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Research Article

EFFECT OF VARYING TRANSPLANTING TIMES ON GRAIN QUALITY CHARACTERISTICS OF RICE (*Oryza sativa* L.) GENOTYPES

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Abstract- To investigate the effect of varying transplanting dates on rice grain quality characteristics and to compare the protein profile of flag leaf at different dates of transplanting in rice (*Oryza sativa* L.) genotypes *viz*., heat tolerant (N22), moderately heat susceptible (IR8) and fine grain rice variety (PR116), were transplanted at three different times i.e., D1 (very early), D2 (early) and D3 (normal date of transplanting). No predictable difference was observed in the electrophoretic pattern of protein bands in all the three cultivars. The 1000-grain weight of paddy, brown rice and milled rice decreased at D1 and D2, particularly in IR8. Grain dimensions were unaffected by different transplanting dates. The head rice recovery was decreased significantly at D1 (53.83- 65.96 %) and D2 in all the three cultivars with the maximum head rice recovery (60.24- 68.60%) being obtained at normal transplanting time. Early transplanting caused significant increase in the percentage of chalky grains (%) in the susceptible genotype IR8 compared N22 and PR 116. Higher temperature during very early and early transplanting times had great impact on chalkiness. The heat susceptible cultivar were observed to be prone to heat stress encountered during very early and early DOT as maximum chalkiness was recorded in IR8 during D1 (12.80 %) followed by D2 (9.60 %) and D1 (6.75 %) respectively.

Keywords-Rice, *Oryza sativa*, transplanting time, grain quality, head rice recovery, chalkiness.

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Introduction

Rice (*Oryza sativa* L.) is the essential food of over half of the world's population. The average temperature of earth has been recorded to rise by 1.4°F over the past century and is predicted to rise another 2 to 11.5°F in coming next hundred years. The detrimental effect of high temperature to rice has become a major problem during the reproductive growth period. During grain filling period rice crop has been exposed to heat stress due to rise in temperature [1,2]. It has been reported that the rice grain yield would decline by 10% for each 1°C increase of minimum temperature during the growing season. It has been shown that adoption of high temperature tolerant cultivars is one of the most effective counter measures to maintain high productivity and stability of rice under the anticipated climate in temperate regions.

The physicochemical characteristics of rice grains are important indicators of grain quality. Grain quality is mainly determined by the combination of many physical as well as chemical characters. The physical quality characters include kernel size, shape, hulling, milling and head rice recovery. The chemical quality is mainly determined by amylose content, gelatinization temperature, gel consistency and cooking behavior [3]. High temperature decreases seed set as well as affects grain quality [4, 5] because of reduction in endosperm sink strength and incomplete grain filling. High quality grains are required to establish rice crops with high yield potential. The impact of heat stress on rice grain quality has received less attention compared to yield. Early sowing is considered as the vector to reduce the growth and yield. On this basis, the present study was planned to obtain a deeper insight into the variation in physicochemical properties of economic importance.

Materials and Methods

The present investigation was carried out in the Department of Plant Breeding and Genetics (Rice Section), Punjab Agricultural University, Ludhiana. Three rice genotypes *viz.* heat tolerant N22 and heat susceptible IR8 fine grain rice variety PR116 were employed in this study. The rice genotypes were sown during mid-March (very early, D1), mid-April (early, D2) and mid-May (normal, D3). The seedlings of rice cultivars were raised in the wooden trays (50 x 40 x 10 cm) in the glasshouse. Thirty- day old seedlings were transplanted in the field in complete randomized block design with three replications. The recommended agronomic practices were followed.

Dehusking and milling: To determine milling characters of crop grains, weighed samples (125 g at 14 per cent moisture content) of cleaned paddy were dehusked in a laboratory sheller equipped with rubber roller (Satake Rice Sheller, Satake Engg. Co., Japan). The shelled rice (brown rice) was weighed and expressed as the percentage of rough rice. Brown rice samples were polished in the Mc Gill Miller No. 2 (USA) in such a way to obtained 6 per cent degree of polish in all the samples and expressed as the per cent of rough rice which gives the value of milled rice recovery. The laboratory model Satake Test Rice Grader (Satake Engg. Co. Ltd., Japan) was used to separate the broken rice from the polished rice. The kernels with more than three- fourth length were considered as head rice. The head rice recovery and broken rice yields were calculated as the percentage of rough rice.

Physicochemical Properties

Thousand Kernel weight: One thousand head rice kernels of paddy, brown and milled rice were counted randomly in triplicate and weighed separately. Mean of three replications was reported.

Length–Breadth ratio (L/B): Length and breadth of milled rice was measured with a dial gauge and their additive measurements (in mm) were taken. L/B ratio was determined by dividing length to breadth. A mean of 10 replications was calculated.

Determination of chalky grains: When 50% or more of the milled rice kernel was opaque (white spot) rather than translucent, it was considered "chalky". Two grams (2 g) milled rice grains were placed on a blue cardboard background to visually identify under white light the grains that had white belly, white centre, white back or a combination of these. The percent chalky grains were determined as: % Chalky grain = weight of chalky grain ×100/2 g.

Cooking properties

Water uptake ratio: Head rice sample (2 g) was cooked in 20 ml distilled water for minimum cooking time in a boiling water bath. The contents were drained and the superficial water on the cooked rice was sucked by pressing the cooked samples in filter paper sheets. The cooked samples were then weighed accurately and the water uptake ratio was calculated and expressed as a percent.

Elongation ratio: Cumulative length of 10 cooked rice kernels was divided by length of 10 uncooked raw kernels and the result was reported as elongation ratio.

Cooked length–breadth ratio: L/B ratio was determined by dividing the cumulative length of 10 cooked kernels by the breadth of 10 cooked kernels. A mean of 10 replications was reported.

Alkali Spreading Value: The test was performed with the method described by Little *et al.* [6] to calculate gelatinization temperature. Six kernels per sample were placed evenly in a Petri dish. Ten ml of KOH (1.7%) solution was added. Petri dishes were kept undisturbed for 23 hours at room temperature (21°C–24°C). Spreading and clearing of rice kernels was observed and the score was given according to the scale [Table-1]. A rating of spreading of 1-3 is classified as high (>74°C), 4-5 as intermediate (70°C-74°C) and 6-7 as low (55°C-69°C) gelatinization temperature. Clearing values are usually 1-unit lower than spreading values.

Table-1 *Characteristics regarding spreading and clearing of rice kernels and corresponding score*

Score	Spreading	Clearing
1.	Kernel not affected	Kernel chalky
2.	Kernel swollen	Kernel chalky, collar powdery
3.	Kernel swollen, collar complete or narrow	Kernel chalky, collar cottony or cloudy
4.	Kernel swollen collar complete and wide	Centre cottony, collar cloudy
5.	Kernel splits or segregated, collar complete and wide	Centre cottony, collar clear
6.	Kernel dispersed merging with collar	Centre cloudy, collar clear
7.	Kernel completely dispersed and intermingled	Centre and collar clear

Gel Consistency: The gel consistency is determined by gel consistency test described by Cagampang*et al.* [7]. According to this method, whole grain of milled rice ground to pass through a 100-mesh sieve. Weighed 100 mg of powder in duplicate in culture tubes (100 x12 mm). Added 0.2 ml of 95 % ethanol containing 0.025 % thymol blue. Added 2.0 ml of 0.2 N KOH and mix with a cyclomixer/vortex mixer. Covered the tubes with glass marbles and heat in a vigorously boiling water bath for 8 minutes making sure that the tube contents reach 2/3rd the height of the tube. Removed from water bath and let them stand for 5 minutes. Then cooled in ice water bath for 20 minutes. Lay the tubes horizontally over a graph paper spread on a table. Do not disturb for one hour. Measure the total length of the gel (mm) from the bottom of the tube to the gel front.
Gel consistency was characterized as follows;

Length of gel (mm)	Gel consistency
40 mm or less	Very flaky rice with hard gel consistency
41 – 61 mm	Flaky rice with medium gel consistency
More than 61 mm	Soft rice with soft consistency

Amylose content: Procedure of Juliano [8] with little modifications was used for estimating the amylose content. A 100-mg powdered dried sample was mixed with 1 mL of 950 mL L^{-1} ethanol and 9 mL of 1 mol L^{-1}NaOH and kept overnight. The total volume was made up to 100 mL with distilled water and mixed well. 0.1 mL of 1 mol L^{-1} acetic acid and 0.2 mL of iodine solution were added to 0.5 mL aliquot and the final volume was made up to 10 mL with distilled water. The colour developed was read at 620 nm against a reagent blank. A standard curve was prepared using potato amylose in the range 40–200 µg.

Crude Protein Content: To determine protein content, the per cent nitrogen estimated in the milled rice kernels by Kjeldahl's method was multiplied by 5.95 to get per cent crude protein [9].

Statistical Analysis: Analysis of variance statistically analyzed the data. For significant mean effects, means were separated using least significant test (LSD).

Results and Discussion

Temperature Profile

In present study, the temperature rise observed was 4.4°C and 2.4°C during N22 D1 and D2 respectively while it was 6.2°C and 4.4°C in IR8 and PR116 during D1 and D2 respectively [Fig-1].

Protein Profiling

SDS-PAGE profile of the proteins in the flag leaf at the dough stage did not show significant changes in the protein bands observed except that varying intensity of these bands was observed [Fig-2]. Major bands were observed between 29.0 kDa and 14.3 kDa.

Changes in the physicochemical characteristics of rice grains at different transplanting time

These characters can be used as selection indices for the improvement in grain quality characteristics of rice. Surveys showed that consumer acceptance of a variety depends primarily on its cooking and eating quality [10].

1000 grain weight

In case of N22, the 1000 grain weight of paddy was observed maximum at D3 and D1 (19.98 g) followed by D2 (19.94 g) [Table-1]. IR8 had maximum value of 1000 grain weight at D3 (28.58 g) followed by D2 and D1 (27.89 and 27.20 g). PR116 also followed the same trend with 25.98, 23.58 and 22.80 g at D3, D2 and D1 respectively. Brown rice and milled rice also followed the same pattern by attaining maximum value during normal transplanting date (D3) followed by early (D2) and very early date of transplanting (D1) [Table-1]. IR8 attained the maximum value of 1000 grain weight of paddy, brown rice and milled rice as compared to N22 and PR116.

Length–breadth ratio (L/B)

Grain length, breadth, and length: breadth ratio did not change with the change in transplanting dates [Table-1]. The maximum length and breadth was observed in PR116 and IR8 respectively, though the difference was statistically non-significant.

Hulling

Hulling % was observed almost similar in IR8 and PR116 at three DOT whereas N22 had lower values comparatively but the difference was non- significant [Table-1].

Milling
In case of milling %, PR116 showed lower values compared to N22 and IR8 where these two cultivars showed similar values at all the three DOT [Table-1]. Again milling % was also observed to be statistically non- significant in all the cultivars at three DOT.

Head Rice Recovery
Head rice recovery (HR) is the proportion of the intact grain which is more than or equal to 3/4th its length. HR indicates that percent of whole grains obtained after milling processing. For quality evaluation, HR recovery is one of the most important characters and more than 65% of HR recovery is desirable. In the present study, heat stress during early and very early transplanting times affected HR significantly in all the cultivars. A decrease was observed in the head rice recovery in D1 and D2 in all the three cultivars with the maximum head rice recovery being attained at D3 [Table-1]. During D1, 53.83- 60.43% head rice recovery was observed. While it was recorded to be 59.19- 65.96% during D2 and 60.24- 68.60% during normal transplanting time (D3) respectively. It has been reported that at ambient CO_2 concentration, higher temperature increased the proportion of broken grains from 10% at 38°C, but the temperature had no effect on the length and breadth of the grain [11].

Chalkiness
Chalkiness is a visually discernible parameter. Higher temperature during very early and early transplanting times had great impact on chalkiness. Early transplanting caused significant increase in the percentage of chalky grains (%) in the susceptible genotype IR8 compared N22 and PR 116. The mean chalky percent was observed to statistically similar in N22 at D1 (4.28 %) and D2 (4.08 %) while 3.65 % chalkiness was observed at D3 [Table-1]. The heat susceptible cultivar were observed to be prone to heat stress encountered during very early and early DOT as maximum chalkiness was recorded in IR8 during D1 (12.80 %) followed by D2 (9.60 %) and D1 (6.75 %) respectively. [Table-1]. It can be observed that heat stress during grain filling facilitates the formation of chalky grains thus lowering consumer acceptability and commercial value of rice cultivars.

Changes in the chemical properties of rice grains at different transplanting time
Gel consistency
Gel consistency (GC) was observed to be statistical significantly higher in N22 compared to IR8 and PR116 at all the transplanting dates being maximum at D2 (94.00 mm) followed by D3 (92.00) and D1 (91.50 mm) [Table-2]. In IR8, maximum GC value was observed in D1 (86.50 mm) followed by D2 (77.00 mm) and D3 (45.00 mm) respectively. While in case of PR116, the maximum value was recorded at D2 (90.50 mm) followed by D3 (80.00 mm) and D1 (79.50 mm).

Water Uptake Ratio
The water uptake ratio was observed to be higher in IR8 compared to N22 and PR116 though the difference was non-significant with respect to DOT. Within date of transplanting higher value was observed during D3 except PR116 [Table-2].

Volume expansion Ratio
Similar trend was observed as that of water uptake ratio. Similar volume expansion ratio was observed in IR8 and PR116 at all the dates of transplanting followed by N22 [Table-2]. Though the difference between cultivars and DOT was non- significant.

Kernel Length after cooking
Kernel length after cooking followed the same trend as that of volume expansion ratio [Table-2].
Elongation Ratio (ER) and Alkali-Spreading Value (ASV) of the grains remained statistically unchanged with different transplanting time [Table-2].

Amylose content Amylose content decreased in all the three cultivars at D1 and D2 [Table-2]. Maximum amylose content was observed in IR8 (27.63 %) during normal transplanting time whereas N22 possessed minimum amylose content at both D1 (13.51 %) and D2 (13.37 %) whereas PR116 possessed amylose content (22.23- 23.21%) in between heat tolerant (N22) and heat susceptible cultivars (IR8) at the three transplanting dates [Table-2]. It has been reported that high temperature decreased amylose content of IR20 and Fujiraka [12] and similar results were obtained in other japonica cultivars [13,14]. Zheng *et al* [15] showed that the effect of high temperature on apparent amylose content and gel consistency in milled rice was cultivar dependent. Under high temperature amylose content increased or remained statistically unchanged in cultivars with high amylose content whereas it decreased in cultivars having lower amylose content. Both drought and heat stress reduce carbohydrate accumulation, which can influence protein concentration, by allowing more N concentration per unit of starch accumulation in the grains [16].

Crude Protein Content Heat stress during grain filling can increase protein concentration, but it significantly decreases the functionality of the protein [17]. In the present study, crude protein content increased from D3 to D1 [Table-2]. N22 showed maximum protein content at D3 (7.04 %) followed by D2 (6.71 %) and D1 (5.16 %) respectively. N22 had statistically higher crude protein compared to IR8 and PR116. Similar trend was followed by PR116 and IR8 being higher at D3 followed by D2 and D1. In case of PR116, the protein content was observed to be higher at D3 (6.92 %) followed by 6.37 % and 5.05 % at D2 and D1 respectively. Similarly, IR8 showed higher protein content at D3 (6.85%) followed by D2 (5.89 %) and D1 (4.65 %) respectively.

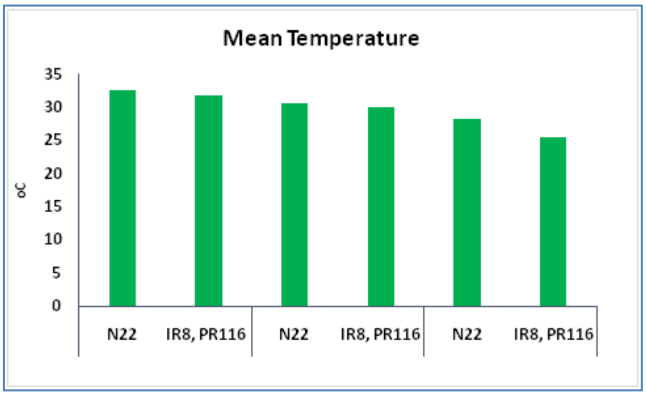

Fig-1 Temperature Profile

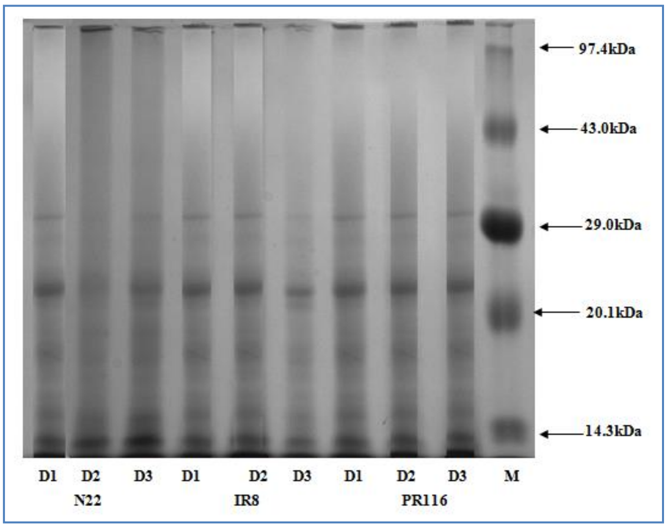

Fig-1 Effect of different dates of transplanting on the electrophoretic pattern of total proteins from flag leaf at dough stage of heat tolerant (N22), moderately heat susceptible (IR8) and fine grain rice variety (PR116). M, protein marker; D1= very early transplanting (mid- April), D2= early transplanting (mid- May) and D3= normal transplanting (mid-June).

Table-1 *Physical and milling characteristics of three rice cultivars at different transplanting time*

Date of transplanting	Variety	1000 Grain weight (g)			Grain length (mm)	Grain breadth (mm)	L/B ratio	Hulling (%)	Milling (%)	Head rice (%)	Chalkiness (%)
		Paddy	Brown rice	Milled rice							
15 May (D1)	N22	19.94±1.14	17.40±1.0	13.96±0.84	5.49±0.49	2.42±0.18	2.26±0.04	78.66±0.86	72.62±0.83	60.43±1.05	4.28±0.02
	IR8	27.20±1.00	25.20±1.0	20.95±0.45	6.73±0.20	2.48±0.02	2.71±0.06	79.47±0.78	73.67±0.73	57.43±1.03	12.80±1.10
	PR116	22.80±1.00	21.70±0.5	17.39±0.39	7.09±0.09	2.24±0.07	3.17±0.13	79.46±0.74	70.93±1.05	53.83±1.03	7.75±0.2
15 May (D2)	N22	19.72±0.17	17.6±1.20	15.53±1.03	5.45±0.12	2.21±0.09	2.47±0.15	78.96±0.82	71.57±0.57	65.96±1.00	4.08±0.03
	IR8	27.89±0.31	25.4±0.80	21.84±0.20	6.69±0.09	2.51±0.14	2.67±0.18	79.51±0.81	72.83±1.03	62.50±1.00	9.60±0.025
	PR116	23.58±1.23	22.60±0.6	18.14±0.04	7.01±0.10	2.26±0.15	3.11±0.16	78.99±1.01	69.58±1.38	59.19±1.01	5.70±0.6
15 June (D3)	N22	19.94±0.56	17.8±1.00	15.74±0.46	5.48±0.02	2.25±0.15	2.44±0.15	79.43±0.89	72.69±1.17	68.60±1.10	3.65±0.02
	IR8	28.58±0.31	25.80±0.2	22.17±0.17	6.71±0.42	2.50±0.30	2.72±0.49	80.31±0.32	74.06±0.55	64.08±1.08	6.75±0.21
	PR116	25.98±0.43	23.80±0.7	18.92±0.82	7.06±0.26	2.23±0.13	3.17±0.06	80.29±0.31	71.70±1.10	60.24±0.96	0.25±0.001
LSD (5%) A= Rice varieties B=Date of transplanting	A	0.68	0.59	0.30	0.40	0.09	0.11	0.17	0.28	1.01	1.07
	B	0.68	0.59	0.30	NS	NS	NS	NS	NS	NS	1.07
	AB	1.19	NS	NS	NS	NS	NS	NS	NS	NS	1.85

Table-2 *Eating and cooking quality of the rice cultivars at different transplanting time*

Date of transplanting	Variety	Water uptake ratio	Volume expansion ratio	GC(mm)	KLAC (mm)	ER	Amylose content%	Crude protein%	ASV
15 April (D1)	N22	8.73±0.09	3.00±0.30	91.50±0.50	8.3±0.1	1.51±0.07	13.51±0.81	5.16±0.005	2
	IR8	31.53±1.01	3.75±0.16	86.50±6.50	10.15±0.05	1.51±0.03	26.03±0.98	4.65±0.15	6
	PR116	30.97±2.15	3.75±0.10	79.50±0.50	11±0.5	1.55±0.02	22.23±0.98	5.05±0.05	6
15 May (D2)	N22	8.77±0.06	3.50±1.0	94.00±1.00	8.2±0.2	1.48±0.04	13.37±1.13	6.71±0.01	2
	IR8	31.04±1.09	3.75±0.07	77.00±1.00	10.35±0.15	1.55±0.02	26.71±0.85	5.89±0.23	6
	PR116	31.41±0.61	3.75±0.06	90.50±0.50	10.65±0.15	1.52±0.06	22.76±0.90	6.37±0.10	6
15 June (D3)	N22	9.52±0.50	3.50±1.10	92.00±1.00	8±0.2	1.46±0.04	14.28±1.08	7.04±0.2	2
	IR8	32.02±0.98	3.75±0.03	45.00±3.00	10.5±0.1	1.56±0.02	27.63±1.17	6.85±0.13	6
	PR116	29.55±2.01	3.75±0.01	80.00±1.00	10.95±0.05	1.55±0.04	23.21±0.41	6.92±0.1	7
LSD (5%)	A	1.21	NS	2.58	0.22	0.03	0.61	0.12	0.000003
A= Rice varieties	B	NS	NS	2.58	NS	NS	0.61	0.12	0.000003
B=Date of transplanting	AB	NS	NS	4.47	NS	NS	NS	0.20	0.000005

± Standard deviation; GC- Gel consistency; KLAC- Kernel length after elongation; ER- Elongatio ratio; ASV- Alkali spreading value

Conclusion

From above it can be concluded that High temperature during grain development caused a decrease in quality of rice grains. High temperature led to changes in the grain length and protein content. Apart from the environmental factors, genotypic variations were also observed. The heat susceptible genotype, IR8 was severely affected in terms of appearance and quality indicating that the mechanism of grain chalkiness under high temperature stress is considerably complicated whereas heat tolerant genotype, N22 was least affected by heat stress encountered during early and very early transplanting times.

Application of Research: Our results have clear implications that heat stress strongly affects rice quality. As, under high temperature conditions, N22 observed to be superior in terms of quality and appearance as compared to heat susceptible (IR8) and fine grain quality variety (PR116). Our result supports the acceptance of heat tolerant cultivars. Therefore, heat tolerant cultivars should be adopted in the time of increasing global warming.

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Author contribution: Sharma Kanu Priya carried out the experimental work and wrote the manuscript. Sharma Neerja designed, provided scientific support, and co-wrote the manuscript.

Abbreviations

ASV: Alkali spreading value
DOT: Date of transplanting
ER: Elongation ratio
GC: Gel consistency
HR: Head rice recovery
KLAC: Kernel Length after cooking
L/B: Length breadth ratio
LSD: Least significant test
SDS- PAGE: Sodium dodecyl sulphate Polyacrylamide gel electrophoresis

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.

Conflict of Interest: None declared

References

[1] Morita S., Wada H. and Matsue Y. (2016) *Plant Production Science,*19, 1–11.
[2] Jagadish S.V.K., Murty M.V.R. and Quick W.P. (2015) *Plant,Cell Environment,*38, 1686–1698.
[3] Khatun M.M, Ali M.H. and Cruz Q.D.D. (2003) *Pakistan Journal of Biological Science*, 6, 511-13.
[4] Shah F., Saddam H, Shah S., Shah H., Bhagirath S.C.,Fahad K., Muhammad Z.I., Abid U., Chao W., Ali A.B, Hesham A., Amanullah, Wajid N., Babar S., Mohsin T.and Jianliang H. (2016) *PLoS One,*11(7), e0159590.
[5] Jagadish S.V., Muthurajan R., Oane R., Wheeler T.R., Heuer S., Bennett J., Craufurd P.Q. (2010) *Journal of Experimental Botany*, 61,143-156.
[6] Little R.R., Hilder G.B. and Dawson E.H. (1958) *Cereal Chemistry,* 35, 111-26.
[7] Cagampang G.B., Perez C.M. and Juliano B.O. (1973) *Journal of Science and Food Agriculture,* 24, 1589-94.
[8] Juliano B.O. (1971) *Cereal Science Today,* 16, 334-340.
[9] Bali A.S. and Uppal H.S. (1999) *Indian Journal of Agricultural Sciences,* 69,

325- 328.

[10] Juliano B.O, Cagampang G.B., Cruz L.J. and Santiago R.G. (1964) *Cereal Chemistry,* 41, 275-85.

[11] Madan P., Jagadish S.V.K., Craufurd P.Q., Fitzgerald M., Lafarge T. and Wheeler T.R. (2012) *Journal of Experimental Botany,* 63, 3843–52.

[12] Resurreccion A.P., Hara T., Juliano B.O. and Yoshida S. (1977) *Soil Science and Plant Nutrition*, 23, 109-12.

[13] Asaoka M., Okuno K. and Fuwa H. (1985) *Agricultural BiolChem,* 49, 373-79.

[14] He G.C, Kogure K. and Suzuki H. (1990) *Japanese Journal of Crop Science,*59, 340-45.

[15] Zheng J.C., Sheng J., Tang R.S., Shi C.L. and Chen L.G. (2007). *Jiangsu Journal of Agricultural Sciences,* 23, 1-4.

[16] Stone P.J. and Nicolas M.E. (1994) *Australian Journal of Plant Physiology*, 21, 887–900.

[17] Corbellini M., Canevar M.G, Mazza L., Ciaffi M., Lafiandra D. and Borghi B. (1997) *Australian Journal of Plant Physiology,* 24, 245-60.