K.; Manohar, S.S.; Gangurde, S.S.; Sriswathi, M.; Sudini, H.K.; Dobariya, K.L.; Bera, S.K.; Radhakrishnan, T.; et al. Improvement of three popular Indian groundnut varieties for foliar disease resistance and high oleic acid using SSR markers and SNP array in marker-assisted backcrossing. Crop J. 2020, 8, 1–15.

[32] Shen, Y.; Jin, L.; Xiao, P.; Lu, Y.; Bao, J. Total phenolics, flavonoids, antioxidant capacity in rice grain and their relations to grain color, size and weight. J. Cereal Sci. 2009, 49, 106–111.

[33] Shridhar, G.; Rajendra, N.; Murigendra, H.; Shridevi, P.; Prasad, M.; Mujeeb, M.A.; Arun, S.; Neeraj, D.; Vikas, S.; Suneel, D.; et al. Modern Diet and its Impact on Human Health. J. Nutr. Food Sci. 2015, 5, 6.

[34] Qiu, Y.; Liu, Q.; Beta, T. Antioxidant properties of commercial wild rice and analysis of soluble and insoluble phenolic acids. *Food Chem.* **2010**, *121*, 140–147. [Google Scholar] [CrossRef]

[35] Ti H, Li Q, Zhang R, Zhang M, Deng Y, Wei Z, Chi J, Zhang Y (2014) Free and bound phenolic profiles and antioxidant activity of milled fractions of different indica rice varieties cultivated in southern China. Food Chem 159:166–174

[36] Verardo, V.; Gmez-Caravaca, A.; Marconi, E.; Segura-Carretero, A.; Garrido-Frenich, A.; Fernmndez-Gutirrez, A. Determination of lipophilic and hydrophilic bioactive compounds in raw and parboiled rice bran. RSC Adv. 2016, 6, 50786–50796.

[37] Xu, F.; Jinsong, B.; Tae-Sung, K.; Yong-Jin, P. Genome-wide Association Mapping of Polyphenol Contents and Antioxidant Capacity in Whole-Grain Rice. J. Agric. Food Chem. 2016, 64, 4695–4703.

[38] Yahfoufi, N., Alsadi, N., Jambi, M., Matar, C. (2018). The immunomodulatory and anti-inflammatory role of polyphenols. *Nutrients* 10, 1618–1640. doi: 10.3390/nu10111618.

[39] Yu J, Xiong H, Zhu X, Zhang H, Li H, Miao J, Wang W, Tang Z, Zhang Z, Yao G, Zhang Q, Pan Y, Wang X, Rashid MAR, Li J, Gao Y, Li Z, Yang W, Fu X, Li Z (2017) OsLG3 contributing to rice grain length and yield was mined by Ho-LAMap. BMC Biol 15:28

[40] Zhai, S.N.; Xia, X.C.; He, Z.H. Carotenoids in Staple Cereals: Metabolism, Regulation, and Genetic Manipulation. Front. Plant Sci. 2016, 7, 1197

[41] Zhang MW, Zhang RF, Zhang FX, Liu RH (2010) Phenolic profiles and antioxidant activity of black rice bran of different commercially available varieties. J Agric Food Chem 58(13):7580–7587

[42] Zhang, H., Tsao, R. (2016). Dietary polyphenols, oxidative stress and antioxidant and anti-inflammatory effects. *Curr. Opin. Food Sci.* 8, 33–42.

[43] Zhang, H.; Shao, Y.; Bao, J.; Beta, T. Phenolic compounds and antioxidant properties of breeding lines between the white and black rice. Food Chem. 2015, 172, 630–639.

[44] Zhu, C.; Kobayashi, K.; Loladze, I.; Zhu, J.; Jiang, Q.; Xu, X.; Liu, G.; Seneweera, S.; Ebi, K.L.; Drewnowski, A.; et al. Carbon dioxide (CO2) levels this century will alter the protein, micronutrients, and vitamin content of rice grains with potential health consequences for the poorest rice-dependent countries. Sci. Adv. 2018, 4, eaaq1012.