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Society for Advancement of Rice Research





Society For Advancement of Rice Research

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- To advance the cause of rice research and development in the country.
- To disseminate knowledge on latest developments in rice research through publications, seminars, lectures and training programmes.
- To provide consultancy in rice production and development.
- To facilitate research and industry collaboration and public private partnership at national level.
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REVIEW ARTICLE

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Understanding the Mechanism of Iron Metabolism and Bioavailability in Cereals towards Biofortification

Suman K, Jaldhani V, Sanjeeva Rao D, Aravind Kumar J, Sruthi K, Mangrauthia SK, Kalyani MB, Papa Rao Vaikuntapu, Sai Prasad SV, Sundaram RM and Neeraja CN*

ICAR-Indian Institute of Rice Research, Rajendranagar, Hyderabad - 500 030

*Corresponding author Email: cnneeraja@gmail.com

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Abstract

Iron (Fe) deficiency remains a critical global health issue, particularly affecting vulnerable populations such as children and pregnant women. Biofortification, the process of enhancing the micronutrients content in staple food crops, holds promise in addressing this challenge. This review aims to elucidate the mechanisms underlying Fe metabolism in cereals, thus focusing on strategies for Fe biofortification. Understanding the molecular mechanisms governing Fe uptake and transport in plants is essential for targeted breeding efforts to enhance the Fe content. Plants employ distinct strategies for Fe uptake from the soil, such as reduction-based and chelation-based approaches, influenced by environmental factors like soil pH. Long-distance Fe transport within plants involves intricate pathways mediated by transporter proteins and regulatory genes. Environmental factors, including soil properties and agricultural practices, influence Fe bioavailability in crops apart from Fe accumulation. Thus, strategies to enhance Fe absorption such as reducing phytic acid content are crucial for improving the nutritional quality of biofortified crops. Various *in vitro*, animal and human studies have assessed the bioavailability of Fe in biofortified crops, highlighting the potential for addressing Fe deficiency through dietary interventions. Combining genetic approaches with an understanding of physiological mechanisms can hasten grain Fe enrichment efforts, resulting in better outcomes through biofortification programmes.

Key words: Cereals, Biofortification, Fe metabolism, Bioavailability.

Introduction

The Green revolution has almost achieved food security across the world addressing the hunger through increasing the production of major staple foods including rice, wheat and other cereals. The prevalent reliance on carbohydrate-rich diets, coupled with restricted dietary diversity due to limited purchasing power in low or middle-income countries, is exacerbating hidden hunger, also known as micronutrient malnutrition (Black *et al.*, 2013). The significance of the nutritional quality of the diets has been underscored by United Nation's (UN) Sustainable Development Goal-2 targeting to eliminate hunger,

accomplish food security and enhanced nutrition and promote sustainable agriculture (Lowe, 2021). According to the World Health Organization (WHO) estimates, around 40% of children below five years, 37% of pregnant women and 30% women between 15 to 49 years suffer Fe deficiency (https://www.who.int/). The requirements for Fe almost doubles between 1 and 6 years of age and also during adolescence/puberty, thus children, adolescents, women of gestation reproductive age and pregnant women are the most vulnerable to Fe deficiency (WHO, 2005; Abbaspour et al., 2014). Almost 70% of the Fe in human body is



found in red blood cells (RBC) as haemoglobin and in muscles as in myoglobin facilitating the circulation and metabolism of oxygen among various tissues (McDowell, 1992; Hurrell, 1997). One-fourth of Fe is stored as ferritin to maintain Fe homeostasis and to support important cellular processes (Knovich *et al.*, 2009). For plants also, Fe is a critical essential element and deficiency of Fe is directly related to the reduction in crop productivity and quality (Grotz and Guerinot, 2006).

Increasing Fe content in food grains

Agronomic biofortification is the application of external Fe salts to the plant parts to increase Fe content in grains. However, it is simple and effective, additional cost required for the purchase of Fe salts and labour for the application hinders the wide adoption of agronomic biofortification. Genetic biofortification is a proven approach to enhance Fe content in cereals especially in pearl millet and wheat (Neeraja et al., 2022). Biofortification can be achieved through either traditional breeding or genetic engineering. The conventional breeding methodology is based on the existence or availability of genetic variability, crossing to combine the high Fe and yields, selection of desirable recombinants from the segregating material and their stabilization to be released as varieties (Vasconcelos et al., 2017). For the use of marker assisted selection (MAS), several attempts were or being made to identify genomic regions and candidate genes associated with high Fe content in target edible tissues using approaches like QTL mapping, GWAS and genomic selection across crops (Srivastava et al., 2020; Gupta et al., 2021; Swamy et al., 2021). Recent identification of ZmNAC78, a transcription factor associated with high Fe levels in maize appears to be promising for MAS (Yan et al., 2023). Genetic engineering approach has demonstrated its potential for enhancing Fe content in cereals, however its adoption is restrained by constraints of regulatory authorities worldwide (Garg et al., 2018). Using

gene editing strategy, one or a few nucleotides can be changed, existing alleles can be replaced and new genes can be inserted precisely and can be inherited stably (Huang et al., 2016). Genome editing for targeted gene editing through Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) involves Cas9/13, RNA-guided DNA endonucleases guided by a single guided RNA (sgRNA) resulting in a complex at the target site (Roy and Soni, 2021; Ahmad et al., 2020). Gene editing of OsNRAMP2 increased the grain Fe content in rice (Chang et al., 2022). Knocking out anti-nutrient genes responsible for accumulation of heavy metals and phytic acid can reduce the accumulation anti-nutrients. Disruption of inositol penta phosphate 2-kinase 1 (IPKI) gene increased grain Fe content in wheat (Aggarwal et al., 2018). Supposed to be transgene free technology, the gene editing appears to be promising with supportive regulatory framework across the world. For targeted breeding efforts to increase Fe content in cereals, deciphering the molecular mechanism of Fe translocation and remobilization into grains is very critical. In the present review, we summarized the uptake and transport mechanisms of Fe and the associated genes in model plants and cereals.

Translocation of Fe from root to loading in grains

Available literature and bioinformatics resources focusing on candidate genes and gene families associated with the translocation of Fe from the roots to the grain-loading process in crops *viz.*, rice, wheat and maize are summarized below.

Fe uptake from soil to roots

Plants employ two distinct strategies for Fe uptake from the soil:

- 1. Strategy I, known as the reduction-based strategy, is activated by non-grass plants when they experience Fe deficiency.
- 2. Strategy II, referred to as the chelation-based strategy, is triggered in grasses (**Figure 1**).



Strategy I: Reduction-based strategy

The Strategy I (reduction-based strategy), is predominantly employed by non-graminaceous plants. This reduction-based strategy hinges on the activity of the Fe-regulated transporter 1 (IRT1). Here is how it works:

- In Strategy I, the available Fe³⁺ in the rhizosphere is first converted into Fe2+ through a reduction process by the plant before it can be taken up.
- When plants are under Fe-deficient conditions, they release protons (H+) into the rhizosphere, which leads to a decrease in pH in the immediate root vicinity. This acidification process is facilitated by ATPases, which utilize ATP to pump protons into the rhizosphere (Kim and Guerinot, 2007).
- As the pH decreases in the rhizosphere, the solubility of ferric oxides (Fe³⁺) increases.
- Furthermore, an enzyme called ferric reductase oxidase 2 (FRO2) aids in the reduction of ferric oxides (Fe³⁺) to ferrous oxide (Fe²⁺), using NADPH-dependent Fe³⁺ chelate reductase. This conversion makes Fe more soluble.
- Subsequently, the soluble ferrous oxide (Fe^{2+}) is transported from the rhizosphere to the roots, primarily through a transporter controlled by IRT1 (**Figure 1**) (Ishimaru *et al.*, 2006).

Strategy II: chelation-based strategy

Strategy II (chelation-based strategy) is primarily employed by graminaceous plants viz., maize, wheat and rice from the grass family for Fe uptake. Here is how it works:

- Strategy II transport activities are controlled by transporters known as mugineic acid (TOM1) and yellow stripe 1 (YS1).
- In Strategy II, ferric ions (Fe³⁺) present in the rhizosphere are transported into the root cytosol with the help of soluble phyto-siderophores. These siderophores are natural Fe chelators with a high affinity for Fe³⁺ transport (Morrissey and Guerinot, 2009).

- Among these chelators, the mugineic acid (MA) belonging to family of phytosiderophores (PS) is particularly effective in binding to Fe³⁺. Different plant species secrete various members of the MA family, depending on their specific needs. For instance, rice primarily releases 2' deoxymugineic acid (DMA), while barley releases two different types of MAs, viz., 3' epihydroxymugineic acid (epi HMA) and 3' epihydroxy 2' deoxymugineic acid (epi HDMA), near the rhizosphere through the TOM1 transporter (Ishimaru et al., 2006).
- These MAs efficiently form Fe³⁺-MA complexes.
- The YS1 transporter then transported the formed complexes into the root (Schaff et al., 2004 and Ishimaru et al., 2006). This chelation-based strategy enhances the uptake of Fe in graminaceous plants.

Certain crops, including rice, are capable of employing a combination of both reduction based (Strategy I) and chelation based (Strategy II) strategies for Fe uptake from the rhizosphere into the roots. Here is how it works

- In this combined strategy, plants directly absorb the soluble ferrous oxide (Fe²⁺) from the rhizosphere, which can be richer in Fe²⁺ compared to Fe³⁺. This is facilitated through transporters like IRT1 and/or IRT2 (Kim and Guerinot, 2007; Sperotto et al., 2012).
- Simultaneously, via Strategy Fe³⁺-MA II, complexes are formed in the rhizosphere. These Fe³⁺-MA complexes are then transported into the root's cytosol using transporters like Yellow stripe -like 15 (YSL15) (Ishimaru et al., 2006).
- Instead of relying solely on direct Fe²⁺ uptake from the rhizosphere, rice successfully utilizes both reduction and chelation-based strategies, ensuring sufficient Fe is absorbed from the rhizosphere into the root cytosol through the Fe³⁺-MA complexes (see Figure 1) (Ishimaru et al., 2006).



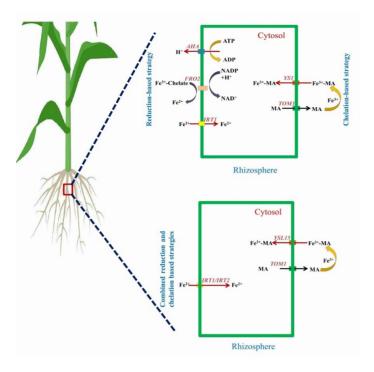


Figure 1: Fe uptake strategy I and II in plants

The expression of different sets of ferric reduction oxidase (FRO) genes in various locations suggests their role in Fe uptake in different plant tissues. FRO2 corresponding gene to the yeast Fe (III) reductase 1 (FRE1) was identified in Arabidopsis based on its sequence similarity (Robinson et al., 1999). FRO2 is a primary FER expressed in the epidermal cells of Fe inadequate roots. In low Fe growth condition, the over expression of FRO2 makes plants more resistant (Connolly et al., 2003). Root specific FRO genes (FRO2, FRO3 and FRO5) are expressed in roots, specifically FRO3 gene expressed in the vascular cylinder of roots. Shoot specific FRO genes (FRO6, FRO7 and FRO8) are expressed in shoot (Feng et al., 2006). The orthologs of IRT1 gene have been identified which combines both Strategy I and II for Fe uptake in rice. Unlike Arabidopsis, the *LeIRT1* and LeIRT2 genes are expressed in the roots of tomato in both the Fe deficient and sufficient roots, especially LeIRT1, shown induction under Fe deficiency (Eckhardt et al., 2001). Ethylene is also associated in the stress adaptation like Fe deficiency in rice, although not in barley (Wu et al., 2011). Additionally,

Methylthioribose kinase (MTK) and S-adenosyl methionine synthetase (SAM) genes are expressed under Fe deficiency conditions in chelation-based strategy plants viz., rice and maize (Liu et al., 2015).

Several endogenous or housekeeping genes have been associated with grain Fe content in rice. For example, the ubiquitin activating enzyme (UBA), a small globular protein involved in the ubiquitination process, has shown a significant positive correlation with Fe concentration in rice grains. This suggests a potential role for UBA in Fe homeostasis, in addition to the reported ubiquitin-conjugating and ligase genes (Bej et al., 2020). In both rice and Arabidopsis, genes related to acquisition, uptake and transport of Fe through both Strategies I and II have been identified and annotated. In Fe deficiency condition, six candidate genes have been associated with Fe in maize and these genes are associated in various aspects of Fe homeostasis, including Fe(III)phytosiderophore transporter, Fe transport to vacuoles and transcriptional factors that regulate Fe-related gene expression (Curie et al., 2001; Kobayashi et al., 2014).

Long-distance Fe transport

Following the Fe transport from the rhizosphere into the root symplast, Fe is needed for chelating compounds. Then the Fe-chelator complexes are transported into the stele, following a diffusion gradient across intercellular connections. At this stage, Fe efflux is required to release Fe into the xylem vessels within the apoplastic space. However, the exact pathway of Fe efflux is not yet fully understood. In plants, especially Arabidopsis, three transporter proteins, known as Feregulated transporters (*IREGs*) or ferroportins (*FPTs*) are localized to the root epidermal cells. It is predicted that these transporters are involved in Fe-dependent nickel detoxification (Schaaf et al., 2006). AtIREG1/ FPT1 is bound to the plasma membrane of stele cells, indicating a potential role in releasing Fe into the xylem tracheas (Kim and Guerinot, 2007). FRD3 is a



citrate efflux long-distance Fe transporter associated to MATE family. MATE gene is expressed in the root pericycle and vascular cylinder, indicating its role in citrate efflux into the root pericycle and xylem vessels. However, in the xylem, the concentration of Fe is reduced and it accumulates in the shoot apoplast due to FRD3's involvement in bypassing longdistance Fe transport. This apoplastic movement of Fe, transfers it from cell to cell through intercellular spaces or walls, allowing Fe to move from the roots to the shoots and from the xylem to the phloem. This may compensate for Fe transport mediated by the xylem (Green and Rogers, 2004). Several genes play a role in the mechanism of Fe uptake from the xylem vessels into the plasma membrane of leaf cells. FRO and ZIP genes are expressed in shoots and the basal part of flowers, signifying their role in Fe uptake in aerial tissues (Vert et al., 2002). The mechanism of Fe transport through the phloem is also noteworthy, as it provides a feasible means of Fe transport, particularly when the Fe levels are insufficient in developing tissues/organs viz., apices, seeds and root tips if relying solely on xylem vessels. In phloem sap, the alkaline pH (>7) is favourable for maintaining Fe and Fe chelates in a soluble form. Phloem transport is also involved in the remobilization of Fe from older to younger leaves, where the alkaline pH in the phloem sap facilitates the binding of Fe to chelators to keep it soluble (Kim and Guerinot, 2007).

Chloronera (*chln*) is a mutant tomato that exhibits the role of nicotianamine in long-distance Fe transport. The *chln* gene encodes NAS in the mutant tomato and illustrates the role of nicotianamine in the transport of Fe over long distances. The *chln* gene was recognized due to the interveinal leaf chlorosis it caused in young leaves, although it led to increased Fe accumulation in roots. This phenomenon suggests that nicotianamine can act as a shuttle, chelating Fe²⁺ from Fe(III)-DMA during phloem loading and unloading, facilitating Fe²⁺/Fe³⁺ transformation and specific Fe(II)-NA

transport within the phloem. OsYSL1 transporters, similar to maize YS1, play a key role in the transport of Fe (III)-PS and Fe (II)-NA complexes. In rice total 18 putative YSL genes are identified in its genome and OsYSL2 is essential gene for transporting Fe(II)-NA and Mn(II)-NA (Koike et al., 2004). The temporal and spatial expression of YSL family genes indicating their role in Fe uptake mechanisms. The expression of AtYSL1 mRNA is increased in the vasculature of roots and shoots, specifically in the xylem tubes and is detected in young siliques and the chalazal zone of the embryo, indicating the role of YSLs in Fe loading of seeds. AtYSL1 and AtYSL3 shows a similar expression pattern in the vasculature of shoots and reproductive organs (Takahashi et al., 2003). Over all in Strategy II plants, the YSL genes are important in the long-distance Fe transport mechanism. In maize, the YS1 gene is expressed in both roots and shoots (Curie et al., 2001). Several OsYSL genes viz., OsYSL2 and OsYSL13 being preferentially expressed in shoots particularly OsYSL6, OsYSL14 and OsYSL16 are over expressed in both roots and shoots (Koike et al., 2004). OsYSL2 is a crucial gene overexpressed in the vascular bundles of the panicle neck and the sieve element cells of the phloem in flowers and developing seeds.

Members of the NRAMP family genes are intermediaries in the uptake of divalent cations (Thomine et al., 2003) and in mutant yeast, AtNRAMP4 can complement the Fe uptake, indicating their role in Fe transport (Thomine et al., 2003). In roots, NRAMP1 is expressed to take up Fe from the soil and is induced by Fe deficiency. Under Fe-deficient conditions, NRAMP1 targets the intracellular membrane and remobilizing the Fe into the cytosol (Thomine et al., 2000). In tomato, NRAMP1 and NRAMP3 genes are localized to vacuolar, intracellular vesicle and plasma membrane (Bereczky et al., 2003). In Arabidopsis, two NRAMP genes viz., AtNRAMP3 and AtNRAMP4 are localized to the vacuolar membrane (Lanquar et al., 2005). The NRAMP1, NRAMP3 and NRAMP4



genes are over expressed in response to Fe deficiency. When the NRAMP3 gene is overexpressed in plants, the over-expression of Fe uptake genes, viz., FRO2 and IRT1, is downregulated, indicating that NRAMP3 remobilizes the vacuolar Fe into the cytosol (Thomine et al., 2003). Fe is stored as ferritin in the plastid stroma of plant cells. Ferritin is a Fe storage protein capable of storing up to 4,500 Fe atoms. Arabidopsis has four genes (AtFer1-4) encoding ferritin. The transcript of AtFer1, 3 and 4 is expressed upon excess Fe treatment in both roots and leaves (Petit et al., 2001). However, despite the abundance of Fe, the mechanism of Fe uptake into chloroplasts is not well understood. Studies of Fe uptake with isolated chloroplasts have suggested that the mechanism is light dependent and requires Fe (III) chelate reductase activity in barley (Bughio et al., 1997).

Contribution of environmental factors

When the soils are aerobic or of higher pH, Fe is oxidized and fixed as insoluble ferric oxides and at the same time, as Fe is highly reactive, when it is present in excess it becomes toxic. Therefore, plants have developed a control system for Fe (Morrissey and Guerinot, 2009; Grillet *et al.*, 2014). Application of animal manure and plant residues modifies properties of the soils and reported to increase Fe and Zn availability, however foliar applications found to be more than soil nutrient application in increasing grain nutrient contents (Wei *et al.*, 2012; Prasad *et al.*, 2014; Velu *et al.*, 2015).

Bioavailability

Research on bioavailability of nutrients has received greater attention in the past decades. For improvement of Fe absorption, a ratio of phytic acid (PA): iron (Fe), <1:1, is requisite without any enhancers (Hurrell and Egli, 2010). Developing the biofortified crop with high nutritive content is not the only concern but also the bioavailability of the nutrients in human gut (Neeraja *et al.*, 2017). Antinutrient *viz.*, phytic acid inhibit absorption of minerals and hinder

the bioavailability of Fe from the ingested food (Kumar et al., 2017). Phytic acid represents 80% of the phosphorous in plants (Bohn et al., 2008). However, variation in the phytic acid content is not attributed to the Fe bioavailability. In maize, by expressing a fungal phytase, 3-fold increase in bioavailable Fe and decrease in phytic acid is reported (Drakakaki et al., 2005). Fe bioavailability is measured by in vitro methods viz., Caco-2 cell model. The caco-2 cells are the colonic carcinoma cells that are morphologically and functionally similar to the epithelial cells lining the small intestine. Animal studies using rodent models have been used in bioavailability studies on Zn and carotenoids, but this model seems to be a poor choice to assess Fe bioavailability. High Fe bioavailability in biofortified food crops was observed using isotopic human studies, however they are time consuming and very expensive, which has limited their use (La Frano et al., 2014). Around 23 articles evaluated the bioavailability in biofortified crops, of which eight were animal studies, seven were in vitro studies and eight were human studies. A combination of these in vitro, animal and human studies will be an effective approach for investigating the efficacy of biofortification programmes (Dias et al., 2018).

Caco-2 cell bioassay was reported as the best approach to evaluate the nutritive quality of Fe biofortified beans (Phaseolus vulgaris) and these varieties have high absorption than normal bean variety. Caco-2 cell model can also disclose the effects of antinutrients like phytic acid. Fe absorption studies in 61 Rwandese women with low Fe status revealed no significant difference in the Fe absorption from Fe rich beans than normal beans, which might be due to high phytic acid and polyphenols in beans (Petry et al., 2012). In Pea, phytic acid decrease by 60% has increased bioavailability of Fe in Caco-2 cell studies and improvement of Fe bioavailability by 50-100% was identified in *lpa* lines than in controls (Warkentin et al., 2012; Liu et al., 2015). Cognitive performance, especially the efficiency of search and the speed



of retrieval on memory tasks, was improved in 18-25 women tested by consuming the Fe biofortified beans (86.1 ppm) compared with the normal beans (Murray-Kolb et al., 2017). High ferritin formation in the Caco-2 cells with digests having FeSO4 and ascorbic acid than the digests with FeSO₄ and citric acid was reported by (Glahn et al., 1998). The effects of Fe status by consuming the Fe-biofortified rice was tested in 191 women in Philippines which resulted in increase of Fe stores in the women (Haas et al., 2005). Meta-analysis on Fe bioavailability in different types of millets, (in vitro and in vivo) showed variation in the Fe levels ranging from 2 to 8 mg/100 g and 13.2% significant increase in haemoglobin levels. Enhancement of Fe bioavailability by 3.4 to 2.2 times is noted in women by following traditional methods like fermentation and germination (Anitha et al., 2021). Randomised efficacy trials in the Fe profiles like serum ferritin, soluble transferrin receptor, total body Fe etc. were conducted in the Philippines, India and Rwanda in different crops like rice, pearl millet and beans. Cognitive performance in attention and memory domains were significantly improved by Fe biofortified crops compared with conventional crops (Finkelstein et al., 2019). When the Indian school children, between 12-16 years of age, fed with Fe biofortified pearl millet continuously for six months, showed increased light physical activity and decreased sedentary time in children (Pompano et al., 2022). Decrease in pathogenic bacteria and increase in beneficial bacteria in the gut is reported by dietary intake of Fe biofortified foods (Gomes et al., 2021). The bioavailability of Fe can be enhanced from 12.1 to 16.4 ppm by following different processing techniques like popping, malting etc. (Neeraja et al., 2017). The Fe bioavailability can also be altered by different methods of cooking and digestion in the intestine. During heating, Fe²⁺ or Fe³⁺ are released from Fe (III) hydroxides in ferritin (Hoppler et al., 2008) and in the food matrix they are chelated by phytic acid (Moore et al., 2018; Perfecto et al., 2018). Since, the primary inhibitors of Fe bioavailability in food

crops are phytic acid and polyphenolic compounds, breeding low phytate genotypes is now being targeted in different crops.

Conclusion

With more than half of the world's women and children suffering from Fe deficiency, all the available strategies should be adopted for improving Fe status in food grains. Biofortification is one of the proven approaches for enhancing Fe content in cereals such as pearl millet and in other crops such as beans. Limited genetic variability for grain Fe content is a major constraint in cereals, thus extensive germplasm screening should be done. High throughput sequencing of germplasm accessions, genome wide association mapping and RNA seq analyses could lead to the candidate genes associated with high grain Fe content. Genome editing offers favorable opportunities to increase the grain Fe content by modifying known candidate genes for Fe metabolism. Converging the physiological and molecular mechanisms of Fe transport and translocation along with methods to improve bioavailability could pave way to the success Fe Biofortification targeting nutritional security.

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RESEARCH ARTICLE

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Genetic Variability for Yield Components, Iron and Zinc contents in Rice (Oryza sativa L.) Genotypes

Tukaram J Bhor1*, Rohit K Jambhulkar2 and Narendra V Kashid1

¹Agricultural Research Station, Vadgaon Maval, Pune-412106, Maharashtra ²College of Agriculture, Pune-411005, Maharashtra *Corresponding author Email: tjbhor1969@gmail.com

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Abstract

The experiment was carried out for 11 characters in 41 rice genotypes during *kharif* 2022 at Agricultural Research Station, Vadgaon Maval, Pune. The highest value of GCV (32.111) and PCV (32.618) observed for grain yield per plant and followed by grain iron (Fe) content, GCV (31.137) and PCV (32.021), grain zinc (Zn) content GCV (27.039) and PCV (28.147), fertile spikelets per panicle GCV (20.462) and PCV (21.232), 1000 grain weight GCV (19.104) and PCV (20.094). The grain yield per plant was observed as the highest broad sense heritability (96.90%) followed by days to 50% flowering (95.60%) and grain iron (Fe) content (94.60%). The highest genetic advance as per cent of mean was observed for the traits grain yield per plant (65.121), grain iron content (62.373), grain zinc content (53.509) and fertile spikelets per panicle (40.625). The significant positive correlation was observed among grain yield per plant and days to 50% flowering (0.3171), panicle length (0.2974), fertile spikelets per panicle (0.5101), total spikelets per panicle (0.4519), spikelet fertility (0.4570), test weight (0.4378) and productive tillers per plant (0.5732). High heritability with high genetic advance observed for the trait grain yield per plant, grain iron (Fe) content, grain zinc (Zn) content, fertile spikelets per panicle and total spikelets per panicle indicated additive gene actions. The total spikelet per panicle (2.9940) was observed that had highest direct positive effect on grain yield per plant and followed by spikelet fertility % (0.7118), test weight (0.6805), productive tillers per plant (0.5068) and panicle length (0.1516).

Key words: Variability, heritability, genetic advance, correlation, direct effect

Introduction

The *Oryza sativa* (rice) species are commonly divided in to three subspecies namely; *Indica*, *Japonica* and *Javanica*. Rice (*Oryza sativa* L.) is one of the three leading cereals in the family Poaceae. It is cultivated during warm wet (*kharif*) and winter (*rabi*) seasons in the tropical and subtropical humid regions of the world. Rice is the staple food for a majority of the people of Asia, providing 50-60 per cent of the total calorie and 30 per cent of the total protein (Mackill *et al.*, 2012).

According to the FAO (2003) and ICMR (2009), the recommended daily allowance (RDA) for zinc and iron is 10 mg for children and 12-15 mg for adults. Due to their effects on growth and development, as

well as other physiological and neurophysiological processes, these micronutrients are important for health and medicine. According to estimates, 49 per cent of people worldwide fail to consume sufficient amounts of zinc. Micronutrients are necessary for both plants and animals to have a balanced diet. Additionally, micronutrients are essential for rice plants' ability to withstand abiotic stresses. From an agronomic perspective, zinc is important to rice for a number or reasons such as nitrogen assimilation and protein metabolism. Approximately 10% of proteins in plants require Zn for structural function and integrity. Low Zn supply limits the rice plant's ability to



convert amino acids to proteins. Iron (Fe) participates in several important metabolic processes such as photosynthesis, chloroplast development, chlorophyll biosynthesis, electron transport and redox reactions. The focus of the green revolution on increasing grain productivity resulted in a decrease in the amount of micronutrients available in a balanced diet. Most of the Iron and Zinc is found in rice bran-aleuronic layer and 80% of it gets removed in polishing. Hence it is essential to have genotypes with high Iron and Zinc content in endosperm.

The increase in rice production is crucial for providing food security. Understanding genetic variability in the experimental material offers a useful opportunity for selection (Singh et al., 2020). Phenotypic variability is the differences between individuals in a population due to genetic composition and growing environment (Sumanth et al., 2017). Planning and execution of any breeding program for improvement on quantitative traits depends on magnitude of genetic variability. Therefore, success on plant breeding activities entirely depends on the existence of genetic variability with respect to desired traits and selection skill of plant breeder (Adhikari et al., 2018). Variability, genetic diversity, expected genetic advances and heritability of the traits are therefore key basis for genetic improvement of the trait.

The level of heritable variation in the traits studied is extremely useful in determining the genotype's potential for future breeding programs. Before planning an appropriate breeding strategy for genetic improvement, it is critical to assess variability for yield and its component characters (Singh *et al.*, 2011). For efficient crop development, understanding the nature and extent of genetic variation in quantitative traits such as yield and its components is critical. Heritability is the ratio of variation due to differences between genotypes to the total phenotypic variation for a trait in a population and shows the component of a character transmitted to future generations. Genetic

advance shows the difference between the mean genotypic values of selected population and the original population from which these were selected. Heritability estimates along with genetic advance is more precise in predicting the genetic gain under selection than heritability alone. In addition relationship between yield and yield attributing traits is prime important for direct and indirect selection of traits which contributes to yield (Aditya and Bhartiya, 2013). Although there are thousands of distinguished genotypes in cultivation, the diverse material available will gives the scope to evolve the cultivars which are having potential for biotic and abiotic stresses along with good quality characters (Khan, 2018). The selected material from evaluations can be utilised for further hybridisation programme. Choosing high-yielding varieties purely on the basis of grain yield will be ineffective unless sufficient data on genetic characteristics is available to use them in hybridization programmes for the further improvement. In the selection process, information on character association, as well as the direct and indirect effects that each character has on yield, will be beneficial. The degree of the association between grain yield and its components, as well as the relative relevance of their direct and indirect effects, are revealed by correlation and path analysis, offering a comprehensive knowledge of their relationship with grain yield (Khan, 2018). About 176 million tons rice is required by 2035, which can be fulfilled by increasing yield potential from 10 to 12.3 tons per hectare (Khush, 2013).

Therefore, the present study was under taken to know information on variability, heritability, genetic advance and traits correlation in promising rice genotypes to develop a high yielding rice genotype. Information on genetic variability is essential for selection, which is the ultimate tool in plant breeding. However, the selection is challenging due to the masking effect of non-heritable variation. Therefore, partitioning of the total variation to understand the



role of the heritable component is very important, which helps the breeder to formulate a sound breeding programme. Correlation is very important factor for any election program. Success in selection depends on the knowledge of the association of component traits with grain yield. However, partitioning the correlation coefficients through path analysis is very important to understand the cause-effect relationship. This helps in understanding the relative importance of component characters for improvement in seed yield of rice. Finally, this type of research could aid the breeder in developing selection strategies to improve grain yield. Given the above scenario, the current investigation is carried out with the objectives of studying the genetic variability, character association and path coefficients in rice genotypes for yield improvement.

Materials and Methods

The present investigation was carried out during *kharif* 2022 at Agricultural Research Station, Vadgaon Maval, Pune. In the present study, forty-one genotypes were assessed to study the variability and genetic parameters for yield and its components, iron and zinc content. List of genotypes used in the present study is presented in **Table 1.** Each genotype was sown in three rows of 4 m length following a spacing of 20 cm between the rows and 15 cm between the plants in randomized block design (RBD) with three replications. Standard agronomic practices were performed uniformly for all the experimental units. Crop was raised following recommended package of practices. Phenological data

for days to 50% flowering was recorded on plots basis for each genotype in each replication. At maturity five plants from each accession were selected randomly for recording data on grain yield per plant and yield component traits, namely, plant height, productive tillers per plant, panicle length, grains per panicle and test weight. In contrast, observations for test weight and iron and zinc content traits were obtained from a random grain sample drawn from each plot and replication using standard procedures. The statistical analysis was done by standard statistical method suggested by Panse and Sukhatme (1995). Mean performance of the genotypes were calculated and the genotypic (GCV) and phenotypic (PCV) coefficients of variation were estimated by using the formula given by Burton (1952). The estimates of PCV and GCV were classified as low (0-10%), moderate (10-20%) and high (>20%) according to Sivasubramanian and Madhavamenon (1973). Heritability in broad sense (h² b) was estimated according to the formula suggested by Johnson et al., (1955) and Hanson et al., (1956) and classified as low (0-30%), moderate (30-60%) and high (> 60%). Estimation of genetic advance was carried out following the formula given by Johnson et al., (1955) and classified as low (< 10%), moderate (10-20%) and high (> 20%). The genotypic correlation coefficients were worked out by adopting the method described by Singh and Chaudhary (1977). Path coefficient analysis was done according to the procedure suggested by Dewey and Lu (1959).

Table 1: List of Rice genotypes used in the study

Sl. No	Entry	Source	Sl. No	Entry	Source	Sl. No	Entry	Source	Sl. No	Entry	Source
1	GNV 1905	AICRP	11	CSR HZR	AICRP	21	BRR-0184-1-IR	AICRP	31	PDKV Red	ARS, Sakoli
		Trial 2021		17-42	Trial 2021		108194-9-1-2-1	Trial 2021		Rice-1	
2	BPT 5204	AICRP	12	HURS 21-7-	AICRP	22	NVSR 3148	AICRP	32	Phule Samruddhi	ARS, Vadgaon
		Trial 2021		IR 105696-1-	Trial 2021			Trial 2021			Maval
				2-3-1-1-B							
3	GNV 1906	AICRP	13	NVSR 526	AICRP	23	RP5401-	AICRP	33	Indrayani	ARS, Vadgaon
		Trial 2021			Trial 2021		JBB-B-622-3-1-1-1-1	Trial 2021			Maval
4	RP 6362-IR15M1298	AICRP	14	RP 6195-MC/	AICRP	24	CR 4365-1-IR	AICRP	34	Pawana	ARS, Vadgaon
	(GID:4289666)	Trial 2021		RIL-A147	Trial 2021		128768-7-2-2-5	Trial 2021			Maval



Sl. No	Entry	Source	Sl. No	Entry	Source	Sl. No	Entry	Source	Sl. No	Entry	Source
5	DRR Dhan 45	AICRP	15	HURS21-6-IR	AICRP	25	RP 6204-MB/	AICRP	35	Phule Maval	ARS, Vadgaon
		Trial 2021		08195-3-1-1-2	Trial 2021		RIL-J65	Trial 2021			Maval
6	IR-64	AICRP	16	RP 6204-MB/	AICRP	26	RP 6196-PC/	AICRP	36	Kundalika	ARS, Vadgaon
		Trial 2021		RIL-J159	Trial 2021		RIL-B162	Trial 2021			Maval
7	Chittimutyalu	AICRP	17	R-RHZ-IA-99	AICRP	27	RP 6253-MV/RIL-	AICRP	37	Bhogawati	ARS,
		Trial 2021			Trial 2021		MV 208	Trial 2021			Radhanagari
8	HURS 17-6-IR	AICRP	18	CSR HZR	AICRP	28	RP6514-	AICRP	38	VDN-1832	ARS, Vadgaon
	82475-110-2-2-1-2	Trial 2021		23-1	Trial 2021		IR128768-7-2-2-4	Trial 2021			Maval
9	NVSR 522	AICRP	19	CSR HZR	AICRP	29	RP 4993-BC/	AICRP	39	Phule Radha	ARS,
		Trial 2021		17-8	Trial 2021		RIL-Z102	Trial 2021			Radhanagari
10	RP 6211-PR/	AICRP	20	HURS-21-3-	AICRP	30	Karjat-4	RARS,	40	Ambemohar-157	ARS, Vadgaon
	RIL-Q181	Trial 2021		IR128773-4-4-	Trial 2021			Karjat			
				2-3-B							
									41	IGP-13-12-19	ZARS, Igatpuri

Results and Discussions

The amount of genetic diversity in the crop is measured by the genotypic coefficient of variation (GCV) as it represents the heritable component of variability. Additionally, the difference between genotypic and phenotypic expression gives the role of environmental in expression of that particular trait. The genotypic differences were significant for almost all the traits studied indicated that there was considerable amount of variability present in genotypes (Table 2). The estimates of genotypic coefficient of variation, range of variability and phenotypic coefficients of variation, the per centage of heritability in a broad sense and the genetic advance reported as a per centage of mean are given in

Table 3. For each of the traits examined in the current analysis a significant range of variance was observed. In forty-one (41) genotypes, the total spikelets per panicle ranged from 109.00-221.50 followed by fertile spikelets per panicle (89.00-201.00), plant height (75.70-172.60 cm), grain zinc content (22.45-68.40 ppm), grain iron content (20.70-66.35 ppm), days to 50% flowering (82.00-116.50), grain yield per plant (9.95-33.50 g), spikelet fertility % (80.10-95.85), 1000 grain weight (11.25-23.65 g), panicle length (17.05-28.75 cm) and productive tillers per plant (6.25-9.85). This indicated that there was a large amount of variation for these traits (Maurya *et al.*, 2022 and Divya *et al.*, 2018).

Table 2: Analysis of variance for eleven different characters

Sl.	Characters		Mean sum of square	
No.	Characters	Replication (d.f.1)	Treatments (d.f.40)	Error (d.f.40)
1.	Days to 50% flowering	18.56	114.97**	2.57
2.	Plant height	1.97	525.29**	25.56
3.	Panicle length	2.74	13.87**	0.64
4.	Productive tillers/plant	1.19	2.09**	0.31
5.	Fertile spikelets/panicle	1705.80	1612.29**	59.50
6.	Total spikelets/panicle	1615.80	1798.75**	82.25
7.	Spikelet fertility %	16.70	33.83**	3.31
8.	1000 grain weight	1.01	28.53**	1.44
9.	Grain yield/plant	0.66	66.23**	1.04
10.	Grain iron (Fe) content	15.28	280.38**	7.84
11.	Grain zinc (Zn) content	2.70	326.48**	13.10

^{*}Significant at 5% level; ** Significant at 1% level



For all characters, it was observed that the values for genotypic coefficients of variation (GCV) were lower than phenotypic coefficients of variation (PCV) (Table 3) indicated that these traits were considerably influenced by environments. The trait spikelet fertility exhibited the lowest GCV (4.474) as well as PCV (4.935), whereas grain yield per plant exhibited the highest GCV (32.111) and PCV (32.618). The broad difference between GCV and PCV were observed for productive tillers per plant indicated that the trait influenced more by environment whereas, days to 50% flowering, spikelet fertility and grain yield per plant recorded

narrow difference that indicated less influence of environment. High GCV and PCV were observed for grain yield per plant followed by grain iron content, grain zinc content and fertile spikelets per panicle. Moderate GCV and PCV were observed for 1000 grain weight, total spikelets per panicle, plant height, productive tillers per plant and panicle length. The trait days to 50% flowering recorded low GCV and PCV. Roy *et al.*, (2021) and Maurya *et al.*, (2022) also reported high GCV and PCV for grain yield per plant and moderate GCV and PCV for 1000 grain weight, total spikelets per panicle, plant height and productive tillers per plant.

Table 3: Estimates of genetic variability parameters for 11 different characters of rice genotypes

Sl. No.	Name of the Character	Range	Mean	G.C.V. (%)	P.C.V. (%)	Heritability (h ²) (bs) %	GA as % of mean (at 5% K)
1.	Days to 50% flowering	82.00-116.50	96.50	7.76	7.94	95.60	15.64
2.	Plant height (cm)	75.70-172.60	100.02	15.80	16.59	90.70	31.00
3.	Panicle length (cm)	17.05-28.75	22.64	11.35	11.89	91.20	22.34
4.	Productive tillers/plant	6.25-9.85	7.69	12.28	14.26	74.10	21.78
5.	Fertile spikelets/panicle	89.00-201.00	136.17	20.46	21.23	92.90	40.62
6.	Total spikelets/panicle	109.00-221.50	155.59	18.82	19.71	91.30	37.05
7.	Spikelet fertility %	80.10-95.85	87.32	4.47	4.93	82.20	8.35
8.	1000 grain weight (g)	11.25-23.65	19.26	19.10	20.09	90.40	37.41
9.	Grain yield/plant (g)	9.95-33.50	17.78	32.11	32.61	96.90	65.12
10.	Grain iron (Fe) content (ppm)	20.70-66.35	37.49	31.13	32.02	94.60	62.37
11.	Grain zinc (Zn) content (ppm)	22.45-68.40	46.29	27.03	28.14	92.30	53.50

Genetic advance and heritability are recognised as important selection parameters. According to Burton (1952), combining genetic advance with heritability estimates, would provide a better idea for effective selection. Heritability is a reliable indicator that shows the transmission of parental values to their offspring. The plant breeder can pick superior traits from diverse genetic groups using the heritability estimates. The high heritability values were recorded for grain yield per plant (96.90%) followed by days to 50% flowering (95.60%), grain iron (Fe) content (94.60%), fertile

spikelets per panicle (92.90%), grain zinc (Zn) content (92.30%), total spikelets per panicle (91.30%), panicle length (91.20%), plant height (90.70%), 1000 grain weight (90.40%), spikelet fertility % (82.20%) and productive tillers per plant (74.10%) character. High heritability estimates suggested that these characters were least influenced by the environment. Similar results were obtained by Divya *et al.*, (2018) for the characters number of productive tillers per plant, 1000 grain weight, number of grains per panicle and grain yield per plant and Shaili *et al.*, (2022) for grain



yield per plant, number of panicles, test weight and plant height.

The highest genetic advance (GA) as per cent of mean was observed for the trait grain yield per plant (65.121) followed by grain iron (Fe) content (62.373), grain zinc (Zn) content (53.509), fertile spikelets per panicle (40.625), 1000 grain weight (37.418), total spikelets per panicle (37.054) and plant height (31.009). The moderate genetic advance (GA) as per cent of mean was exhibited by the traits such as panicle length (22.342), productive tillers per plant (21.787) and days to 50% flowering (15.649). The low values were recorded by the trait spikelet fertility % (8.354). High heritability (h²) with moderate genetic advance as per cent of mean showed for the characters fertile spikelets per panicle, total spikelets per panicle, 1000 grain weight and plant height suggested that both additive and nonadditive gene effects were involved in the genetic regulation of these traits. High heritability (h²) along with higher genetic advance (GA) as per cent of mean was recorded for the characters viz., grain yield per plant, grain iron (Fe) content, grain zinc (Zn) content, fertile spikelets per panicle, 1000 grain weight, total spikelets per panicle, plant height and days to 50% flowering indicated that these traits were under control of additive gene action. The selection could be practised for these traits for improvement of genotype. Panigrahi *et al.*, (2018) reported additive gene action for 1000 grain weight and Shaili *et al.*, (2022) for test weight, grain yield and plant height.

Correlation studies

The genotypic correlation coefficient values between yield and its related characters are presented in Table 4. The highest and significant positive correlation of grain yield per plant was found with productive tillers per plant (0.5732) followed by fertile spikelets per panicle (0.5101), spikelet fertility (0.4570), total spikelets per panicle (0.4519), test weight (0.4378), days to 50% flowering (0.3171) and panicle length (0.2974). Plant height (0.1903) showed nonsignificant positive correlation with grain yield per plant. When making selections to improve yield, it is recommended that these characters be given top priority. Similar type of associations were reported earlier by Pachauri et al., (2017) for days to 50% flowering, panicle length and number of productive tillers per plant. All the studied characters had a positive correlation with grain yield per plant.

Table 4: Genotypic correlation coefficients of 8 characters of 41 genotypes of rice on grain yield

	Days to	Plant	Panicle	Productive	Fertile	Total	Spikelet	Test	Grain
Sl.	50%	height	Length	tillers per	spikelets	spikelets	Fertility	weight	yield per
No.	flowering	(cm)	(cm)	plant	per panicle	per panicle	%	(g)	plant (g)
	1	2	3	4	5	6	7	8	9
1.	1.0000	0.3488**	0.1818	0.0555	0.5392**	0.4892**	0.4603**	-0.1501	0.3171**
2.		1.0000	0.4913**	-0.0603	0.2235*	0.2067	0.1771	0.0583	0.1903
3.			1.0000	-0.0471	-0.0523	-0.0697	0.0003	0.4095**	0.2974**
4.				1.0000	0.1982	0.1770	0.1280	-0.0642	0.5732**
5.					1.0000	0.9811**	0.4734**	-0.4045	0.5101**
6.						1.0000	0.2974**	-0.4441	0.4519**
7.							1.0000	0.0336	0.4570**
8.								1.0000	0.4378**
9.									1.0000

^{*, **} Significant at 5(0.2172) and 1(0.2829) per cent, respectively



Number of productive tillers per plant exhibit positive association with grain yield per plant, panicle length and number of productive tillers per plant, also exhibited high direct effect on yield. This result was in conformity with the earlier findings of Ramanjaneyulu et al., (2014). Correlation analysis of yield contributing characters shows that all the characters under study were significantly and positively correlated with grain yield per plant except, plant height. These results were agreement with Iqbal et al., (2018). Kamana et al., (2019) reported positive direct effect of spikelet fertility, days to 50% flowering, plant height and 1000 grain weight on grain yield.

Path analysis: The path coefficient evaluation using genotypic correlation indicates the interrelationships between the characters, which are illustrated in **Table 5.** The character total spikelet per panicle (2.9940) showed highest direct positive effect on

grain yield per plant followed by spikelet fertility % (0.7118), test weight (0.6805), productive tillers per plant (0.5068) and panicle length (0.1516). Hence, direct selection for these characters will be beneficial for yield improvement programme. The characters days to 50% flowering (-0.0340), plant height (-0.0563) and fertile spikelet per panicle (-2.5506) observed negative direct effect on grain yield per plant. Similar types of findings were reported by Singh and Ekka (2019). Days to 50 per cent flowering showed positive correlation with grain yield per plant (0.3171) but it showed negative indirect effect with plant height (-0.0119), panicle length (-0.0062), productive tillers per plant (-0.0019), fertile spikelet per panicle (-0.0183), total spikelet per panicle (-0.0166) and spikelet fertility % (-0.0157). Days to 50% flowering showed positive indirect effect with test weight (0.0051).

Table 5: Direct (diagonal) and indirect (above and below diagonal) path effects of different characters towards grain yield at genotypic level in rice

CI	Days to	Plant	Panicle	Productive	Fertile	Total	Spikelet	Test	Grain
Sl. No.	50%	Height	length	tillers per	spikelet	spikeletper	Fertility	Weight	Yield per
110.	flowering	(cm)	(cm)	plant	per panicle	panicle	%	(g)	plant (g)
	1	2	3	4	5	6	7	8	9
1.	-0.0340	-0.0119	-0.0062	-0.0019	-0.0183	-0.0166	-0.0157	0.0051	0.3171**
2.	-0.0196	-0.0563	-0.0277	0.0034	-0.0126	-0.0116	-0.0100	-0.0033	0.1903
3.	0.0276	0.0745	0.1516	-0.0071	-0.0079	-0.0106	0.0001	0.0621	0.2974**
4.	0.0281	-0.0306	-0.0239	0.5068**	0.1004	0.0897	0.0649	-0.0326	0.5732**
5.	-1.3752	-0.5701	0.1334	-0.5054	-2.5506	-2.5024	-1.2075	1.0318	0.5101**
6.	1.4646**	0.6189**	-0.2087	0.5300**	2.9374**	2.9940**	0.8905**	-1.3297	0.4519**
7.	0.3277**	0.1261	0.0002	0.0911	0.3370**	0.2117	0.7118**	0.0239	0.4570**
8.	-0.1021	0.0397	0.2787*	-0.0437	-0.2753	-0.3022	0.0229	0.6805**	0.4378**

(Residual effect = 0.1035) *, ** Significant at 5 and 1 per cent, respectively

Productive tillers per plant showed significant positive correlation (0.5732) with grain yield, it also had the significantly positive direct effect (0.5068) on grain yield. The character influences the yield positively indirect effect with days to 50% flowering (0.0281), fertile spikelet per panicle (0.1004), total spikelet

per panicle (0.0897), spikelet fertility % (0.0649). It also had negative indirect effect on grain yield with plant height (-0.0306), panicle length (-0.0239) and test weight (-0.0326). Total spikelets per panicle had significant positive direct effect (2.9940) on grain yield per plant and indirect effect through days to



50% flowering (1.4646), plant height (0.6189), productive tillers per plant (0.5300), fertile spikelets per panicle (2.9374) and spikelet fertility (0.8905). It also observed that character had negative indirect effect like Panicle Length (- 0.2087) and Test weight (-1.3297). Test weight had positive direct effect (0.6805) on grain yield per plant and had positive significant correlation (0.4378) on grain yield. It had positive indirect effect through plant height (0.0397), panicle length (0.2787), spikelet fertility % (0.0229).

The residual effect determines how well the causal factors account for the variability of the dependent factors, in this case, the grain yield. The residual effect in the current study was relatively low (0.1035), indicating that the characters selected were sufficient for explaining variability in rice grain yield. The productive tillers per plant, total spikelet per panicle, spikelet fertility % and test weight identified as important components of rice grain yield. Similar type of association was recorded by Akinwale *et al.*, (2011) for productive tillers per plant and test weight and, Hasan *et al.*, (2011) for productive tillers per plant, spikelet fertility % and test weight and,

Conclusion

The trait grain yield per plant and grain iron content observed high magnitude of GCV and PCV this indicates that there is opportunity for improvement through selection. There was a high heritability with a high genetic advance as per cent of mean for the traits grain yield per plant, grain iron content, grain zinc content, fertile spikelets per panicle, 1000 grain weight, total spikelets per panicle, plant height and days to 50 per cent flowering indicated that these characters were primarily governed by additive gene action and selecting for these traits would be more effective in achieving desired genetic improvement. Grain yield per plant had significant and positive correlation with days to 50 per cent flowering, panicle length, fertile spikelets per panicle, total spikelets per panicle, spikelet fertility, test weight and productive

tillers per plant. In the path analysis, highest and significant positive direct effect on grain yield per plant was recorded through total spikelet per panicle followed by spikelet fertility per cent, test weight, productive tillers per plant and panicle length. The direct selection of these traits will be beneficial for yield improvement programmes.

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RESEARCH ARTICLE

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Genetic Variability Studies in Rice Genotypes Jyothsna B¹, Roja V ^{2*}, Krishnaveni B¹, Prasanna kumari V³ and Pranaya J¹

¹Department of Genetics and Plant Breeding, ANGRAU, Maruteru ²Department of Molecular Biology and Biotechnology, ANGRAU, Maruteru ³Department of Plant Pathology, Acharya N.G. Ranga Agricultural University *Corresponding author Email: v.roja@angrau.ac.in

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Abstract

Seventy-four rice genotypes were evaluated to assess their genetic variability, heritability and genetic advance as per cent of mean. The analysis of variance revealed significant differences for all the characters under the study except number of productive tillers per plant, indicating the presence of high genetic variability among the genotypes. The genotypes per se performance revealed BPT 3130, BPT 3164, BPT 3264, BPT 3092, BPT 3275, BPT 2824, BPT 2954, BPT 3276, BPT 3095 and BPT 3178 exhibiting significant superiority over the check Samba Mahsuri for the characters *viz.*, grain yield per plant, test weight and panicle length. The high estimates of GCV and PCV were observed for grain yield per plant. High heritability coupled with high expected genetic advance as per cent of mean was observed for test weight and grain yield per plant suggesting that selection for the improvement of these traits may be rewarding.

Keywords: Analysis of variance, GCV, PCV, heritability, genetic advance as per cent of mean

Introduction

Rice (Oryza sativa, 2n=2x=24) occupies a notable position among food grains by directly feeding the majority of people more than any other crop. The demand for rice production is increasing day by day because of the increase in the number of rice consumers. To meet this demand, it is necessary to enhance the production and productivity of rice cultivars. The success of any breeding programme depends on the degree of variability present in the germplasm. Before initiating any breeding programme, knowledge on the nature and magnitude of genetic variation governing the inheritance of characters and as well as adopting appropriate selection techniques is essential for genetic improvement. Genetic parameters such as Genotypic Coefficient of Variation (GCV) and Phenotypic Coefficient of Variation (PCV) are useful in detecting the amount of variability present in the population (Idris et al., 2012). It is very difficult to judge whether detected variability is heritable or not. Heritability indicates the extent of transmissibility of a character into future generations. Heritability can be either broad sense or narrow sense. Broad sense heritability is the relative magnitude of genotypic and phenotypic variances for the traits and has a predictive role in selection procedures (Allard, 1960). This gives an idea of total variation ascribed to genotypic effects, which are the exploitable portion of variation (Mba and Dixon, 1995). The magnitude of variation due to the heritable component is very important because it would be a guide for the selection of parents for crop improvement (Dutta et al., 2013) and also plays a vital role to plan an efficient breeding program for the genetic improvement of quantitative traits (Seyoum et al., 2012). The estimate of heritability alone is



not very much useful because it includes the effect of both additive and non-additive genes. The genetic advance is a useful indicator of the progress that can be expected as a result of exercising selection on the pertinent population (Vanniarajan *et al.*, 1996).

Identification of effective selection criteria is very important in any breeding programme for effective yield improvement. The present investigation was undertaken in this context to elucidate information on variability, heritability and genetic advance in rice genotypes for utilization in the further breeding programme.

Materials and Methods

The material for the present study consists of 74 rice genotypes including yield check Samba Mahsuri. These genotypes were evaluated in an augmented randomized complete block design during *kharif*, 2021 at Agricultural College Farm, Bapatla and Andhra Pradesh. Observations were recorded for the characters, plant height (cm), number of productive tillers per plant, panicle length (cm), spikelet fertility (%), test weight (g), grain yield per plant (g) and days to 50 per cent flowering.

Statistical analysis

The mean data of all the characters were subjected to analysis of variance (ANOVA) based on the model proposed by Federer (1956) using Windostat Version 9.3. Different genetic parameters such as genotypic and phenotypic coefficient of variation, heritability, genetic advance and genetic advance as per cent of mean were estimated by using the following formula.

Estimation of genotypic and phenotypic variances

The genotypic and phenotype variances were calculated as per the formulae proposed by Burton and Devane (1953).

Phenotypic standard deviation (
$$\sigma p$$
) = $\sqrt{\sigma_p^2} = \sqrt{\sigma_g^2} + \sqrt{\sigma_e^2}$

GCV=
$$\frac{\text{Genotypic standard deviation } (\sigma g)}{(\text{General mean } (x \ \overline{)})} \times 100$$

PCV=
$$\frac{\text{Phenotypic standard deviation } (\sigma p)}{(\text{General mean } (x \))} \times 100$$

GCV and PCV values were categorized as low (<10%), moderate (10-20%) and high (>20%) (Subramaniam and Menon, 1973).

Estimation of heritability (broad sense)

Heritability in broad sense (h²) was computed and categorized as per the suggestions of Hanson *et al.*, (1956) and Johnson *et al.*, (1955) respectively.

GCV=
$$\frac{\text{Genotypic variance } (\sigma 2g)}{\text{(Phenotypic variance } (\sigma 2p)} \times 100$$

Heritability was classified as low (below 30%), medium (30-60%) and high (above 60%) as suggested by Johnson *et al.*, (1955).

Estimation of genetic advance

From the heritability estimates, the genetic advance (GA) was calculated by the following formula given by Lush (1940) and Johnson *et al.*, (1955).

Genetic Advance (GA) =
$$K \times \sigma_p \times h_{(b)}^2$$

Where,

K=Selection differential at 5% selection intensity which accounts to a constant value of 2.06

 $h_{(b)}^2$ = Heritability in broad sense.

 σ_{p} = Phenotypic standard deviation.

Estimation of Genetic advance as per cent of mean (GAM)

Genetic advance as per cent of mean was calculated as per the formula

$$GAM = \frac{Genetic advance}{General mean (x)} X 100$$

The degree of genetic advance as per cent of mean was classified as suggested by Johnson *et al.*, (1955) as low (<10%), moderate (10-20%) and high (>20%).



Results and Discussions

Analysis of variance

The ANOVA results indicated significant variability among the genotypes for all the characters studied except for the trait number of productive tillers per plant. Similar results were reported by Chandramohan *et al.*, (2016). Analysis of variance for the yield and yield component traits was furnished in **Table 1**.

Table 1: Analysis of variance for yield and yield component traits

Sources of	d. f	DFF	PH	NPT	PL	SF	TW	GYP			
variation	u. 1		Mean sum of squares								
Blocks	4	3.576	20.401	1.175	0.376	4.09	0.066	1.591			
Entries	73	94.345 **	143.158 **	4.296	4.262 **	26.372 **	9.456 **	45.571 **			
Genotypes	69	49.023 **	82.952 **	3.962	3.952 **	24.139 **	5.014 **	45.524 **			
Checks	3	797.333 **	1138.847 **	6.467	3.729 *	47.576 **	110.531 **	53.010 **			
Checks vs	1	1112.584 **	1310.292 **	20.829 *	27.280 **	116.870 **	12.714 **	26.518 *			
Genotypes	1	1112.364	1310.292	20.829	27.280	110.870	12./14	20.318			
Error	12	3.375	8.573	2.342	1.05	4.944	0.089	3.421			

^{*}Significant at 5% level; **Significant at 1% level; DFF: Days to 50% flowering; PH: Plant hieght; NPT No. of productive tillers; PL: Panicle length; SF: Spikelet fertility %; TW: Test weight; GYP: Grain yield for plot.

Mean performance

Mean values of the seventy-four genotypes along with the standard yield check Samba Mahsuri for the studied seven quantitative characters were presented in **Table 2**. Genotype TN1 was found earliest in flowering (85 days) suggesting that this genotype can be used as a donor in the hybridization programme for evolving short-duration rice variety. Genotype Samba Mahsuri and BPT 3068 were recorded as the shortest and tallest, respectively. The highest mean performance for productive tillers per plant was observed in BPT 2808 (15 tillers) followed by BPT 2824 (13 tillers). Genotype BPT 3137 (29.80 cm) exhibited the highest

mean performance for panicle length. The maximum grain yield per plant was observed 41.71 g by BPT 2854 indicating that these genotypes can be used in a hybridization programme to achieve a desirable increment in yield. A critical analysis on the mean performance of the genotypes studied revealed that BPT 3130, BPT 3164, BPT 3264, BPT 3092, BPT 3275, BPT 2824, BPT 2954, BPT 3276, BPT 3095, BPT 3178 exhibited significant superiority over the check Samba Mahsuri for the characters grain yield per plant, test weight, panicle length and were also on par for spikelet fertility and productive tillers per plant.

Table 2: Mean performance of rice genotypes for yield and yield components

Sl. No	Genotypes	DFF	PH	NPT	PL	SF	TW	GYP
1	BPT 1235	90	101.90	7	24.04	88.47	18.83	24.30
2	BPT 2231	116	98.40	10	25.30	93.21	19.39	26.81
3	BPT 2295	122	109.80	12	22.26	88.54	15.13	30.55
4	BPT 2411	113	116.00	8	25.25	79.69	19.56	23.01
5	BPT 2595	121	102.20	8	22.88	92.42	14.81	18.25
6	BPT 2620	102	108.40	7	23.10	90.07	16.01	20.88
7	BPT 2677	98	103.00	8	24.90	85.97	21.44	25.62
8	BPT 2764	121	105.80	8	23.50	88.72	16.48	15.54



Sl. No	Genotypes	DFF	PH	NPT	PL	SF	TW	GYP
9	BPT 2766	110	104.40	8	22.10	89.75	14.93	19.61
10	BPT 2776	116	107.80	10	22.90	94.39	16.03	27.05
11	BPT 2782	114	95.80	8	20.88	96.23	15.14	20.75
12	BPT 2808	112	101.00	15	21.80	87.82	15.81	30.52
13	BPT 2824	110	109.80	13	23.88	86.76	16.17	34.82
14	BPT 2846	111	101.40	6	22.30	84.45	15.28	32.12
15	BPT 2848	109	117.80	8	20.04	87.79	14.38	24.65
16	BPT 2849	111	111.00	8	24.28	79.60	19.38	16.12
17	BPT 2854	112	111.20	12	21.00	92.61	20.28	41.71
18	BPT 2863	114	105.00	9	23.40	91.82	16.35	29.08
19	BPT 2950	110	115.60	5	22.00	86.63	19.80	13.65
20	BPT 2954	96	130.80	9	22.90	93.65	18.68	34.48
21	BPT 2958	108	105.60	7	20.90	82.92	19.53	23.50
22	BPT 3032	101	104.40	8	22.20	91.08	22.19	22.61
23	BPT 3033	115	106.00	10	20.50	86.10	15.00	20.14
24	BPT 3061	110	117.60	8	24.20	94.60	15.57	19.34
25	BPT 3068	110	137.20	9	26.30	93.02	16.93	19.72
26	BPT 3074	95	114.80	9	21.90	92.94	16.94	30.51
27	BPT 3081	106	103.00	6	20.50	90.09	16.26	18.33
28	BPT 3086	110	122.20	5	28.70	86.29	16.57	16.86
29	BPT 3092	90	125.40	10	24.70	93.14	22.28	36.89
30	BPT 3095	114	118.80	8	24.80	96.32	21.19	34.04
31	BPT 3111	106	110.00	8	24.80	88.21	19.08	27.29
32	BPT 3113	107	104.20	9	26.50	84.07	18.13	32.16
33	BPT 3114	110	117.80	9	23.90	93.59	15.84	20.92
34	BPT 3115	110	97.60	12	21.70	94.67	14.58	30.71
35	BPT 3118	111	98.20	11	21.80	79.31	14.61	30.28
36	BPT 3120	110	93.60	9	21.40	86.53	20.02	23.50
37	BPT 3121	106	101.00	11	22.25	88.29	15.61	28.62
38	BPT 3129	110	109.40	7	22.38	81.92	16.55	24.04
39	BPT 3130	110	111.60	11	23.70	91.74	17.04	41.02
40	BPT 3133	109	115.40	10	22.80	94.07	15.78	27.01
41	BPT 3135	102	128.00	8	24.30	91.68	16.96	27.94
42	BPT 3136	107	116.20	7	24.10	87.02	17.85	26.95
43	BPT 3137	114	132.40	6	29.80	79.31	14.59	22.32
44	BPT 3145	119	107.60	7	25.10	91.73	23.61	20.33
45	BPT 3147	114	116.60	9	22.20	85.71	14.60	24.97
46	BPT 3148	110	109.60	6	21.90	85.40	19.60	28.08
47	BPT 3150	110	102.80	7	25.40	89.78	18.02	23.06
48	BPT 3151	101	106.80	9	22.30	76.74	16.06	32.93



Sl. No	Genotypes	DFF	PH	NPT	PL	SF	TW	GYP
49	BPT 3159	114	112.80	10	25.10	91.97	18.53	29.28
50	BPT 3164	105	105.20	12	22.75	87.87	20.40	40.86
51	BPT 3168	110	116.40	7	22.63	85.29	16.92	18.54
52	BPT 3170	106	114.20	7	22.60	85.54	19.32	23.61
53	BPT 3172	96	104.20	7	21.30	88.86	14.34	20.33
54	BPT 3178	106	100.60	11	24.20	91.53	18.90	33.56
55	BPT 3208	108	107.40	8	21.60	93.59	17.29	21.48
56	BPT 3244	105	108.60	10	22.60	90.76	15.54	27.54
57	BPT 3260	111	112.40	11	25.00	76.82	16.99	31.80
58	BPT 3261	113	111.40	6	26.83	85.70	19.30	15.91
59	BPT 3262	98	122.60	6	26.50	89.62	21.03	18.20
60	BPT 3263	112	126.00	9	25.70	90.80	17.68	23.03
61	BPT 3264	96	115.00	11	23.60	91.85	17.03	37.25
62	BPT 3269	98	109.40	8	21.70	95.64	14.69	20.36
63	BPT 3270	118	118.20	8	28.60	87.02	17.85	22.92
64	BPT 3274	118	116.00	6	25.30	88.73	18.89	27.61
65	BPT 3275	119	114.40	8	22.20	76.85	15.63	36.32
66	BPT 3276	113	116.40	7	23.60	94.13	19.19	34.25
67	BPT 3277	108	100.60	7	21.20	82.35	17.79	20.09
68	BPT 3279	106	108.20	10	24.20	93.41	16.42	25.33
69	BPT 3291	110	101.00	7	22.90	89.78	16.35	21.21
70	BPT 4358	117	93.50	7	23.00	82.38	13.64	12.87
71	Improved Samba Mahsuri	96	92.06	9	21.84	87.42	13.99	19.87
72	Krishnaveni	108	97.56	8	22.82	90.14	19.29	24.83
73	TN 1	85	123.46	10	22.86	92.60	24.43	25.87
74	Samba Mahsuri (Yield Check)	113	91.66	11	21.06	94.23	15.45	27.40
	Min	85	91.66	5	20.04	76.74	13.64	12.87
	Max	122	137.20	15	29.80	96.32	24.43	41.71
	Mean	108	109.87	9	23.40	88.65	17.46	25.73
	CV %	6.99	8.62	13.60	8.70	5.61	13.64	15.68
	C.D. (0.05)	2.53	4.03	2.11	1.41	3.06	0.41	2.55

Variability, Heritability and Genetic advance as per cent of mean

The estimates of phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability (broad sense) and genetic advance as per

cent of mean (GAM) are presented in **Table 3** and **Figures 1 and 2**. The results are discussed here under. GCV and PCV are high for grain yield per plant (22.66% and 23.77%) indicating the existence of sufficient variation among the genotypes for potential yield improvement through selection. High GCV,



PCV for grain yield per plant were also observed in the findings of Nithya *et al.*, (2020), Bhargava *et al.*, (2021), Chamar *et al.*, (2021), Gupta *et al.*, (2021) and Priyanka *et al.*, (2023). The traits like days to 50% flowering (5.84% and 5.59%), plant height (7.52% and 7.04%), panicle length (7.86% and 6.54%), spikelet fertility (5.12% and 4.46%) exhibited low phenotypic and genotypic coefficient of variation indicating the presence of less genetic variability as a result of which there is less scope for selection. Similar results were observed by Mohan *et al.*, (2016), Jan *et al.*, (2017), Adhikari *et al.*, (2018), Nithya *et al.*, (2020), Bhargava *et al.*, (2021) and Teja *et al.*, (2023).

High estimates of heritability combined with high genetic advance was observed for test weight (97.83%, 23.40) and grain yield per plant (90.90%, 44.51) provides evidence that this was under the control of additive gene effects and selection may be effective. Similar results were also reported earlier by Anyaoha et al., (2018), Nithya et al., (2020), Chamar et al., (2021), Bhargava et al., (2021), Gupta et al., (2021) and Priyanka et al., (2023). Similarly high heritability coupled with moderate genetic advance was observed for days to 50% flowering (91.65%, 11.03), plant height (87.57%, 13.57), panicle length (69.17%, 11.20) indicating the preponderance of additive and

non-additive gene actions in the expression of this trait. The results were in conformity with Gampala et al., (2015), Mohan et al., (2016), Adhikari et al., (2018) and Bhargava et al., (2021) whereas spikelet fertility (75.91%, 8.01) had exhibited high heritability combined with low genetic advance as per cent of mean due to the favourable influence of environment rather than the genotype, indicating the possibility of improvement of the trait through heterosis breeding rather than simple selection. High heritability coupled with low genetic advance as per cent of mean for spikelet fertility was reported by Parimala et al., (2021).

Conclusion

In a nutshell, based on the results obtained, the studied rice genotypes showed adequate variability. High PCV and GCV were recorded for grain yield per plant indicating the existence of large variation among the genotypes for potential yield improvement through selection. High heritability coupled with high genetic advance as per cent of mean was observed for the traits test weight and grain yield per plant revealing that these traits are governed by additive gene action and therefore selection can be practiced based on phenotypic performance.

Table 3: Variability, heritability and genetic advance as per cent of mean for yield and yield component traits

S.	Character	Coefficient	t of variation	Heritability	Genetic advance as Per cent of mean	
No.	Character	PCV (%)	GCV (%)	(%)		
1	Days to 50 per cent flowering	5.84	5.59	91.65	11.03	
2	Plant height	7.52	7.04	87.57	13.57	
3	Number of productive tillers	22.39	13.43	35.97	16.58	
4	Panicle length	7.86	6.54	69.17	11.20	
5	Spikelet fertility	5.12	4.46	75.91	8.01	
6	Test weight	11.61	11.49	97.83	23.40	
7	Grain yield per plant	23.77	22.66	90.90	44.51	



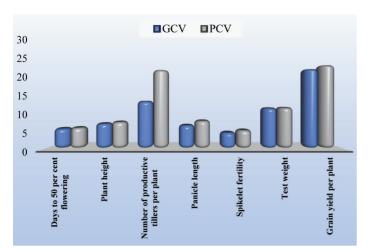


Figure 1: Variability parameters for yield and yield component traits

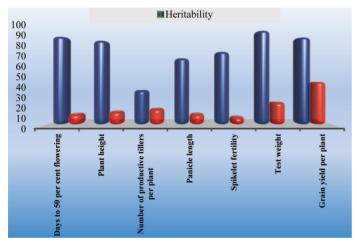


Figure 2: Heritability and Genetic advance as per cent mean for yield and yield component traits

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RESEARCH ARTICLE

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Genetic Diversity Analysis of Rice Germplasm (Oryza sativa L.) Using Morphological Markers

Sudarshana Negi, Neelam Bhardwaj* Kajal Bhardwaj and Deepika Sud

Department of Genetics and Plant Breeding, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur-176062 *Corresponding author Email: neenabhardwaj@gmail.com

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Abstract

The present investigation was carried out to assess the genetic diversity among 35 rice germplasm lines for 19 different traits including quantitative and grain quality traits. The genotypes were grouped into 8 clusters and out of these maximum number of genotypes were placed in cluster I (25 genotypes) followed by Cluster II (4 genotypes). The highest intra-cluster distance was observed for cluster I whereas, highest inter-cluster distance was recorded between cluster VII and cluster VIII. Cluster III, cluster V and cluster VIII showed the highest cluster mean values for most of the traits studied. The maximum contribution towards the genetic divergence was exhibited by spikelets per panicle followed by 1000 grain weight, days to 75% maturity and plant height. These traits, hence could be focused for selection improving grain yield. The germplasm lines falling in different clusters with high mean for grain yield and other component traits can be utilized for hybridization programme to obtain elite segregants.

Keywords: Rice germplasm, Markers, Genetic diversity, Cluster analysis

Introduction

Rice is the staple food for more than half the world's population of 7.8 billion people and plays a pivotal role in food security of many countries. More than 90% of the global production and consumption of rice is in Asia. It is the primary food source for more than one-third of the world's population and is grown on 11% of the world's cultivated area. It provides minerals, vitamins and fiber. To meet the challenge of producing more rice from limited available lands, we need varieties or hybrids which grow better under adverse conditions and possess high yield potential. Genetic diversity is the pre-requisite for any crop improvement programme because it helps in the development of superior recombinants, through selection of parents having wider variability for different characters. Since, the last few centuries, rice has faced loss in diversity due to replacement of native

varieties with high yielding varieties (Choudhary et al., 2013). Genetic divergence analysis evaluates the genetical distance among the selected genotypes and shows the relative contribution of specific traits towards the total divergence. A higher heterosis could be achieved from crosses between genetically distant parents (Falconer, 1960). Therefore, the present investigation is aimed to assess the nature and magnitude of genetic divergence present in the 35 germplasm lines of basmati rice and to select suitable diverse genotypes as parents for further utilization in crop improvement programmes.

Materials and Methods

Experimental Material and Plan

Experiment was conducted during *kharif*, 2022 at Chaudhary Sarwan Kumar Himachal Pradesh Krishi



Vishvavidyalaya Rice and Wheat Research Centre, Malan. The experimental material comprised of 35 germplasm lines of rice. The nursery was sown on 28th May and 21 days old seedlings were transplanted in the field in Randomized block design with three replications. Standard agronomic practices and plant protection measures were taken as per schedule. Each germplasm line was raised in a 3 rowed plot of 3 m length with a spacing of 20 × 15 cm respectively.

Morphological Markers

Observations were recorded on five randomly selected plants per replication for different quantitative traits: days to 50 per cent flowering (DF), days to maturity (DM), total tillers per plant (TT), effective tillers per plant (ET), plant height (PH), panicle length (PL), spikelets per panicle (SPP), grains per panicle (TG), spikelet fertility per centage (SFP), 1000- grain weight (TW) biological yield per plant (g), harvest index per cent and grain yield per plant (GYPP); four quality traits: grain length (GL), grain breadth (GB), L/B ratio (LB) and amylose content (AC).

Statistical analysis

Genetic divergence was estimated by Mahalanobis' D² statistics (1936). The germplasms lines were grouped into a number of clusters by Tocher's method described by Rao (1952). Each character was ranked on the basis of values in all the combination of genotype for estimation of contribution of individual characters towards divergence.

Results and Discussions

Cluster distances and composition

It is the task of grouping a set of genotypes in which accessions falling in the same group are more similar to each other than to those in other groups or clusters which is very helpful in diversity analysis. In the present investigation with nonhierarchical Euclidean cluster analysis, 35 rice germplasm lines were

grouped into eight different clusters based on the inter se genetic distances. Out of eight clusters two clusters were polygenotypic while six were monogenotypic based on genetic divergence. Maximum number of genotypes were placed in cluster I (25 genotypes) followed by cluster II (4 genotypes), cluster III (1 genotype) and cluster IV (1 genotype), cluster V (1genotype), cluster VI (1 genotype), cluster VII (1 genotype), cluster VIII (1genotype). Genotypes falling under each cluster are presented in Table 1 which indicates cluster 1 represent maximum genotypes. Similar studies were taken up by Chakravorty et al., (2013) who assessed genetic divergence among 51 rice genotypes and grouped them into 11 clusters using D² analysis and Akhter et al., (2022) who assessed genetic divergence among rice genotypes and grouped them into different clusters using D² analysis. Rao et al., (2018) conducted diversity analysis in rice germplasm and divided the genotypes into 8 clusters. Tushara et al., (2022) also conducted similar studies on grouping of rice germplasm into different clusters.

Table 1: Grouping of rice genotypes into different clusters on the basis of Mahalanobis D² -analysis

Clusters	No. of genotype	Genotypes		
Cluster 1	25	HPR-5021, HPR-5024, HPR-2612, HPR-5022, HPR-5007, HPR-5015, HPR-5008, PB-1509, HPR-3221, Kasturi, HPR-5020, HPR-2750, HPR-5023, HPR-5016, HPR-5001, HPR-5018, HPR5017, HPR-5003, HPR5012, HPR-5009, HPR-5013, HPR-5026, HPR-2749, HPR-5019, HPR-5025		
Cluster 2	4	HPR-5004,HPR-5005, HPR-5002, HPR-5006		
Cluster 3	1	HPR-2696		
Cluster 4	1	HPR-5014		
Cluster 5	1	HPR-5010		
Cluster 6	1	HPR-3225		
Cluster 7	1	HPR-5011		
Cluster 8	1	HPR-5027		



The average intra and inter-cluster distances are presented in **Table 2**. The highest intra-cluster distance is observed for cluster I (18.31), followed by cluster II (12.62) and no intra-cluster distance is observed for cluster III, cluster IV, cluster V, cluster VI, cluster VII and cluster VIII as these clusters possess only one genotype each. Presence of high intra-cluster distances revealed that genotypes within the same cluster were quite diverse; hence the selection of parents within the cluster would be effective. The highest inter-cluster distance was 42.92, which was observed between clusters VII and cluster VIII followed by VI and VII

(39.80), V and VIII (35.02), V and VI (34.53), IV and VIII (34.17), II and VIII (33.01), I and VIII (32.02) and III and V (31.71) indicating that, hybridization between the most diverse genotypes would yield desirable segregants with the accumulation of favorable genes in the segregating generations, while, least inter cluster distance was observed between cluster V and VII (18.48) indicating that the genotypes belonging to these clusters were comparatively less diverse. Similar studies were also conducted by Kishore *et al.*, (2018), Vennila *et al.*, (2011), Chakravorty *et al.*, (2013) and Devi *et al.*, (2015).

Table 2: Average intra and inter-cluster distance

Clusters	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI	Cluster VII	Cluster VIII
Cluster I	18.31	21.67	22.78	22.39	23.19	23.65	22.93	32.03
Cluster II	0	12.62	26.43	26.38	24.48	30.09	30.18	3 3 . 0 1
Cluster III		0	0	29.97	31.78	21.17	30.99	28.55
Cluster IV				0	21.95	25.22	22.87	34.17
Cluster V					0	3 4 . 5 3	18.48	35.02
Cluster VI						0	26.14	39.80
Cluster VII							0	42.92
Cluster VII								0

Cluster means for different characters:

The cluster means of rice genotypes falling under different clusters are presented in **Table 3**. Analysis of cluster means indicates existence of considerable differences in the mean values of different traits. The highest values of total tillers per plant, effective tillers per plant, biological yield per plant and amylose content were observed in cluster III. Cluster V showed highest value for plant height, flag leaf length, 1000-grain weight and grain breadth. Cluster VIII showed highest value for days to 50% flowering, days to 75% maturity, harvest index and grain yield per plant Cluster IV showed highest values for flag leaf breadth, grain length, L/B ratio. Cluster VI showed highest value for panicle length and spikelets

per plant and cluster VII showed highest value for spikelet fertility and grains per panicle. The cluster IV exhibited minimum values for most of the traits *viz.*, days to 75% maturity, flag leaf length, total tillers per plant, effective tillers per plant, spikelet fertility, biological yield per plant and grain yield per plant while, cluster VI showed minimum values for plant height, grain length and grain breadth. Thus, various traits contribute to the total divergence in cluster, III, V and VII and the genotypes comprising these clusters seem to be quiet promising for many of the traits under study. Similar studies in rice were also done by Sharma *et al.*, (2011), Chakravorty *et al.*, (2013), Kumar (2015) and Amudha and Ariharasutharsan (2021).



Table 3: Cluster means of eight clusters for yield and yield related traits of rice genotypes

Cluster/ Character	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI	Cluster VII	Cluster VIII	Mean	Min	Max
Days to 50% flowering	87.78	88.58	94.00	82.33	87.00	86.00	80.00	108.00	89.21	80.00*	108.00**
Days to 75% maturity	121.01	120.75	118.00	108.00	122.67	119.33	122.00	124.67	119.55	108.00*	124.67**
Plant height (cm)	120.69	128.74	108.13	122.20	146.87	85.93	112.53	134.93	120.00	85.93*	146.87**
Flag leaf Length (cm)	36.05	36.48	33.83	29.53	37.30	30.20	32.77	34.37	33.82	29.53*	37.30**
Flag leaf width (cm)	1.48	1.48	1.53	1.87	1.73	1.50	1.73	1.43	1.59	1.43*	1.87**
Total tillers per plant	8.39	8.02	11.47	6.60	8.53	11.30	9.27	7.60	8.90	6.60*	11.47**
Effective tillers per plant	7.93	7.34	10.47	6.47	8.13	10.40	8.47	7.47	8.34	6.47*	10.47**
Panicle length (cm)	25.64	25.42	24.30	24.97	24.07	26.70	23.80	23.13	24.75	23.13*	26.70**
Spikelets per plant	180.87	136.27	182.90	193.73	182.47	199.40	195.67	172.40	180.46	136.27 *	199.4 0**
Spikelet fertility (%)	91.96	94.29	94.57	76.23	97.10	78.07	98.17	96.03	90.80	76.23 *	98.17**
Grains per panicle	166.29	128.31	172.93	147.73	177.13	155.63	192.00	165.60	163.20	128.31*	192.00**
1000 grain weight (g)	23.73	23.23	20.63	26.20	31.13	22.73	30.17	26.23	25.51	20.63*	31.13**
Harvest index (%)	34.95	34.45	26.07	37.83	25.90	34.30	37.37	40.17	33.88	25.90*	40.17
Biological yield per plant (g)	43.82	37.13	58.63	33.23	55.27	40.6	39.6	44.93	44.15	33.23*	58.63**
Grain length (mm)	6.33	5.63	5.70	7.17	6.60	5.13	6.50	6.63	6.21	5.13*	7.17 **
Grain breadth (mm)	1.82	1.82	1.93	1.73	2.20	1.60	2.13	1.97	1.90	1.60*	2.20**
L/B ratio	3.52	3.12	2.96	4.15	3.00	3.22	3.06	3.39	3.30	2.96*	4.15**
Amylose content (%)	23.31	22.04	25.89	24.85	22.99	24.50	25.40	21.53	23.81	21.53*	25.89**
Grain yield per plant (g)	15.23	12.66	15.30	12.60	14.33	13.97	14.40	17.73	14.53	12.60*	17.73**

Trait contribution towards the divergence

A comparison of contribution of different traits towards genetic diversity was estimated based on ranking method. Contribution of different traits to total divergence is presented in Table 4. The maximum contribution towards the genetic divergence was exhibited by spikelets per panicle followed by 1000 grain weight, days to 75% maturity, plant height, days to 50% flowering, spikelet fertility, grain yield per plant, amylose content, grains per panicle, flag leaf length, grain length, flag leaf width, grain breadth, L/B ratio, harvest index and biological yield. Hence, spikelets per panicle, 1000 grain weight, plant height and days to 50% flowering, were found to be potential contributors to genetic divergence. The results are supported by the earlier findings of Devi et al., (2015) and Rachappanavar (2017) who revealed similar findings.

Table 4: Relative contribution of individual trait towards divergence among rice genotypes

Traits	Rank	Contribution
Days to 50% flowering	48	7.62%
Days to 75% maturity	74	11.75%
Plant height	74	11.75%
Flag leaf Length	22	3.49%
Flag leaf width	8	1.27%
Total tillers	0	0.00%
Effective tillers	0	0.00%
Panicle length	0	0.00%
Spikelets per panicle	185	29.37%
spikelet fertility	31	4.92%
Grains per panicle	24	3.81%
1000 grain weight	88	13.97%
Harvest index	2	0.32%
Biological yield	1	0.16%
Grain length	10	1.59%
Grain breadth	6	0.95%
L/B ratio	5	0.79%
Amylose content	25	3.97%
Grain yield per plant	27	4.29%



Conclusion

A significant range of variation is evident among thirty-five rice genotypes evaluated in the present study. The thirty-five rice genotypes were grouped into eight clusters which was in consonance with the clustering pattern obtained by Mahalanobis D² statistics. The parents for hybridization program should be selected on the basis of magnitude of genetic distance, contribution of different characters towards the total divergence and magnitude of cluster means for different characters performance having maximum heterosis. Genetic diversity studies revealed that maximum number of genotypes were placed in cluster I. The highest intra-cluster distance was observed for cluster I, whereas, highest intercluster distance was recorded between cluster VII and cluster VIII followed by V and VII, IV and VIII, II and VIII, I and VIII and III. Hence, crosses between the genotypes of these clusters are expected to manifest high heterosis along with, accumulation of favorable genes in subsequent segregating generations.

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RESEARCH ARTICLE

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Unveiling Genetic Variation in Rice Hybrids Through Hierarchical Clustering and Principal Component Analysis

Vijay Kumar Reddy C, Amarnath K and Ravi Kumar BNVSR*

Acharya N. G. Ranga Agricultural University, Regional Agricultural Research Station, Nandyal, Andhra Pradesh *Corresponding author Email: bnvsr.ravikumar@angrau.ac.in

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Abstract

The present investigation was carried out with 67 rice hybrids along with eight checks (four varietal and four hybrid checks) to ascertain the extent of genetic diversity for yield and yield associated traits through multivariate techniques like hierarchical clustering and principal component analysis (PCA). By using Wards method of clustering, 67 rice hybrids along with eight checks were aggregated into eight clusters based on different traits in which cluster VI comprised of 15 hybrids is the largest one followed by cluster III and IV with 12 hybrids. The hybrids in cluster I and II had highest values for test weight and effective bearing tillers /m² respectively. Similarly, the hybrids in cluster III recorded maximum values for plant height and Grain yield. In PCA, the total variation was bisected into 10 major principal components (PCs) in which PC1, PC2, PC3 and PC4 with eigen values more than one describing 24.76%, 23.26%, 14.54 and 13.22%, respectively attributing for overall variation of 75.80%. From the present study, the hybrids *viz.*, NRH 24, NRH 46, NRH 40, NRH 38, NRH 53, NRH 2; Hybrid checks HC2 (US 314), HC4 (HRI 174) and varietal check VC1 (BPT 5204) were identified to be genetically potential for commercial exploitation to enhance yield and its attributing traits in rice.

Keywords: Rice, Genetic diversity, Cluster analysis, Principal component analysis

Introduction

Rice (*Oryza sativa* L.), a global food grain and an important staple food crop for half of the global population. Globally, India holds second place in rice production next to China. The major rice producing states in India include West Bengal, UP, Andhra Pradesh, Punjab, Telangana and Tamil Nadu. Besides, West Bengal and Uttar Pradesh produce 30% of total quantity of rice produced in the country. In India, the rice crop reported a production of 203.6 million tonnes from 47.8 million ha with average productivity of 4259 Kg/ha (https://ipad.fas.usda.gov/countrysummary/Default.aspx?id=IN&crop=Rice). In Andhra Pradesh, the crop is cultivated in total area of 2.13 million ha with production of 12.63

million tonnes and productivity of 5932 million tonnes (Agricultural statistics at a glance, 2022-23, Directorate of Economics and Statistics, Government of Andhra Pradesh, 2022-23). In the present national scenario, the population growth rate is reached to 1.58% and the requirement of rice was estimated to be around 140.7 million tonnes by 2025 (http://worldfood.apionet.or.jp). In order to make India self-sufficient in rice, enhancement of rice productivity to larger extent is a prime requisite (Hossain, 1996; Mishra, 2002). Despite this, task is quite challenging for breeders as the options available are very limited. Hence, breeders need to identify genetically diverse and potential hybrids for their inclusion in crop



improvement programme by divergence studies for yield and its attributing traits.

Although yield is a complex and challenging trait which depends mainly on environment and other different variables. For an efficient selection, breeder can reduce the number of characteristics and this can be made possible by using a method known as PCA. PCA is one of the multivariate techniques utilized in data analysis that converts data into a series of new orthogonal variables called principal components by linearly combining the variables that account for majority of variance in the original variables (Abdi and Williams, 2010) and it comprehends non-parametric strategy from a complicated set of data (Tiwari et al., 2022). The main advantage of using PCA over cluster analysis is that each genotype can be assigned to one group only (Mohammadi, 2002). The cluster analysis is a pertinent method for concluding relationship among hybrids and amount of genetic distance from each other (Mellingers, 1972). Divergence analysis using PCA and hierarchical cluster analysis has been shown to be effective in determining potential hybrids useful for hybridization (Chaudhary et al., 2015). The prime objective of this investigation was to unveil genetic diversity and identify the best divergent hybrids for their inclusion in commercial rice improvement programme.

Materials and Methods

The experimental material consisted of 67 rice hybrids along with eight checks (four varietal and four hybrid checks) which were evaluated at Regional Agricultural Research Station (RARS), Nandyal, Andhra Pradesh, India during *kharif*, 2023. All the hybrids were evaluated in Augmented Block Design with plot size of 10 m² per hybrid with a spacing of 20 cm x 15 cm. All the agronomic practices recommended by Acharya N.G. Ranga Agricultural University were followed to raise a healthy crop. Data was collected from five competitive and randomly selected plants for recording yield and yield attributes *viz.*, Days to 50% flowering (DFF), Days to maturity (DM), Plant

height (cm) (PH), Effective bearing tillers/m² (EBT/m²), Panicle length (cm) (PL), Filled Grains per panicle (FGP), Unfilled grains per panicle (UFGP), Total grains per panicle (TGP), Spikelet fertility % (SF %), Test weight (g) (TW) and grain yield (Kg/m²)(GY) whereas DFF and DM data was recorded on plot basis. PCA (Hotelling, 1936) and cluster analysis were used to identify the most contributing traits for variation and diversity among hybrids, respectively (Peeters and Martinelli, 1989). The data generated by evaluation of hybrids s forwarded to statistical analysis to estimate genetic diversity through PCA and hierarchical clustering using JMP 17.0 statistical software (SAS Institute Inc., Cary, NC, USA).

Results and Discussions

The performance of 67 rice hybrids together with eight checks evaluated for eleven yield and yield attributes is presented in Table 1. The hybrid NRH 3 was found to be early flowering (81 days), while VC2(MTU 1262) was identified as late flowering (114 days). DM ranged from 109 days (NRH2) to 144 days (NRH 51). NRH 36 was found as taller (123 cm) whereas VC1 (BPT 5204) noticed as shorter (87.70 cm) with an average of 106.98 cm. The hybrid NRH 43 was found to bear a greater number of effective tillers (726 / m²), while least number of effective tillers (238 / m²) was displayed by hybrid, NRH 8. The mean value of PL was found to be 25.19 cm with range of 21.22 cm (HC2) to 27.86 cm (NRH 55). The TGP ranges from 160 (HC 2) to 599 (NRH 40) with mean value of 337. The mean SF % was recorded as 84.55% in which NRH 34 topped the list with 93.64% and NRH 17 occupied the bottom position with 60.06%. The TW ranged from 11.27 g (NRH 40) to 23.27 g (NRH 11) with average TW value of 15.98 g. The mean GY was recorded as 1.27Kg/m² with ranges from 0.612 Kg/m² (NRH 26) to 2.152 Kg/m² (NRH 5) among the studied hybrids. Vasudeva Reddy et al., (2023) reported same kind of experimental results while evaluating hybrids for yield and its component traits in rice.



Table 1. Per se performance of 67 hybrids along with eight hybrids for yield and its component traits

Compiliation			(ma)	m/107		5)	0 1)	(111)	
5 29	92	120	103.8	389	24.90	193	50	243	79.57	20.24	1.246	-13.24
AE 1325 8	82	109	95.3	310	24.90	296	22	318	95.96	15.20	0.870	-39.42
IR 79156 A / AE 1305	81	110	97.2	277	25.90	301	21	322	93.60	16.52	1.002	-30.22
	82	110	92.3	277	24.34	298	33	331	90.02	14.23	1.152	-19.78
APMS 6A / RTCNP 103 9	26	125	109.4	442	26.06	239	19	258	92.56	16.23	2.152	49.89
APMS 6A / RTCNP 152 8	88	115	106.5	422	23.94	240	25	265	90.55	17.29	1.304	-9.17
8 29	85	113	6.96	479	25.40	336	42	377	88.98	14.47	1.542	7.35
	87	125	95.5	238	24.90	307	26	333	92.30	16.21	1.354	-5.71
5	96	125	8.96	403	25.70	270	26	296	91.14	13.80	1.020	-28.97
151 9	66	128	114.6	432	27.80	294	50	344	85.53	19.36	1.631	13.61
167 9	26	125	105.1	640	25.08	296	43	339	87.32	23.27	1.782	24.09
APMS 9A / RTCNP 168	66	126	111.2	353	25.68	245	99	310	78.80	20.95	1.186	-17.39
	101	130	103.7	653	25.94	204	45	249	81.90	16.23	0.963	-32.94
1	102	129	109.0	317	24.70	271	82	353	89.92	15.83	1.846	28.54
	95	124	113.4	370	26.50	315	37	352	89.49	18.20	1.470	2.37
	87	116	103.4	436	25.76	277	26	303	91.55	18.30	1.752	22.01
APMS 11A / RTCNP 152 9	06	116	109.9	376	26.52	206	137	343	90.09	19.35	0.881	-38.68
APMS 11A / RTCNP 167 1	101	133	103.5	386	26.22	214	37	251	85.26	15.50	1.709	19.02
APMS 11A/RTCNP 170 9	95	125	110.6	389	26.64	197	63	260	75.67	18.77	1.201	-16.39
APMS 11A / RTCNP 176 9	86	128	105.9	574	25.40	201	32	233	86.27	16.77	1.494	4.04
	95	123	115.3	989	25.04	216	52	268	80.60	19.45	1.134	-21.03
	97	125	106.5	614	25.48	257	41	298	86.25	18.93	1.422	-0.97
6	66	128	99.1	548	26.14	248	52	300	82.67	17.74	1.368	-4.74
APMS 14A / RTCNP 167 1	111	137	104.7	409	24.18	189	92	265	71.42	17.32	0.695	-51.61
<u> </u>	06	117	106.1	389	26.50	370	26	396	93.39	15.54	0.987	-31.27
Ś	92	117	98.5	330	25.30	336	53	388	86.45	15.55	0.612	-57.38
15A / NICRA P3 8	83	112	106.3	350	26.46	340	39	380	89.63	17.74	1.674	16.57
	98	116	106.3	429	25.00	335	42	377	88.76	16.30	1.602	11.56
15A / RTCNP 169 1	107	136	108.7	393	24.32	218	83	301	72.38	15.86	1.220	-15.03
67 1	102	130	114.9	541	26.70	301	41	342	88.00	14.21	1.467	2.16
	88	118	102.6	396	26.00	324	27	350	92.40	14.26	0.828	-42.34
	94	124	88.9	488	24.10	197	52	249	79.12	13.24	1.035	-27.92
APMS 16A / RTCNP 170 1	105	135	114.1	409	24.96	250	85	336	74.57	16.22	1.040	-27.58
16A / RTCNP 37 9	95	124	119.1	703	25.70	324	22	346	93.64	13.93	1.233	-14.14
APMS 16A / RTCNP 66 9	66	128	106.1	895	24.56	224	43	267	83.92	14.52	1.233	-14.14
67 1	100	128	123.0	535	25.66	260	28	288	90.29	15.64	1.693	17.90
APMS 16A / SN 698	96	125	100.5	422	26.40	339	77	415	81.51	11.51	0.906	-36.91
CM 449	90	118	92.4	350	22.40	422	105	528	90.08	13.30	0.690	-51.95
	97	127	101.7	376	24.20	352	57	409	86.05	15.40	1.521	5.92
\dashv	96	125	106.4	403	26.90	485	114	599	80.93	11.27	1.122	-21.87
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S. No.	Entry	Cross Combination	DFF	DM	PH (cm)	EBT/m ²	PL(cm)	FGP	UFGP	TG/P	SF %	TW (G)	GY (kg/sq.m)	Heterosis %
42	NRH 42	APMS 17A / RTCNP 66	102	130	114.6	673	24.40	198	42	240	82.50	14.75	1.242	-13.51
43	NRH 43	APMS 17A / RTCNP 67	6	125	101.5	726	24.80	277	48	325	85.21	14.28	1.341	-6.62
44	NRH 44	APMS 18A / CM 460	6	127	108	409	25.10	496	72	268	87.26	13.84	1.038	-27.72
45	NRH 45	APMS 18A / RTCNP 145	86	127	106	528	26.46	356	51	407	87.47	14.18	1.146	-20.19
46	NRH 46	APMS 18A / RTCNP 166	106	133	6.111	449	27.10	286	132	418	68.46	15.69	1.366	-4.89
47	NRH 47	APMS 18A / RTCNP 172	26	125	6.801	380	24.90	212	<i>L</i> 8	298	96.07	17.96	1.673	16.52
48	NRH 48	APMS 18A / RTCNP 176	66	130	114.7	373	25.04	301	103	404	74.48	14.83	1.293	-9.99
49	NRH 49	APMS 18A / RTCNP 66	93	120	118.3	475	25.50	273	36	309	88.33	16.61	0.99	-31.06
50	NRH 50	APMS 18A / RTCNP 84	104	128	107.2	350	24.68	320	74	394	81.17	16.30	0.958	-33.30
51	NRH 51	APMS 18A / RTCNP 97	105	134	115.8	413	25.74	318	98	404	78.77	16.42	1.304	-9.17
52	NRH 52	APMS 18A / RTCNP 99	104	132	105.9	370	26.00	394	29	462	85.40	14.41	1.643	14.38
53	NRH 53	APMS 19A / BM 563	91	119	108.7	403	25.42	469	61	531	88.47	15.11	0.840	-41.50
54	NRH 54	APMS 19A / BM 571	86	127	1.601	422	24.78	394	35	430	91.76	12.19	1.512	5.29
55	NRH 55	APMS 19A / RTCNP 103	6	126	113.4	383	27.86	392	99	458	85.64	14.90	1.278	-11.00
99	NRH 56	APMS 19A / RTCNP 133	66	129	112.5	462	25.34	478	82	556	85.98	16.64	1.506	4.87
57	NRH 57	APMS 19A / RTCNP 167	93	120	<i>L</i> '601	502	24.56	194	32	226	85.84	11.89	1.381	-3.83
58	NRH 58	APMS 19A / RTCNP 171	94	124	111.1	363	22.10	205	68	294	69.75	14.76	1.019	-29.03
59	NRH 59	APMS 19A / RTCNP 172	94	127	110.1	383	25.28	263	92	338	99'./_	17.20	1.418	-1.24
09	NRH 60	APMS 19A / RTCNP 173	100	130	109.7	399	25.08	296	59	355	83.33	17.56	1.361	-5.20
61	NRH 61	APMS 19A / RTCNP 174	86	127	111.1	396	22.58	286	23	309	92.49	17.67	1.471	2.45
62	NRH 62	APMS 19A / RTCNP 175	66	129	113.3	389	24.46	198	41	239	82.69	18.33	1.116	-22.28
63	NRH 63	APMS 19A / RTCNP 176	86	130	115.3	373	25.68	310	92	386	80.37	13.63	1.480	3.05
64	NRH 64	APMS 19A / RTCNP 66	95	124	119.9	455	25.70	247	33	280	88.21	15.03	1.412	-1.67
65	NRH 65	APMS 19A / RTCNP 67	90	118	121.4	561	24.30	236	25	261	90.41	15.68	1.386	-3.48
99	NRH 66	APMS 19A/RTCNP 97	86	128	1.901	406	26.44	299	71	370	80.78	14.89	1.339	-6.73
29	NRH 67	APMS 19A / RTCNP 99	106	132	110.8	380	24.50	325	71	396	82.04	11.60	1.417	-1.30
89	VC1	BPT 5204	111	140	87.7	502	23.04	175	22	197	88.73	14.30	1.160	
69	VC2	MTU 1262	114	144	106.9	386	24.76	363	42	405	89.62	12.57	0.990	
70	VC3	NDLR 7	100	129	97.4	360	23.72	250	22	272	91.77	13.19	1.315	
71	VC4	NDLR 8	96	126	103.9	432	23.96	175	30	205	85.19	15.59	1.366	
72	HC1	US 312	96	125	107.2	393	26.48	296	37	333	88.88	18.16	0.831	
73	HC2	US 314	85	115	106.6	452	23.92	143	18	160	89.03	20.43	1.250	
74	HC3	27 P 63	96	126	108.9	403	21.22	267	38	305	87.54	14.98	1.356	
75	HC4	HRI 174	101	130	9.901	386	24.28	165	30	195	84.72	20.49	1.436	
Mean			96	125	106.9	435	25.19	285	52	337	84.55	15.98	1.27	
Maximum	un		114	144	123	726	27.86	496	137	599	93.64	23.27	2.15	
Minimum	um		81	109	87.7	238	21.22	143	18	160	90.09	11.27	0.61	
SD			6.74	6.82	7.22	100.35	1.20	77.44	26.81	87.90	82.9	2.36	0.29	
SE			0.778	0.788	0.834	11.588	0.139	8.942	3.096	10.150	0.783	0.274	0.034	

DFF-days to 50% flowering, DM-days to maturity, PH-plant height, EBT/m²- Effective bearing tillers/m², PL-panicle length, FGP-filled grains per panicle, UFGP-unfilled grains per panicle, TGP-total grains per paniele, SF %- spikelet fertility %, TW-test weight, GY- grain yield



Hierarchical cluster analysis was conducted with 67 rice hybrids along with eight checks using Wards method which provides the best result to get the finest possible classification. The cluster analysis revealed the aggregation of hybrids into eight clusters (Table 2 and Figure 1). The cluster means computed for eleven major yield attributing characters revealed the existence of ample amount of variation among the clusters (Table 3). The highest and lowest cluster means were recorded for the traits EBT/m² (631.4) and GY (1.00), respectively. Maximum cluster mean of overall traits was noticed in cluster VII (174.60) followed by cluster VIII (154.20). In contrast, the least cluster mean was displayed by cluster I

(121.70). This clearly infers the existence of ample amount of genetic divergence in the hybrids of these clusters. Further, among the eight divergent clusters, the highest numbers of hybrids were grouped in cluster VI with 15 hybrids followed by 12 hybrids in cluster III and IV. The hybrids in cluster IV showed highest mean values for DFF, DM, PH, EBT/m² and SF %. The hybrids of cluster III and cluster VIII showed maximum value for GY. Ravikumar *et al.*, (2015), Tejaswani *et al.*, (2016), Tejaswini *et al.*, (2018), Muthuramu and Sakthivel (2018), Dhakal *et al.*, (2020), Kusuma Kumari *et al.*, (2021) and Amudha and Ariharasutharsan (2021) also documented same kind of clustering of accessions into distinct clusters.

Table 2: Grouping of different hybrids into different clusters

Cluster	No. of hybrids	Hybrids
I	5	NRH 1, NRH 12, NRH 19, NRH 62,HC4
II	9	NRH 11, NRH 13, NRH 35, NRH 42, NRH 43, NRH 20, NRH 22, NRH 23, NRH 21
III	12	NRH 5, NRH 18, NRH 10, NRH 15, NRH 30, NRH 45, NRH 55, NRH 34, NRH 36, NRH 64, NRH 49, NRH 65
IV	12	NRH 2, NRH 3, NRH 4, NRH 25, NRH 31, NRH 26, NRH 41, HC1, NRH 7, NRH 28, NRH 16, NRH 27
V	11	NRH 6, HC2, NRH 61, HC3, NRH 8, NRH 9, VC3, NRH 32, NRH 57, VC4, VC 1
VI	15	NRH 14, NRH 47, NRH 48, NRH 63, NRH 51, NRH 50, NRH 59, NRH 60, NRH 66, NRH 46, NRH 24, NRH 29, NRH 33, NRH 58, NRH 17
VII	6	NRH 37, NRH 40, NRH 44, NRH 53, NRH 56, NRH 38
VIII	5	NRH 39, NRH 54, NRH 52, NRH 67,VC2

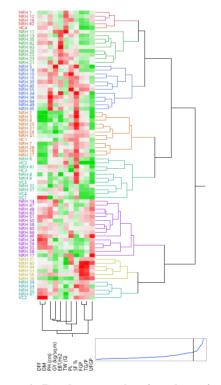


Figure 1: Dendrogram showing clustering by Wards method

DFF-days to 50% flowering, DM-days to maturity, PH-plant height, EBT/m²- Effective bearing tillers/ m², PL-panicle length, FGP-filled grains per panicle,

UFGP-unfilled grains per panicle, TGP-total grains per panicle, SF %- spikelet fertility %, TW-test weight, GY- grain yield



Table 3: Cluster means	of various	characters of	rice hy	vbrids under study	
Table 5. Cluster means	oi various	characters of	1100 11	ybiids dilaci stady	

Cluster No.	DFF	DM	PH	EBT/m ²	PL	FGP	UFGP	TGP	SF %	TW	GY	Mean
Cluster I	96.9	125.7	109.1	381.5	25.2	199.5	50.0	249.4	80.3	19.8	1.2	121.7
Cluster II	98.3	126.9	106.4	631.4	25.2	235.6	44.2	279.9	84.1	17.3	1.3	150.1
Cluster III	96.8	125.5	114.7	484.3	26.2	287.6	37.1	324.7	88.7	15.8	1.5	145.7
Cluster IV	87.1	115.5	101.2	373.5	25.7	322.4	34.6	357.1	90.4	16.2	1.2	138.6
Cluster V	94.8	124.5	101.2	417.9	23.8	227.5	28.6	256.1	88.5	15.3	1.3	125.4
Cluster VI	100.5	129.3	110.5	386.1	25.1	263.0	87.7	350.7	74.7	16.3	1.3	140.5
Cluster VII	94.8	123.8	104.8	408.1	25.3	448.2	84.6	532.8	84.0	13.6	1.0	174.6
Cluster VIII	103.7	132.2	106.1	386.8	24.8	365.7	54.6	420.3	87.0	13.2	1.4	154.2
Mean values	96.6	125.4	106.7	433.7	25.2	293.7	52.7	346.4	84.7	15.9	1.3	

Bold figures indicate maximum and minimum values in each character. DFF-days to 50% flowering, DM-days to maturity, PH-plant height, EBT/m²- Effective bearing tillers/m², PL-panicle length, FGP-filled grains per panicle, UFGP-unfilled grains per panicle, TGP-total grains per panicle, SF %- spikelet fertility %, TW-test weight, GY- grain yield

The constellation plot based on Wards method (Figure 2) depicts relationship among the 67 hybrids together with eight checks. The hybrids are grouped as end points and every cluster join as a new point with lines drawn will act as membership in constellation plot. The plot divided the total hybrids into 8 clusters with membership of 5,9,12,12,11,15,6 and 5. The Clustering pattern divulged that majority of hybrids congregated in cluster VI (15), followed by Cluster III (12) and Cluster IV (12). The hybrids with longer line representing greater genetic distance between the clusters. Further, the identified hybrids with maximum genetic distance are considered as superior and exploited commercially in yield improvement programme in rice.

The PCA an authentic tool utilized for successful selection of divergent genotypes in crop improvement programme. The results of PCA revealed the significance of first four PCs in discriminating 67 rice hybrids along with eight checks. The first four PCs, PC1, PC2 PC3 and PC4 exhibited eigen value

greater than one explaining 75.80% of total variation. The eigen values and total cumulative per centage of variances explained by PCs is furnished in **Table 4**. PC1 with eigen value of 2.724 contribute 24.76% of the total variability, PC2, PC3 and PC4 with eigen value of 2.559, 1.604 and 1.455 attributed 23.62%, 14.54% and 13.22% of the total variability, respectively. The first PC displayed high positive weight to UFGP (0.850), DFF (0.645), TGP (0.624) and DM (0.622). The second PC displayed highest positive loading to DFF (0.569), DM (0.563) and EBT (0.438). Likewise, the third and fourth PCs gave positive loading to GY (0.588) and TW (0.661), respectively (**Table 5**).

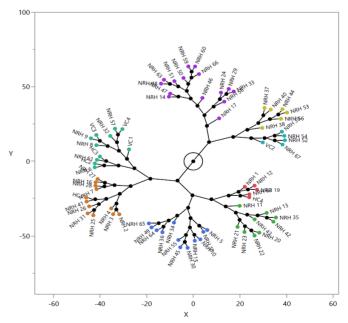


Figure 2: Constellation plot of 67 hybrids along with eight checks into Eight clusters based on Euclidean distance



Table 4: Total variances explained by different principal components in rice hybrids

Components	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
Eigen values	2.724	2.559	1.604	1.455	0.862	0.682	0.56	0.506	0.033	0.012
Proportion variance %	24.76	23.262	14.547	13.229	7.83	6.21	5.14	4.60	0.30	0.11
Cumulative variance %	24.76	48.03	62.577	75.806	83.65	89.86	94.99	99.59	99.89	100.00

Table 5: Factor loading of different characters with respect to different principal factor in rice hybrids

Principal	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
Components	101	1 C2	103	104	103	100	107	100	109	1 C10
DFF	0.645	0.569	0.150	-0.402	0.075	0.184	0.106	0.076	0.129	-0.007
DM	0.622	0.563	0.164	-0.420	0.154	0.193	0.095	0.060	-0.126	0.008
PH (cm)	0.283	0.344	0.509	0.379	-0.095	-0.398	0.403	-0.257	-0.002	0.0007
EBT/m2	-0.115	0.438	0.435	-0.120	-0.654	-0.161	-0.273	0.247	-0.007	0.0007
PL	0.219	-0.201	0.500	0.480	-0.182	0.571	-0.119	-0.234	-0.000	-0.0001
FGP	0.414	-0.807	0.336	-0.087	0.029	-0.064	0.076	0.209	-0.006	-0.0320
UFGP	0.850	-0.034	-0.321	0.338	-0.023	-0.130	-0.184	0.035	0.010	0.0741
TGP	0.624	-0.722	0.198	0.026	0.018	-0.096	0.011	0.195	-0.002	-0.0056
SF %	-0.629	-0.389	0.500	-0.383	0.055	0.079	0.190	0.050	0.014	0.0740
TW (G)	-0.292	0.384	-0.021	0.661	0.128	0.168	0.235	0.477	-0.001	0.0029
GY (Kg/sq.m)	-0.145	0.277	0.588	0.149	0.581	-0.186	-0.400	0.010	0.007	-0.0034

DFF-days to 50% flowering, DM-days to maturity, PH-plant height, EBT/m²- Effective bearing tillers/m², PL-panicle length, FGP-filled grains per panicle, UFGP-unfilled grains per panicle, SF %- spikelet fertility %, TW-test weight, GY- grain yield

The greater portion of the variance (24.76%) was noticed in PC1and was strongly convinced DFF, DM, PH, PL, FGP, UFGP, TGP. Similarly, PC2 was influenced by DFF, DM, PH, EBT/m², TW and GY. Likewise, PC3 and PC4 are primarily influenced by UFGP, TW and DFF, DM, EBT/m², FGP, SF, respectively. Similar kind of results are in agreement with findings of Nachimuthu *et al.*, (2014), Allam *et al.*, (2017), Riaz *et al.*, (2018), Umadevi *et al.*, (2019), Sudeepthi *et al.*, (2020), Singh *et al.*, (2020), Pushpa *et al.*, (2021), Christina *et al.*, (2021), Dhanuja *et al.*, (2022), Wenkata Ratnam *et al.*, (2022), Lakshmi *et al.*, (2022), Mushtaq and Kumar (2023), Nayak *et al.*, (2023), in rice.

The interaction between the characters and the genotypes that perform better for the traits are depicted in the biplot diagram. The length of the vector for each trait represents its offering to total divergence, longer the vector length, more is the contribution of

concerned traits. The biplot depicted the relationship of 67 rice hybrids along with eight checks for 11 traits (Figure 3). The trait TG/P displayed greater vector length implying its contribution to the total divergence followed by DFF, SF %and FGP. These results are in congruence with the research findings of Lakshmi *et al.*, (2022), Tiwari *et al.*, (2022) and Gayathridevi *et al.*, (2023).

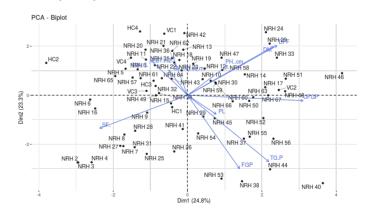


Figure 3: Biplot comprising of 67 rice hybrids along with eight hybrids studied for eleven yield and yield attributing traits



DFF-days to 50% flowering, DM-days to maturity, PH-plant height, EBT/m²- Effective bearing tillers/m², PL-panicle length, FGP-filled grains per panicle, UFGP-unfilled grains per panicle, TGP-total grains per panicle, SF %- spikelet fertility %, TW-test weight, GY- grain yield

The angle formed by the vectors of the traits indicates the association between the traits. A right angle (90°) between the vectors denotes no correlation, while an obtuse angle (>90°) denotes a negative correlation and an acute angle (<90°) between vectors suggests a positive correlation. All the traits studied displayed positive correlation with grain yield per plant except UFGP which noticed no correlation. From the biplots, the 11 yield and yield attributing traits were divulged into four groups. GY, TW and EBT were grouped in same cluster. DFF, DM and PH were grouped in same cluster. The traits UFGP, PL, TGP and FGP were grouped in another cluster. Whereas, SF % alone grouped as one cluster. The selection of hybrids with desirable highest score (0.588) for grain yield in PC3 will be desirable for developing high grain yielders in rice. The study showed that NRH 24, NRH 46, NRH 40, NRH 38, NRH 53, NRH 2; Hybrid checks HC2 (US 314), HC4 (HRI 174) and varietal check VC1 (BPT 5204) were located at extreme ends of distinct quadrants of the plot. Hence, theses hybrids and checks were recognized as highly divergent and found to be potential for exploitation in hybridization programme to enhance heterotic potential in rice crop. These results are in congruence with findings of Rahimi et al., (2013), Pandit et al., (2016), Sharafi et al., (2018), Divya et al., (2022), in rice.

Conclusion

PCA concluded that the first four PCs with eigen values more than one describing 24.76%, 23.26%, 14.54 and 13.22%, respectively attributed 75.80% of total variation. The cluster analysis exhibited

high genetic diversity, indicating a great chance for crop improvement by employing hybrids from other clusters. NRH 24, NRH 46, NRH 40, NRH 38, NRH 53, NRH 2 were identified as promising hybrids and can be used in developing diverse and heterotic inbred lines. Besides, the hybrids *viz.*, NRH 5 (49.89%), NRH 14 (28.54%), NRH 11(24.09%), NRH 16 (22.01%), NRH 18 (19.02%), NRH 36 (17.90%), NRH 27 (16.57%), NRH 47 (16.52%) were recognized as best heterotic hybrids for yield over best hybrid check HC4 (HRI 174) that governed by dominant genes and hence these hybrids can be advanced to evaluate under multi-location trails and further forwarded for commercial exploitation.

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RESEARCH ARTICLE

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Estimates of Heterosis, Inbreeding Depression and Transgressive Segregation in Rice (*Oryza sativa* L.) Under Sodic Soil

Shiv Prakash Shrivastav¹*, Verma OP¹ and Kuldeep Srivastava²

¹Department of Genetics and Plant Breeding, Acharya Narendra Deva University of Agriculture and Technology, Narendra Nagar (Kumarganj), Ayodhya -224 229 (UP), India.

²ICAR-Indian Institute of Vegetable Research, Varanasi (UP), India.

*Corresponding author Email: ms.shiv92@gmail.com

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Abstract

The present investigation was carried out at the Main Experimental Station of Acharya Narendra Deva University of Agriculture and Technology, Narendra Nagar (Kumarganj), Ayodhya (U.P.) India. A field experiment was conducted by using a line x tester set of 63 F₁s and 63 F₂s derived by crossing 21 rice genotypes/varieties as lines (females) with three testers (males) viz., Narendra Usar Dhan 3, CSR 23 and IR 28 with 2 check varieties (Jaya and CSR 43) of rice (Oryza sativa L.) in randomized complete block design with three replications to work out the heterosis, transgressive segregation and inbreeding depression effects for various attributes under the sodic soil condition. Among these, F₁s viz., NDRK 5037 x Narendra Usar Dhan 3, NDRK 5062 x IR 28, NDRK 5062 x CSR 23, NDRK 5037 x CSR 23 and NDRK 5040 x Narendra Usar Dhan 3 were showed significant positive standard heterosis for grain yield per plant over SV₁ and SV₂. All these crosses also had highly significant inbreeding depression for grain yield per plant in F₂ generation. Inspite of grain yield of these F₁s had significant heterosis and inbreeding depression for some of the other yield contributing characters also. This study indicated the presence of non additive gene action in the inheritance of grain yield per plant and some of the other yield contributing characters. Tolerant breeding populations showed similar banding pattern whereas susceptible exhibited similar banding pattern but possesses wide variations between tolerant and susceptible. At 35 kDa the medium to dark bands were present in parents, F₁s, F₂s, transgressive segregants and checks while in highly inbreeding depressed cross combinations, variable range of the bands were observed viz., absence of bands, light, medium and dark bands. The data offer a valuable resource for advancing the understanding and facilitating the utilization of additive and non-additive information for rice improvement.

Keywords: Rice, heterosis, transgressive segregation, inbreeding depression, SDS PAGE, protein profiling, breeding populations and sodic soil.

Introduction

Rice (*Oryza sativa* L.), is a staple food for more than 50% of the world population (Jiang *et al.*, 2020, Shrivastav *et al.*, 2023). It forms the breath of life '*prana*' for the human being. Rice is a high caloric food, which contain 75% starch, 6-7% protein, 2-2.5% fat, 0.8% cellulose and 5-9% ash. India has the largest area of 46.38 million hectare constituting

28.26% of the land under rice in the world and rank second in total production 130.29 million tonnes next to China with an average productivity of 2809 Kg/ha (DAC and FW, 2021-2022). It showed that the average productivity of rice is very low in our country. Therefore, there is immence need to develop high yielding, multiple resistance and wider adoptive



hybrid varieties. Taking the above points under consideration the present investigation was carried out to sort out the best heterotic hybrids for yield and its component characters. Heterosis is a common phenomenon in which an F_1 hybrid performs better than either inbred parent (Shrivastav *et al.*, 2022, Modunshim *et al.*, 2022).

Heterosis or hybrid vigor refers to the phenomenon that the heterozygous first filial generation (F₁) performs better than its parental inbred lines in target traits. With the development of the first commercial hybrid maize variety in the 1930s (Ayalneh 2020) and the development of rice hybrid varieties in the early 1970s in China, exploitation of heterosis in crop plants has achieved remarkable yield advantages over inbred lines and remains a crucial approach to increase agricultural production for global food demand in response to rapidly increasing global population and changing climate (Gu *et al.*, 2023).

In crop genetic research, the mechanism of heterosis has always been a key topic and several hypotheses have been proposed the dominance hypothesis, which proposes the masking of deleterious recessive parental alleles in the hybrid; the overdominance hypothesis, which attributes heterosis to the superiority of heterozygotes over parental homozygotes at individual loci; and the epistatic hypothesis, which postulates the contribution of positive epistatic interactions between non-allelic genes. However, there are no reports explaining the genetic mechanisms responsible for heterosis of rice grain quality traits (You *et al.*, 2022).

Inbreeding depression and heterosis are related phenomena of fundamental importance to evolutionary biology and applied genetics. Inbreeding depression refers to reduced fitness of progenies resulting from inbreeding (Stebbins 1958; Wright 1977). In contrast, heterosis, or hybrid vigor, is defined as the superiority of an F_1 hybrid over its parents (Stuber, 1994). Both heterosis and inbreeding depression are widely observed in both animal and plant kingdoms.

In evolution, inbreeding depression may contribute to formation of reproductive barriers between species and populations, while heterosis may be an important force in maintenance of genetic variation in populations. In applied genetics, exploitation of heterosis has played a major role in the genetic improvement of many crop plants and animals (Falconer, 1981; Stuber, 1994). Heterosis and inbreeding depression are considered two aspects of the same phenomenon (Falconer, 1981; Mather and Jinks, 1982). Heterosis is clearly related to heterozygosity, but it has long been debated how heterozygosity results in heterosis. Two predominant theories were proposed as the genetic basis of heterosis. The overdominance hypothesis (Shull, 1908; East, 1936) states that heterozygosity at single loci confers properties that are superior to either homozygote. In contrast, the dominance hypothesis (Bruce, 1910; Keeble and Pellew, 1910; Jones, 1917) proposed that dominant factors from either parent mask deleterious recessive mutations from the other parent in the heterozygous F₁. In both cases, the inbreeding depression is due to segregation and expression of deleterious recessive mutations in inbred progenies (Allard, 1960; Simmonds, 1979). A third, less widely embraced hypothesis suggests that heterosis may arise from epistasis between alleles at different loci (Stuber, 1994; Goodnight, 1999).

Transgressive segregation is common in plant breeding populations, where small minority of recombinants are outliers relative to parental phenotypes. While this phenomenon has been attributed to complementation and epistatic effects, the physiological, biochemical and molecular bases have not been fully illuminated. The phenomenon of transgressive segregation, which is observed in both natural and artificial populations created by plant breeding, is characterized by the occurrence of minority phenotypic outliers relative to parental range across segregating or recombinant population derived from genetically divergent parents. In addition to the classic explanations attributing



complementation and epistatic interactions as major mechanisms behind transgressive traits, the possible roles of coupling and uncoupling effects and genetic network rewiring have also been recently proposed (Vega and Frey, 1980; Rieseberg *et al.*, 1999; Dittrich-Reed and Fitzpatrick, 2013; de Los Reyes, 2019). Combined with the paradigms of genomic biology, the potential of transgressive individuals for enhanced yield of crops have been established, but its true potential for adaptive traits is yet to be determined (DeVicente and Tanksley, 1993).

The soil sodicity is a major factor that adversely affects the growth and yield of crop plant. Approximately one third of the land area on which rice grown is affected by salinity. Approximately 10% of the world's total land area (950 million ha), 20% of the world's arable land (300 million ha) and 50% of the total irrigated land (230 million ha) are affected by soil salinization. Further, it is expected to influence 50% of total cultivated land in 2050 at a dis-quieting rate. Every year almost 12 billion US\$ are globally lost due to salt stress that significantly affects the agricultural production (Shrivastav *et al.*, 2022a).

Materials and Methods

This experiment was carried out at the Main Experimental Station of A.N.D. University of Agriculture and Technology, Narendra Nagar (Kumarganj), Ayodhya (U.P.) India. The experimental material was based on a line x tester set of 63 hybrids (F₁s) and 63 F₂s developed by crossing 21 lines (females) viz., NDRK 5004, NDRK 5093, NDRK 5040, NDRK 5062, NDRK 5037, NDRK5025, NDRK 50059, NDRK 5081, NDRK 50047, NDRK 5039, IR 66946-3R-178-1-1 (FL 478), Sushk Samrat, IR 85897, Pant 10, CSR 10, Sarjoo 52, Narendra 2064, Narendra Usar Dhan 2, Deepak, Sundri and Pusa Basamati 1 with 3 testers (males) viz., Narendra Usar Dhan 3, CSR 23 and IR 28. An attempt was made to make a sixty three cross combinations during kharif season 2016 for F₁s production and grown in *kharif* season

2017 for F₂s production and *kharif* season 2017 again fress F₁s generated by crossing The 63 F₁s and 63 F₂s along with their parents including two checks, Jaya and CSR 43 (total set of 152 genotypes) were studied to work out the heterosis, transgressive segregation and inbreeding depression effects of their various attributes on grain yield under the sodic soil in randomized complete block design with three replications during Kharif 2018 (Arunachalam, 1974). Data on various attributes viz., days to 50% flowering, chlorophyll content, leaf nitrogen, leaf temperature, flag leaf area (cm²), plant height (cm), panicle bearing tillers per plant, panicle length(cm), spikelets per panicle, grains per panicle, spikelet fertility (%), biological yield per plant (g), harvest-index (%), L:B ratio, 1000-grain weight (g), amylose content, protein content and grain yield per plant (g) were recorded and analysed as per Kempthorne (1957). Heterobeltiosis and standard heterosis estimated as per Fonseca and Patterson (1968) and inbreeding depression as per Hill (1966). For the transgressive segregation the observations were recorded on fifteen randomly selected plants taken from each genotype of each replication. The per centage of superior segregants in particular cross is recorded by calculating the number of plants exceeding mean value of best check to the total number of plants in a cross.

The selected genotypes of different breeding populations were used to work out the SDS PAGE protein profiling of rice seed following method described by Laemmli, (1970).

Results and Discussions

The heterosis breeding has been used extensively in improving yield potential through development of hybrid cultivars in most of the allogamous crops and some autogamous crops like rice as well. The exploitation of heterosis for developing high yielding commercial hybrids in rice has been found highly fruitful inspite of its autogamous nature because significant heterosis is encountered in F₁ hybrids and successful and economical technology for commercial hybrid seed



production is available. A wide range of variation in the estimates of heterobeltiosis and standard heterosis was observed for grain yield per plant and other related traits. Top five F₁s that showed good heterotic potential for grain yield and yield contributing traits over Jaya (SV1) as well as CSR 43 (SV2) were NDRK 5037 x Narendra Usar Dhan 3, NDRK 5062 x IR 28, NDRK 5062 x CSR 23, NDRK 5037 x CSR 23 and NDRK 5040 x Narendra Usar Dhan 3 (**Table 1**). The findings will help promote rice improvements in context to establishment of heterotic patterns as a requirement for a sustainable long-term success of hybrid rice breeding (Shrivastav *et al.*, 2022a).

The estimates of inbreeding depression of eighteen characters of sixty-three crosses are presented in **Table 2.** The range of inbreeding depression for grain yield per plant was ranged from 4.39 (NDRK 5004 x Narendra Usar Dhan 3) to 38.74% (NDRK 5037 x CSR 23). The depressed cross was 38.74% (NDRK 5037 x CSR 23) followed by 31.98 (NDRK 5047 x Narendra Usar Dhan 3), 30.95 (Sundri x IR 28), 30.90 (NDRK 5004 x IR 28) and 30.56% (NDRK 5025 x CSR 23). Among 63 crosses all were positively significant while none had negatively significant inbreeding depression.

Table 1: Most promising crosses based on mean performance, heterobeltiosis and standard heterosis (SV₁ and SV₂), SCA effect, GCA effect of parent for grain yield / plant

S. No.	Crosses	per se Performance	Heterosis over better- parent	Heterosis over SV ₁	Heterosis over SV ₂	SCA effect	GCA effect of parent	Traits for which these the cross also exhibited desirable heterosis
1	NDRK 5004*	19.73**	26.47**	45.32**	36.10**	3.08**	НхН	CC, FLA, PL, S/P, G/P, SF, BY/P, HI,
1	NarendraUsarDhan 3	17.73	20.47	13.32	30.10	3.00	HAH	1000-GW, AC
2	Sarjoo 52 * NarendraUsarDhan 3	19.61**	25.41**	44.44**	35.27**	1.92**	НхН	LN, PBT/P, PL, S/P, G/P, SF, BY/P, HI, AC, PC
3	NDRK 5040* NarendraUsarDhan 3	19.48**	24.89**	43.51**	34.40**	3.68**	НхН	CC, S/P, G/P, SF, BY/P, HI,
4	NDRK 5062 * IR 28	18.92**	21.26**	39.33**	30.49**	2.10**	HxL	CC, PH, PL, S/P, G/P, BY/P, L/B
5	CSR 10* NarendraUsarDhan 3	18.65**	19.55**	37.37**	28.65**	0.63**	НхН	CC, PH, PBT/P, PL, S/P, G/P, SF, BY/P, HI
6	NDRK 5062* CSR 23	18.47**	18.38**	36.02**	27.39**	0.50*	HxA	CC, LN, PL, S/P, G/P, BY/P
7	Narendra 2064* Narendra UsarDhan 3	17.80**	7.81**	31.11**	22.79**	1.26**	HxH	CC, FLA, PH, PBT/P, S/P, G/P, BY/P, 1000-GW, AC
8	NDRK 5039* NarendraUsarDhan 3	17.61**	12.88**	29.71**	21.48**	1.73**	НхН	DF, CC, PL, S/P, BY/P, L/B, 1000-GW, AC
9	NDRK 5059* IR 28	17.12**	17.39**	26.10**	18.10**	3.35**	HxL	CC, LN, FLA, PH, PL, G/P, SF, BY/P, L/B, AC
10	NDRK 5037* NarendraUsarDhan 3	16.82**	7.84**	23.91**	16.05**	1.22**	HxH	CC, FLA, PH, PL, S/P, G/P, SF, BY/P, HI, L/B, PC

Traits: DF=Days to 50% flowering, CC=Chlorophyll content (spad value), LN= Leaf nitrogen (spad value), LT= Leaf temperature (spad value), FLA=Flag leaf area(cm²), PH=Plant height (cm), PBT/P=Panicle bearing tillers / plant, PL=Panicle length (cm), S/P=Spikelets / panicle, G/P= Grains per panicle, SF=Spikelet fertility (%), BY/P= Biological yield / plant (g), HI=Harvest index (%), L/B=L/B ratio, 1000-GW=1000- grain weight (g), AC= Amylose content, PC= Protein content and GY/P=Grain yield / plant(g)



Table 2: Estimates of inbreeding depression (%) in different yield and yield contributing traits of rice under sodic soil

Grain yield/ plant (g)	4.39**	17.59**	30.90**	21.54**	16.97**	15.50**	26.17**	26.49**	10.10**	23.17**	20.14**	18.27**	13.99**	11.30**	15.51**	12.88**	15.25**	23.80**	4.80**	8.28**	5.82**	4.42**	23.26**	21.51**	8.89**	25.26**	18.33**	15.65**	11.03**	12.64**	14.49**	27.00**	26.07**
Protein content (%)	0.63** 4.	0.58** 1	0.45** 30	0.51** 2	0.57** 10	0.53** 1:	0.26** 20	0.26** 20	0.28** 10	0.54** 2.	0.64** 20	0.66** 18	0.27** 1.3	0.30** 1.	0.53** 1.	0.56** 12	0.56** 1.	Н	1.17** 4.	0.66** 8	0.72** 5.	0.48** 4.	0.38** 2.	-	1.02** 8.	2.33** 2.	1.13** 18	0.38** 1.	0.32**	0.36** 1.	0.35** 1	0.39** 2'	0.36** 20
Amylose content	1.18** 0	0.99**	1.40** 0	1.08**	0.55** 0	1.26** 0	1.19** 0	2.20** 0	2.00** 0	1.90** 0	2.34** 0	3.66** 0	3.48** 0	3.49** 0	3.93** 0	2.86** 0	3.43** 0	3.46** 0	1.17** 1	0.84** 0	1.67** 0	1.43** 0	29.26** 0	0.84** 0	0.54**	0.43** 2	0.50** 1	1.30** 0	1.58** 0	1.35** 0	0 **09.0	12.44** 0	0.90** 0
1000-grain A weight (g)	0.51	0.13 0.	0.18	0.09	0.52 0.	0.41	0.15	0.56 2.	0.49 2.	0.20	0.22 2.	0.18 3.	0.18	0.31 3.	0.28 3.	0.37 2.	0.23 3.	0.22 3.	0.45	0.22 0.	0.37	0.16	0.24	0.21 0.	0.38 0.	0.35 0.	0.23 0.	0.41	0.12	0.29	0.21 0.	1.35** 1.	1.04*
L/B 1	0.72**	0.11	0.11	0.54**	0.42**	0.22**	0.32**	0.73**	0.54** (0.33** (-0.11 (3.73** (1.06**	1.53** (1.13**	-2.91**	-1.34** (-0.34**	-6.53**	0.59**	1.96**	-5.29**	_	-1.70** (0.28**	1.26**	2.82** (0.89**	-1.08** (0.86**	0.80**	0.76** 1	2.01** 1
Harvest index (%)	1.42 0	7.87** 0	29.14** 0	13.39** 0	2.97** 0	7.47** 0	20.29** 0	_	7.00** 0	19.73** 0	16.01**	14.35** 3	2.39	4.63**	-0.82	3.28**	11.75**	Н	-3.96**		3.58** 1	2.33	17.99**	19.50**	5.55** 0	21.62** 1	11.52** 2	9.13**	- 6.45**	12.03**	16.67** 0	24.99** 0	21.89** 2
Bio- logical F yield/ plant (g)	2.96** 1	10.61** 7	2.34* 2	9.45**	14.41** 2	8.62** 7	7.30** 2	2.52* 2	3.45** 7	4.12**	4.69**	4.30** 1.	11.93** 2	7.08** 4	16.07**	9.85** 3	4.00**	П	8.39**	3.13** 5	2.29* 3	2.19* 2	6.40**	2.46* 1	3.50** 5	4.48** 2	7.63** 1	7.09**	4.86** 6	0.68	-2.61** 1	2.70** 2.	5.26** 2
Spikelet 1. fertility (%)	4.17** 2.				-9.49** 14	-3.87** 8.	17.03** 7.	7.87** 2.			4.85** 4.	4.78** 4.	8.47** 11	7.62** 7.	8.87** 16	2.97* 9.			5.92** 8.	6.43** 3.		-1.43 2.	-3.65* 6.	-1.42		-3.03* 4.		2.96* 7.	-0.08 4.	5.39** 0.	-0.89		
Grains/ S _F fe	5.43** 4.1	3.86* 2.08)3 0.31	-12.18** 0.04	-5.00** -9.		19.61** 17	11.82** 7.8	3.83* 0.06	6.87** 2.33	7.97** 4.8	8.28** 4.7	18.81** 8.4	18.72** 7.6	17.03** 8.8	13.58** 2.9	15.78** 1.07		10.12** 5.9	10.11** 6.4	6.58** 3.39	7.67** -1.	6.46** -3.	8.17** -1.	7.86** 2.63	6.34** -3.	8.25** 0.18	7.46** 2.9	5.04** -0.	8.89** 5.3	5.10** -0.	6.40** 2.18	9.31** 0.40
Spike- G _i lets/ pa			2.03	-12.18** -12		00:00 *1		П					11.28** 118.	12.02** 18.	\vdash	10.93** 113.	14.90** 15.																
	1.33	1.76	1.71		4.01*	3.74*	3.04	4.34*	3.74*	4.63**	3.29	3.69*			**96.8		\vdash	\Box	4.49**	3.93*	3.32*	**86.8	9.75**	9.48**	5.34**	**96'8	7.95**	4.63**	5.09**	3.68*	5.88**	4.33*	**96.8
Panicle length (cm)	1.06*	1.81**	1.60**	2.75**	2.02**	1.90**	4.11**	2.93**	3.09**	2.23**	3.00**	2.17**	2.70**	2.23**	1.23*	1.63**	1.50**	3.21**	3.19**	2.39**	1.37**	2.04**	2.78**	2.87**	1.93**	2.15**	2.11**	2.13**	2.49**	2.32**	2.67**	1.96**	1.55**
Panicle bearing tillers/ plant	8.65**	10.03**	16.57**	8.78**	9.40**	10.06**	15.96**	15.78**	16.26**	17.16**	17.29**	17.49**	11.72**	10.12**	12.74**	10.59**	11.42**	12.31**	7.33**	7.73**	1.40**	10.14**	9.03**	11.50**	15.05**	15.92**	14.71**	14.91**	8.73**	13.61**	7.44**	7.39**	5.45**
Plant height (cm)	-0.93	-0.30	0.63	5.36**	99.0	1.34	-0.17	-1.14	-0.29	-1.31	-0.71	-1.22	-0.32	-0.59	-0.19	0.32	0.11	0.20	3.26**	3.48**	1.11	0.92	1.36	-0.60	0.39	-0.18	0.26	0.05	0.15	0.10	-0.18	99.0-	0.00
Flag leaf area (cm²)	0.92**	2.40**	0.86**	-1.05**	-1.05**	-1.03**	-2.19**	-5.48**	-2.22**	3.90**	2.38**	3.88**	0.74**	2.97**	1.47**	-2.92**	-0.55	-0.91**	-2.54**	-2.61**	0.00	3.25**	3.00**	4.09**	5.78**	3.47**	2.06**	2.88**	4.83**	3.86**	2.64**	2.77**	0.51
Leaf tem- perature	9.23**	8.53**	-0.29	1.55**	**02.9	-6.61**	5.56**	3.71**	7.96**	0.32	4.73**	-8.18**	10.07**	6.41**	1.81**	-3.92**	0.27**	6.55**	0.12	5.55**	1.49**	-4.16**	-5.21**	-1.42**	-6.71**	-0.55**	-6.20**	-7.92**	-4.00**	-5.04**	1.84**	-6.05**	0.02
Leaf nitro- gen	-12.08**	-8.80**	-0.62**	-17.72**	-19.06**	-23.28**	-8.44**	_	8.94**	-11.46**	-17.38**	-1.76**	9.30**	**86.9-		1.10**	-18.56**	*	-1.19**	0.00	0.55**	**96.6	20.44**	15.90**	-4.05**	-10.59**	-8.20**	-3.55**	-14.27**	-9.78**	-4.01**	-3.78**	-10.11**
Chlorophyll	4.90**	-6.85**	1.68**	26.96**	-20.78**	4.06**	13.68**	-7.09**	2.66**	-1.25**	-20.35**		7.96**	11.41**	8.58**	14.12**	17.06**		16.60**	6.48**	-1.00**	7.79**	16.70**	10.76**	34.47**	17.29**	17.61**	13.32**	12.62**	12.61**	9.92**	16.53**	18.31**
Days to 50% flower- ing	-2.01	-0.40	-1.54	-2.81	-0.78	-0.80	-0.79		0.39	-0.39	-3.23*	-2.37	-1.59	0.00	0.00	-2.79	-3.23*		0.79	-3.23*	-1.59	-1.99	-4.15**	-3.60**	-0.39	-2.33	-0.38	-2.02	-1.93	-1.59	-1.59	-2.42	-6.02**
Crosses	NDRK 5004*NarendraUsarDhan 3	NDRK 5004*CSR 23	NDRK 5004*IR 28	FL478*NarendraUsarD- han 3	FL478*CSR 23	FL478*IR 28	NDRK 5093*Narendra U sarDhan 3	23		SushkSamrat*Naren- draUsarDhan 3	SushkSamrat*CSR 23	SushkSamrat*IR 28	IR 85897*NarendraU-sarDhan 3	IR 85897*CSR 23	IR 85897*IR 28	Pant 10*NarendraU- sarDhan 3	Pant 10*CSR 23		CSR 10*NarendraU-sarDhan 3	CSR 10*CSR 23	CSR 10*IR 28	NDRK 5040*Naren- draUsarDhan 3	NDRK 5040*CSR 23	NDRK 5040*IR 28	Sarjoo 52*NarendraU-sarDhan 3	Sarjoo 52*CSR 23	Sarjoo 52*IR 28	NDRK 5062*Naren- draUsarDhan 3	NDRK 5062*CSR 23	NDRK 5062*IR 28	Narendra 2064*NarendraUsarDhan 3	*CSR 23	Narendra 2064*IR 28
S. S.	-	7	3	4	w	9	7	∞	6	10	=	12	13	14	15	16	17		19	20	21	77	23	54	25	56	27	78	56	30	31	32	33



34 NDRK 5037*Naren-	-1.59	5.95**	13.64**	-2.32**	2.46**	-0.62	8.73**	2.85**	10.16** 8	8.56**	-1.78	2.47**	20.44**	0.53**	0.27	0.73**	2.55**	22.43**
35 NDRK 5037*CSR 23	-4.38**	14.86**	9.17**	2.35**	4.20**	0.21	0.00	1.93**	8.22**	11.06**	3.09*	6.21**	34.58**	1.31**	99.0	0.71**	0.51**	38.74**
36 NDRK 5037*IR 28	**44**	10.14**	8.80**	-2.47**	3.07**	92.0-	9.26**	1.77**	11.17** 1	11.66** 0	0.55	3.80**	27.67**	1.37**	0.31	0.28**	0.52**	30.40**
37 NDRK 5025*Naren- draUsarDhan 3	-5.18**	21.27**	-2.86**	-6.12**	0.12	0.54	13.52**	2.11**	8 **60.01	8.13**	-2.16	9.86**	14.39**	1.42**	1.11*	1.25**	0.59**	22.77**
38 NDRK 5025*CSR 23	-1.21	21.94**	-2.38**	-5.81**	2.00**	80.0	14.35**	1.73**	12.36** 1	10.90**	-1.67	6.25**	26.00**	1.53**	0.53	1.43**	0.46**	30.56**
39 NDRK 5025*IR 28	-2.38	15.42**	7.15**	-6.34**	1.34**	0.16	9.42**	-	\neg	-		10.22**	-	-4.60** (0.31	0.88**	0.40**	20.45**
40 NDRK 5059*Naren-draUsarDhan 3	-1.57	14.95**	-5.37**	1.66**	1.15**	-0.06	13.45**	2.07**	16.39** 1	19.80** 4	4.08**	12.12**	19.79**	0.84**	0.62	0.66**	0.24**	29.53**
41 NDRK 5059*CSR 23	-1.22	16.29**	0.56**	-3.78**	2.32**	-0.74	16.22**	2.27**	11.24** 1	14.97** 4	4.20**	6.92**	0.84	0.44**	-0.63	0.55**	0.23**	7.74**
42 NDRK 5059*IR 28	-1.58	16.13**	-3.97**	-6.20**	2.18**	0.00	14.91**	2.01** 1	15.35** 2	20.35** 5	5.96**	8.57** (0.46	0.92**	0.16	0.48**	0.31**	9.02**
43 NDRK 5081*Naren-draUsarDhan 3	-4.45**	18.79**	8.82**	-1.53**	-5.96**	-0.83	12.04**	2.99**	19.19** 2	23.93** 5	5.86** 1	12.95**	15.91**	-2.73**	0.19	0.82**	0.33**	26.81**
44 NDRK 5081*CSR 23	-4.03**	18.42**	15.74**	-3.92**	1.57**	-0.55	13.50**	2.04**	19.92**	29.20** 1	11.58**	11.11**	**62.9	1.33**	0.35	1.20**	0.31**	17.19**
45 NDRK 5081*IR 28	-9.36**	19.84**	15.65**	-0.32	3.02**	-0.30	13.26**	2.10**	20.49** 2	29.81** 1	11.72** 5	5.67**	17.09**	0.14*	0.44	0.59**	0.35**	21.75**
46 NDRK 5047*NarendraUsarDhan 3	-3.17*	25.76**	3.86**	-4.26**	3.06**	0.22	4.70**	2.74**	8 **29.5	8.52** 3	3.05*	1.60	30.82**	0.72**	0.07	0.81**	0.61**	31.98**
47 NDRK 5047*CSR 23	4.44**	19.89**	33.77**	**69.0-	1.02**	0.03	9.21**	1.91**	4.68**		14.00**	6.67**	_	1.40**	0.14	0.52**	0.72**	16.45**
48 NDRK 5047*IR 28	-3.57**	12.80**	23.94**	-1.22**	2.62**	0.45	9.19**	3.02**	3.82* 5	9.07** 5	5.45** 7	7.94**	15.79**	1.10**	0.23	0.81**	0.70**	22.33**
49 NDRK 5039*Naren-draUsarDhan 3	0.00	9.23**	23.45**	5.60**	0.21	-0.39	14.19**	2.90**	4.89**	22.93** 1	18.92**	-2.67*	20.14**	0.19**	0.36	0.71**	0.45**	18.03**
50 NDRK 5039*CSR 23	-1.20	23.99**	26.81**	-7.48**	2.76**	0.40	18.24**	1.91**	4.34**	7.00** 2	2.78	5.22**	12.02**	-1.15**	0.20	0.59**	0.47**	16.66**
51 NDRK 5039*IR 28	-1.19	24.43**	**06.61	**96.0	1.68**	0.05	17.20**	1.13*	3.85* 3	3.57	-0.36	10.24**	17.80**	1.50**	0.58	0.76**	0.49**	26.11**
52 NarendraUsarDhan 2*NarendraUsarDhan 3	-2.38	13.23**	-11.11**	0.26	2.56**	-0.43	7.47**	1.84**	6.71**	7.14** 0	0.44	2.31*	10.14**	-0.22**	0.30	0.43**	0.77**	12.39**
53 NarendraUsarDhan 2*CSR 23	-2.79*	17.13**	9.10**	1.88**	1.73**	-0.79	**60.8	2.86**	7.37**	-5.14**	-13.51** 4	4.10**	9.81**	0.78**	0.81	0.76**	0.31**	13.72**
54 NarendraUsarDhan 2*IR 28	-1.21	10.37**	-64.65**	4.16**	2.61**	-0.14	8.16**	2.64**	13.03** 1	10.65**	-2.75	5.88**	6.44**	0.76**	0.30	1.01**	0.23**	12.01**
55 Deepak*NarendraU-sarDhan 3	-2.01	-2.76**	-34.29**	0.47	4.22**	0.24				5.82**	-5.40** 4		15.36**	1.51**	0.61		0.27**	19.16**
56 Deepak*CSR 23	-2.40*	0.92**	-36.24**	1.23**	2.16**	-1.80	10.76**	1.65**	10.43** 1	14.07** 4	4.04**	9.57**	5.36**	-0.22**	09:0	0.65**	0.35**	14.53**
57 Deepak*IR 28	-2.83*	-1.78**	-19.27**	1.47**	-0.45	-1.31	8.83**	2.48**	11.28** 5	9.65**	-1.83	15.69**	11.86**	2.22**	0.22	0.86**	0.22**	25.96**
58 Sundri*NarendraUsarD- han 3	-4.78**	8.11**	-18.24**	-1.31**	0.43	0.00	11.07**	1.87**	13.13** 1	13.62** 0	0.57	- 19.05**	-0.16	0.37**	0.44	0.61**	1.09**	19.01**
59 Sundri*CSR 23	-2.72*	9.04**	-1.36**		2.60**	-0.45	10.87**	1.72** 1	11.65** 1	15.21** 4	4.06** 1	18.26** 5	5.94** (0.75** (0.47	0.95**	**66.0	23.09**
	-3.57**	6.58**	7.94**	1.19**	4.17**	0.15			*		-2.35				0.92*		0.92**	30.95**
61 Pusa Basmati 1*Naren-draUsarDhan 3	-1.74	10.18**	4.90**		2.05**	0.42	10.28**		4.03*	15.02** 1	11.43** 4	4.46**	18.70**	1.23**	0.26		0.13**	22.34**
62 Pusa Basmati 1*CSR 23	-1.02	17.73**	1.82**	2.38**	3.23**	-0.63	$\overline{}$	1.92**	14.06** 2	\rightarrow	11.20** 1			0.41**	0.18	0.70**	0.17**	19.61**
63 Pusa Basmati 1*IR 28	-4.00**	17.24**	1.81**	3.55**	3.81**	0.28	10.59**	2.76**	\rightarrow	19.93** 7	\rightarrow	4.40**	22.31** (0.53**	0.47	**69.0	0.46**	25.63**
Range of Min.	-9.36	-20.78	-64.65	-8.18	-5.96	-1.80	0.00	1.06	-12.18	-12.18	-13.51	-2.67	-3.96	-6.53	-0.63	0.28	0.13	4.39
Inbreeding Max. depression	0.79	34.47	33.77	10.07	5.78	5.36	18.24	4.12	20.49	29.81	18.92	19.05	34.58	3.73	1.35	29.26	2.55	38.74
No. of crosses with significant positive inbreeding depression	0	54	27	29	45	3	62	63	57	28	30	59	99	4	4	63	63	63
No. of crosses with signif- icant negative inbreeding depression	22	6	35	28	11	0	0	0	Т	8	9	7	1	16	0	0	0	0



Majority of the crosss combinations possessing high heterosis also had high estimates of inbreeding depression. Further, it indicated that both the heterosis as well as inbreeding depression is closely related phenomenon with preponderance of non additive gene action. The similar findings have also been reported by Alam *et al.*, (2004), Verma and Srivastava (2005), Sharma *et al.*, (2013) and Venkanna *et al.*, (2014). The presence of high heterosis for economically important characters is not only useful for developing hybrids, synthetic or composites through exploitation of heterosis, but also helps in obtaining transgressive segregants for development of superior homozygous lines.

In genetics, transgressive segregation is the formation of extreme phenotypes, or transgressive phenotypes, observed in segregated hybrid populations compared to phenotypes observed in the parental lines. The transgressive segregation was estimated as appearance of these transgressive (extreme) phenotypes either positive or negative in terms of fitness. The estimates of transgressive segregation for eighteen characters of sixty-three crosses are presented in **Table 3**.

For grain yield per plant twenty-one crosses over better parent, twenty-eight crosses over SV_1 and nineteen crosses over SV_2 exhibited positive and significant Transgressive segregation, due to increased *per se* performance in F_2 generation. As such selection of promising lines in segregating generation, especially to pick up the transgressive segregants by maintaining more

number of progenies would be best perspective method to make progress for this trait. The top five transgressive segregants over better parent were NDRK 5004 x Narendra Usar Dhan 3 (26.47), Sarjoo 52 x Narendra Usar Dhan 3 (25.41), NDRK 5040 x Narendra Usar Dhan 3 (24.89), NDRK 5062 x IR 28 (21.26) and CSR 10 x Narendra Usar Dhan 3 (19.55) in grain yield per plant and these same crosses have best heterotic potential as well as high amount of inbreeding depression; it clearly indicated that preponderance of non additive gene action (additive x additive) in such type of cross combinations and showing transgressive segregation as well due to heritable and fixable nature of gene action. These results are similar to those of Verma and Srivastava (2005), Saleem et al., (2008) and Seetharam et al., (2013). For amylose content none of the crosse over better parent, twenty-four crosses over SV, and forty-two over SV, exhibited positive and significant residual heterosis, due to increased per se performance in F₂ generation. As such selection of promising lines in segregating generation, especially to pick up the transgressive segregants by maintaining number of progenies would be more persepective method to make progress for this trait. For protein content three crosses over better parent, forty-five over SV₁ and nine over SV₂ exhibited positive and significant residual heterosis. As such selection of promising lines in segregating generation, especially to pick up the transgressive segregants by maintaining number of progenies would be more persepective method to make progress for this trait. In F₂s segregating population, some of the crosses were found as transgressive segregants.



Table 3: Extent of transgressive segregation in F_2 s for 18 characters in rice under sodic soil

S. No.	Crosses	Days to 50% flow- ering	Chlo- rophyll content	Leaf ni- trogen	Leaf tem- perature	Flag leaf area (cm²)	Plant height (cm)	Panicle bearing tillers/plant	Panicle length (cm)	Spike- lets/ panicle	Grains/ panicle	Spikelet fertility (%)	Biologi- cal yield/ plant (g)	Harvest In- dex(%)	L/B ratio	1000-grain weight (g)	Amylose	Protein content (%)	Grain yield/ plant (g)
-	NDRK 5004*Nar- endraUsarDhan 3	-1.93	8.08	3.73	-10.49**	-0.38	-7.39**	1.60	-0.53	-2.63	-6.36**	-2.99	-2.96	26.33**	-21.56**	2.34	-0.31	-5.34**	26.47**
7	NDRK 5004*CSR 23	-3.09	-9.64	0.00	-5.68*	1.12	-1.47	0.64	-2.26	7.21**	-0.57	-11.22	3.51	-17.45**	-0.43	3.22	-0.24	-1.66*	0.59
6	NDRK 5004*IR 28	1.54	-15.34*	-0.62	-7.53**	-0.54	-7.33**	-8.01 **	-2.26	10.58**	-0.59	-12.70	9.65**	-14.01**	-3.64	2.53	-0.59	-2.92**	-5.84*
4	FL478* NarendraU-sarDhan 3	1.59	-20.53**	4.40	-6.37*	-8.23**	**99.6-	2.46	-3.35	-23.25**	-25.67**	-2.30	-14.81**	-1.91	-25.47**	1.04	-30.56 **	-0.02	-1.32
w	FL 478*CSR 23	3.60	-8.65	13.84	-3.98	1.53	5.23**	1.76	-3.35	-18.49**	-16.24**	-0.40	-3.81	1.26	4.08	89:0-	-40.94 **	0.56	0.40
9	FL 478* IR 28	0.40	-19.57**	13.21	-1.05	-9.11**	0.44	0.70	-3.98	-24.82**	-22.79**	1.94	0.95	-8.70*	1.43	0.51	-33.12 **	0.59	-7.85**
7	NDRK 5093*Nar- endra U sarDhan 3	0.39	-12.33*	-1.10	-2.61	-11.02**	2.21*	** 09.7-	-1.77	-23.03**	-38.88**	-19.83	-5.93*	-17.17**	-24.82**	-13.35**	-39.83 **	-2.40**	**09'9-
∞	NDRK 5093*CSR 23	-1.57	31.09**	-13.74	-3.55	8:00	0.51	** 68.7-	-5.57*	-11.53**	-21.37**	-11.34	19.59**	-29.49**	96:0	-5.96*	-49.40 **	0.61	-15.65**
6	NDRK 5093*IR 28	1.57	24.08**	-4.95	-5.56*	-15.14**	5.04**	-9.65 **	-6.39**	8.73**	-0.79	-22.70	24.44**	-8.85*	-2.56	-13.71**	-43.24 **	-0.72	13.55**
10	SushkSamrat*Nar- endraUsarDhan 3	-0.77	-19.49**	0.54	-6.93*	-4.69**	4.08**	-10.83 **	-0.99	-9.65**	-17.11**	-7.45 **	-31.11**	-14.76**	-26.36**	-0.42	-43.78 **	-11.68**	-33.06**
=	SushkSamrat*CSR 23	-1.54	1.55	2.72	*67-9-	-4.52**	-0.42	** 09.8-	-4.51	3.26	-4.56**	-7.80 **	25.77**	-13.59**	*06.9-	4.32*	-52.63 **	-9.03	8.49**
12	SushkSamrat*IR 28	-0.38	-14.84**	-5.98	**89.8-	-3.98**	5.20**	** 28.6-	1.41	0.00	-13.71**	-12.87	9.88*	-12.28**	9.95**	2.92	-47.53 **	-9.91	-3.92
13	IR 85897*NarendraUsarDhan 3	-1.92	-13.80*	-11.70	-6.92*	-0.78	2.16*	-18.35 **	-2.96	-12.06**	-19.80**	-8.00 **	-28.89**	9.14*	-24.33**	2.65	-69.71 **	0.17	-14.06**
14	IR 85897*CSR 23	-4.21 *	-2.91	-2.13	*/0.9-	-3.52**	2.16*	-7.56 **	-5.66*	-2.76	**69.6-	-7.36 **	8.25*	-0.93	12.79**	3.56	-74.40 **	1.79*	7.36**
15	IR 85897*IR 28	-1.53	-0.12	-6.38	-4.77	-1.32	2.43*	0.00	-2.43	-0.79	-1.29	-2.99	5.62	-3.84	5.46	3.04	-71.34 **	1.47	1.14
16	Pant 10*Naren- draUsarDhan 3	0.39	-20.90**	-0.55	-3.70	-12.53**	-0.32	-17.63 **	3.15	-14.25**	-19.07**	4.80 **	-11.85**	18.25**	-36.78**	-5.48*	-71.91 **	0.25	4.17
17	Pant 10*CSR 23	-0.39	-8.72	8.79	-8.12**	4.28*	-1.40	** 96.6-	3.41	-5.51*	-5.70**	-0.46	6.19	-26.09**	1.97	-1.83	-75.97 **	0.24	-8.54**
18	Pant 10*IR 28	-1.95	-1.24	4.95	-5.70*	-15.79**	9.40**	4.73	-1.18	2.08	3.49*	-1.44	2.65	-21.17**	10.33**	-5.28*	-73.34 **	0.26	0.58
19	CSR 10*Naren- draUsarDhan 3	-0.40	-12.58*	-7.57	-1.97	-15.27**	-9.66**	2.02	2.50	29.85**	28.12**	-1.13	-2.96	23.26**	-41.58**	0.43	-25.52 **	-5.04**	19.55**
20	CSR 10*CSR 23	2.40	19.47**	-6.49	-5.04	4.37*	5.72**	-0.29	2.06	22.44**	35.33**	0.48	-6.06*	-12.54**	-9.27*	2.82	-36.69 **	-1.47	5.20*
21	CSR 10*IR 28	2.00	20.09**	-2.70	-4.44	-18.94**	0.48	1.15	\$.59*	27.02**	31.25**	-1.57	-3.03	11.97**	80.9	8.39**	-28.84 **	-2.79**	13.48**
22	NDRK 5040*Nar- endraUsarDhan 3	1.59	2.63	-13.81	-4.60	-9.05**	-1.64	-7.15 **	-2.95	-2.19	-2.93*	-1.82	-0.74	21.82**	-38.49**	-3.53	-62.33 **	-1.41	24.89**
23	NDRK 5040*CSR 23	-0.40	-16.40**	-14.76	-2.75	-2.01	-2.82**	3.56	-2.33	9.02**	11.40**	-0.86	20.62**	-22.63**	**/9.6	0.21	-77.12 **	0.83	-6.63**
24	NDRK 5040*IR 28	2.78	-14.58**	-11.90	-4.40	-13.34**	-0.30	0.40	-5.28	14.44**	11.41**	-2.58	22.68**	-19.15**	10.85**	-0.23	-64.42 **	-0.20	-0.86



Crosses	Days to 50% flow- ering	Chlo- rophyll content	Leaf ni- trogen	Leaf tem- perature	Flag leaf area (cm²)	Plant height (cm)	Panicle bearing tillers/plant	Panicle length (cm)	Spike- lets/ panicle	Grains/ panicle	Spikelet fertility (%)	Biologi- cal yield/ plant (g)	Harvest In- dex(%)	L/B ratio	1000-grain weight (g)	Amylose content	Protein content (%)	Grain yield/ plant (g)
Sarjoo 52*Naren- draUsarDhan 3	-1.15	-18.72**	7.29	-3.33	-6.61**	0.45	** 90.7-	66.0	18.18**	21.70**	-0.77	-2.82	22.97**	-41.99**	-2.30	0.92	09.0	25.41**
Sarjoo 52*CSR 23	0.38	-8.05	-2.08	-9.03**	-2.91	0.24	-6.76 **	-3.26	31.31**	29.01**	** 44.4	**98.6-	-18.73**	-6.49	-0.88	1.27	3.77**	-6.61**
Sarjoo 52*IR 28	1.15	-17.34**	3.13	-2.30	-7.72**	1.11	-6.18 **	-1.28	30.91**	31.13**	-0.27	-14.79**	9.13*	-4.23	09:0-	1.11	0.64	-3.11
NDRK 5062*Nar- endraUsarDhan 3	-0.78	-16.45**	7.94	-0.68	-4.12*	0.83	1.46	0.91	-5.50**	-12.08**	-7.85 **	-2.96	0.25	-44.67**	-0.18	-3.82	1.45	6.67**
NDRK 5062*CSR 23	3.53	1.09	10.05	-4.08	-3.90*	0.33	0.58	1.42	-11.01**	-29.38**	-20.96	11.38**	-4.11	6.49	-0.54	-17.94 **	0.51	18.38**
NDRK 5062*IR 28	0.00	66.0	1.06	-1.70	-7.93**	-0.48	1.75	3.36	-8.81**	-23.13**	-16.07	18.70**	2.17	8.73*	1.46	-8.01 **	0.59	21.26**
Narendra 2064*Na- rendraUsarDhan 3	0.00	-19.01**	-1.62	-2.78	-1.93	3.48**	0.54	-1.45	21.36**	-0.22	-17.42	16.30**	-23.08**	-18.96**	5.50**	2.51	0.15	7.81**
Narendra 2064*CSR 23	-0.78	-24.54**	3.78	1.86	-1.87	2.93**	1.35	1.74	23.55**	1.12	-17.82**	28.57**	-32.19**	11.35**	2.12	-9.01 **	0.49	-12.82**
Narendra 2064*IR 28	3.13	-25.34**	5.95	-5.19	0.33	2.55*	2.96	1.74	21.76**	-6.47**	-22.87**	28.57**	-35.32**	18.77**	4.68**	2.58	0.48	-16.94**
NDRK 5037*Nar- endraUsarDhan 3	-2.30	-12.10*	-13.64	-2.64	-0.73	-2.78*	0.88	-2.74	30.71**	30.13**	-0.71	16.18**	-7.85	-23.03**	-22.05**	-4.15	-0.66	7.84**
NDRK 5037*CSR 23	0.38	-20.30**	-5.05	-8.81**	-2.70*	-1.35	0.58	-3.48	32.82**	28.85**	-3.25*	11.03**	-40.43**	8.76**	-10.41**	-18.44 **	1.51*	-18.46**
NDRK 5037*IR 28	-0.77	-9.59*	-16.16	-4.74	1.11	0.47	0.29	-6.43**	25.14**	23.08**	-1.92	11.76**	-21.83**	19.07**	-22.59**	-9.10 **	1.37	-11.98**
NDRK 5025*Nar- endraUsarDhan 3	4.76*	-19.86**	-6.25	-3.93	-20.18**	-0.38	0.00	-0.13	38.51**	34.20**	-2.40	-5.19	-31.63**	-43.69**	-14.57**	-13.54 **	-5.27**	-14.17**
NDRK 5025*CSR 23	0.00	-22.39**	-10.42	-2.82	-8.91**	0.14	0.00	0.38	38.07**	32.07**	-3.65*	17.65**	-40.97**	2.12	-3.94	-27.05 **	-1.96*	-30.53**
NDRK 5025*IR 28	3.20	-15.32**	-11.98	-4.67	-24.99**	-0.41	-0.27	92.0	35.01**	28.27**	-4.30**	20.59**	-21.49**	-6.75	-12.54**	-18.54 **	-3.21**	-5.43*
NDRK 5059*Nar- endraUsarDhan 3	0.39	-10.41*	7.69	-6.54*	**98.6-	-1.85	-4.07*	4.11	-0.44	-3.91**	-2.65	-14.07**	-26.88**	-42.39**	3.87	1.92	-1.51	-20.66**
NDRK 5059*CSR 23	-3.49	-11.51*	-2.75	-4.18	-1.54	-2.44*	-6.23**	1.03	9.91**	8.29**	-4.62**	21.00**	-12.68**	-10.33**	3.83	1.78	0.04	5.65*
NDRK 5059*IR 28	-0.39	-16.83**	0.55	-2.49	-12.99**	-1.72	**69.5-	0.15	-1.18	-0.55	1.42	28.00**	-8.37*	13.36**	3.19	1.95	0.11	17.39**
NDRK 5081*Nar- endraUsarDhan 3	-1.15	-17.86**	-10.14	-3.93	-32.01**	-14.01**	0.80	-2.73	-12.28	-17.60**	-5.22**	-10.37**	-22.05**	-48.01**	-1.23	-4.07	89.0-	-12.52**
NDRK 5081*CSR 23	-1.15	-14.19**	-12.08	-1.82	-19.81**	-3.25*	0.27	-1.77	-0.25	-12.25**	-12.25**	31.96**	-14.47**	-1.72	-1.98	-18.51 **	0.84	16.26**
NDRK 5081*IR 28	-1.53	-16.07**	-19.32*	-4.47	-37.76**	-0.40	-1.06	1.77	-0.26	0.00	-8.44**	43.01**	-23.57**	-3.57	-2.00	-9.18 **	0.77	9.28**
NDRK 5047*Nar- endraUsarDhan 3	3.17	-15.51**	-16.67*	-3.81	-9.33**	0.28	1.75	1.61	-12.50	-18.58**	-11.19**	**68.8-	-26.85**	-33.12**	-5.50**	-5.19	08.0	-27.86**
NDRK 5047*CSR 23	3.60	-7.48	-29.05**	-6.97*	86:0	-1.67	0.58	-0.81	-3.01	-9.78**	-11.18**	29.90**	-12.34**	-5.48	-6.42**	-19.48 **	-0.06	13.99**
NDRK 5047*IR 28	4.40*	-0.12	-22.86**	-4.03	-10.92**	0.73	0.87	4.51	0.00	-10.34**	-9.59**	28.89**	-22.16**	-1.58	-4.67	-9.65 **	0.93	0.24



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S.S.	Crosses	Days to 50% flow- ering	Chlo- rophyll content	Leaf ni- trogen	Leaf tem- perature	Flag leaf area (cm²)	Plant height (cm)	Panicle bearing tillers/plant	Panicle length (cm)	Spike- lets/ panicle	Grains/ panicle	Spikelet fertility (%)	Biologi- cal yield/ plant (g)	Harvest In- dex(%)	L/B ratio	1000-grain weight (g)	Amylose content	Protein content (%)	Grain yield/ plant (g)
49	NDRK 5039*Nar- endraUsarDhan 3	-1.59	-0.42	-27.60**	-7.44**	-9.87**	2.36*	-7.64**	2.47	6.58**	-11.25**	-16.39**	14.07**	-1.05	-15.70**	6.02**	3.65	-1.75*	12.88**
20	NDRK 5039*CSR 23	0.40	-21.85**	-27.15**	-5.47	1.62	0.64	-9.72**	2.94	21.55**	17.38**	-4.96**	13.39**	-13.13**	0.31	4.51*	3.45	-0.05	13.77**
51	NDRK 5039*IR 28	1.99	-22.35**	-28.96**	-3.23	-13.34**	-2.23*	-9.72**	2.94	23.12**	-6.63**	-23.50**	1.79	-17.27**	16.18**	2.39	3.31	0.02	-14.44**
52	NarendraUsarDhan 2*NarendraUsarD- han 3	1.98	6.84	-16.67*	-4.72	-8.90**	-2.12*	2.22	-6.22**	-14.69**	-20.54**	-6.03**	-5.93*	-8.93*	-26.77**	-0.25	3.72	-2.73**	-7.71**
53	NarendraUsarDhan 2*CSR 23	1.98	0.51	-12.04	-7.57**	0.41	-4.57**	0.95	-8.11**	0.75	-0.85	-1.81	6.36	-22.98**	-2.54	-0.03	3.64	0.84	-7.92**
25	NarendraUsarDhan 2*IR 28	-1.19	8.56	-11.57	-5.03	-10.44**	-3.70**	0.00	-5.41*	-7.32 **	-7.36**	-2.22	1.82	-0.51	0.11	0.86	3.97	-0.21	1.21
55	Deepak*Naren- draUsarDhan 3	-1.17	-25.95**	21.85	0.71	-37.05**	-17.27**	1.06	2.52	-7.89 **	-12.96**	-4.96**	-20.74**	-7.41	-25.47**	2.12	-8.63 **	-0.70	-26.18**
99	Deepak*CSR 23	-0.39	-4.67	10.19	-3.06	-21.94**	6.64**	-0.70	2.36	3.26	-4.27**	-8.54**	7.22	-8.25*	-16.25**	2.07	-22.43 **	-0.01	-1.86
57	Deepak*IR 28	-1.17	-6.09	8.05	-0.94	-37.71**	0.34	1.41	-1.10	10.54**	-7.21**	-15.29**	-5.49	-14.78**	-26.69**	2.60	-13.12 **	90.0	-19.76**
58	Sundri*NarendraU-sarDhan 3	4.37*	-29.60**	7.36	-3.93	-33.34**	-4.69**	-29.19**	2.60	-24.56	-31.78**	-8.78**	-24.44**	12.33**	-13.34**	-2.11	4.66	-5.10**	-13.61**
59	Sundri*CSR 23	6.02**	-8.33	-9.20	*09.9-	-17.26**	-2.57*	-17.97**	3.90	-18.30**	-25.36**	-8.87**	-3.09	-15.01**	-14.47**	-4.90*	-0.60	-2.15**	-17.58**
60	Sundri*IR 28	4.40*	-6.28	-7.36	-6.36*	-36.41**	-0.35	-12.26**	-1.30	-8.58 **	-4.35*	-6.92**	22.22**	-28.95**	5.07*	-5.82**	4.96	-3.16**	-13.68**
61	Pusa Basmati 1*NarendraUsarD- han 3	2.82	-12.30*	-17.11	-5.76*	-38.41**	-20.11**	-17.99**	-5.13*	-11.18**	-30.81**	-21.78**	-20.74**	-8.68*	-21.40**	-7.74**	1.79	-2.11**	-27.82**
62	Pusa Basmati 1*CSR 23	4.23*	4.77	-13.37	-7.32*	-28.18**	-5.80**	-8.76**	-10.12**	-6.52 **	-26.50**	-22.60**	13.40**	-29.14**	-2.52	4.81*	1.74	0.34	-19.78**
63	Pusa Basmati 1*IR 28	0.70	5.13	-12.83	-5.98*	-41.76**	-7.25**	4.73	-1.91	1.70	-22.71**	-23.64**	10.13*	-35.17**	4.83	-7.72**	1.74	-0.63	-24.33**
Range	ge Min.	-4.21	-29.60	-29.05	-10.49	-41.76	-20.11	-29.19	-10.12	-24.82	-38.88	-23.64	-24.44	-40.97	-48.01	-22.59	-77.12	-11.68	-30.53
of trans- gressive segrega- tion	ans- sive ega-	6.02	31.09	21.85	1.86	5.09	9.40	4.73	5.59	38.51	35.33	1.94	43.01	26.33	19.07	8.39	4.96	3.77	26.47
No. of cr significar regation	No. of crosses with significant positive seg- regation	9	4	0	0	8	15	0	1	25	17	0	29	6	12	9	0	9	21
No. c signi segre	No. of crosses with significant negative segregation	1	35	∞	23	43	19	26	6	22	37	41	15	43	27	16	36	16	30

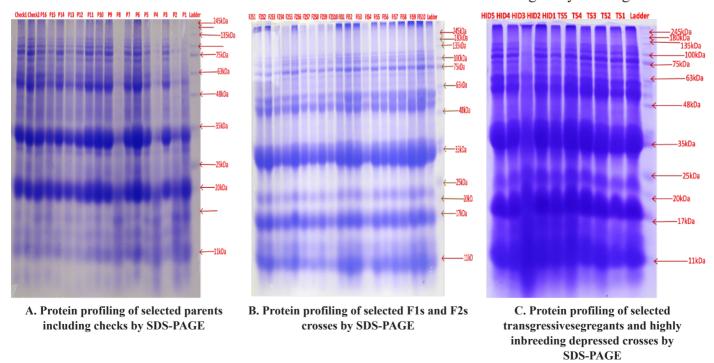
 $^{\ast},\,^{\ast\ast}$ significant at 5 and 1 per cent probability levels, respectively



The most promising crosses based on mean performance, heterobeltiosis and standard heterosis (SV1 and SV2), SCA effect, GCA effect of parent for grain yield per plant and the traits for which these crosses also exhibited desirable heterosis in F₁s and F₂s have been depicted in **Table 1**. The ten most promising crosses viz., NDRK 5037 x Narendra Usar Dhan 3, NDRK 5062 x IR 28, Sarjoo 52 x Narendra Usar Dhan 3, Narendra 2064 x Narendra Usar Dhan 3, NDRK 5062 x CSR 23, NDRK 5004 x Narendra Usar Dhan 3, NDRK 5037 x CSR 23, NDRK 5040 x Narendra Usar Dhan 3, NDRK 5093 x Narendra Usar Dhan 3 and Narendra 2064 x CSR 23 in F₁s while in F₂s are NDRK 5004 x Narendra Usar Dhan 3, Sarjoo 52 x Narendra Usar Dhan 3, NDRK 5040 x Narendra Usar Dhan 3, NDRK 5062 x IR 28, CSR 10 x Narendra Usar Dhan 3, NDRK 5062 x CSR 23, Narendra 2064 x Narendra Usar Dhan 3, NDRK 5039 x Narendra Usar Dhan 3, NDRK 5059 x IR 28 and DRK 5037 x Narendra Usar Dhan 3. It indicated that additive and non-additive genetic effects were responsible for increased grain yield in these F₁s over the SV₁ and SV₂.

Such type of hybrid could be meaningful for heterosis breeding and desirable segregants could also be screened out in succeeding generations as a substantial part of variance which was considered as fixable one. Heterosis for grain yield in these crosses could be due to desirable heterotic response for component traits such as days to 50% flowering, chlorophyll content, leaf nitrogen, leaf temperature, flag leaf area, plant height, panicle bearing tillers / plant, panicle length, spikelets / panicle, grains per panicle, spikelet fertility, biological yield / plant, harvest index, L/B ratio, 1000- grain weight, amylose content, protein content, indicating genetic association of these character with grain yield. The hybrid combinations showing non additive gene action, may be exploited through the use of CMS system, since the stable CMS with perfect restoration in rice are available.

Therefore, the yielding ability in the present set of material might be enhanced due to higher level of manifestation from yield, physiological and few quality contributing traits. Further, rice workers have also observed that grain yield might be due to



Where, P-Parents, F1S-F1S crosses, F2s-F2s segregants, TS-Transgressive segregants, HID- Highly inbreeding depressed

Figure 1: Protein profiling of selected rice genotypes of different breeding populations by SDS-PAGE



heterotic response through all other yield contributing traits. Similar findings have also been reported by Janardanam *et al.*, 2001; Punitha *et al.*, 2004; Verma and Srivastava, 2005; Singh *et al.*, 2007; Roy *et al.*, 2009 Chougule *et al.*, 2012 and Sudeepthi *et al.*, 2018.

Seed protein profiling is the most promising tool in determining the molecular polymorphism and genetic homology. Seed storage proteins help in cultivar identification by avoiding the external environmental influences. Electrophoretically detectable proteins in rice grains possess the potential of characterizing the germplasm by their taxonomic and evolutionary aspects. This study was aimed at exploiting the genetic variations among 48 (parents+F₁s+ F₂s+transgressive segregants+checks) elite rice genotypes through electrophoretical separation of grain proteins by sodium dodecyl sulphate polyacryamide gel electrophoresis (SDS PAGE) at 12% (**Figure 1**).

At 35 kDa the medium to dark bands were present in parents, F₁s, F₂s, transgressive segregants and

checks while in highly inbreeding depressed cross combinations, variable range of the bands were observed *viz.*, absence of band, light, medium and dark bands.

Majority of these populations have two distinct protein bands in parents; three in F₁s and F₂s; and only one in transgressivesegregants and highly inbreeding depressed cross combination, which indicated that these proteins are developed in the salt tolerant breeding populations and play an important role in salt tolerance. The SDS-PAGE in combination with 2-D electrophoresis is further suggested for documenting contrasting variations of isoforms of protein peptides.

The RM values and banding pattern of different breeding populations viz., parents, F_1s , F_2s , transgressive segregants including checks have been depicted in **Tables 4**, **5** and **6**. Seed protein showed variability in banding pattern of polypeptide at 12% acrylamide gel during SDS-PAGE.

Table 4: Relative mobility	at 12%	SDS-PAGE	of selected	parents in rice

kDa	Length	R.M.								Pare	ents									
value	of gel (cm)	Value	Check 1	Check 2	P16	P15	P14	P13	P12	P11	P10	P9	P8	P 7	P6	P5	P4	Р3	P2	P1
	0																			
100	1	0.10	+++	+++	+++	+	++	+	+	+++	+++	+++	-	++	+++	+	-	++	-	-
75	2	0.13	+++	+++	+++	++	+++	+	++	+++	+++	+++	-	++	++	+++	-	+++	-	-
71	3	0.15	+	++	+	-	++	+	+	++	++	++	-	++	+++	+	-	++	-	-
63	4	0.20	+	+	+	-	+	-	+	+	+	+	-	+	++	+	-	+	-	-
55	5	0.24	++	++	++	+	++	+	++	++	++	++	-	++	++	++	-	++	-	+
35	6	0.40	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	++	+++	+++	+++	++	+++	++	+++
25	7	0.45	+	+	+	+	+	+	+	+	++	++	-	+	++	+	-	+	+	++
20	8	0.60	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	++	+++	+++	+++	++	+++	+	+++
17	9	0.70	-	-	+	+	-	-	-	-	+	-	++	-	+	+	+	-	++	++
11	10	0.80	+	+	+	+	+	-	-	+	+	+	+	+	+++	-	+	-	+	+

⁺ Low intensity of the band, ++Medium intensity of the band, +++ High intensity of the band, -No band was found.

Where, P= Parents

P1-NDRK 5037, P2-NDRK 5062, P3-Sarjoo 52, P4-Narendra 2064, P5-NDRK 5004, P6-NDRK 5040, P7-NDRK 5093, P8-CSR 10, P9-NDRK 5039, P10-NDRK 5059, P11-NDRK 5047, P12-Sundri, P13-NDRK 5025, P14-Narendra UsarDhan 3, P15-CSR 23, P16- IR 28, Check1- Jaya, Check2- CSR 43

Result revealed that majority the parents including checks visualized high to medium intensity of bands on 0.40 and 0.60 RM values (**Table 5**). Similarly, elite hybrids and their segregants showed high to medium intensity of band on 0.50 and 0.75 RM value (**Table 6**). Further, elite

transgressive segregants showed high to medium intensity of band on 0.11, 0.22, 0.40 and 0.65 RM value. However, highly inbreeding depressed crosses showed high to medium intensity of band only on 0.65 RM value.



Table 5: Relative mobility at 12% SDS-PAGE of promising F₁s and F₂s in rice

kDa	Length of	R.M.										Cro	sses									
value	gel (cm)	Value	F2S1	F2S2	F2S3	F2S4	F2S5	F2S6	F2S7	F2S8	F2S9	F2S10	F1S1	F1S2	F1S3	F3S4	F1S5	F1S6	F1S7	F1S8	F1S9	F1S10
	0																					
135	1	0.14	-	+	-	++	++	+	-	+	+	+++	+++	+++	+	+	+	+	+	+	+++	+
100	2	0.17	+	++	-	++	++	+	+	+	+	+++	+++	+++	+	++	+	++	++	++	+++	+
75	3	0.20	+	++	+	+++	+++	++	++	++	++	+++	+++	+++	++	+++	+++	+++	+++	+++	+++	+++
63	4	0.28	++	++	+	+	++	+	+	+	+	++	+++	+++	+	++	++	++	++	++	++	++
48	5	0.34	+++	+++	+	+	++	+	+	+	+	++	+++	+++	+	++	++	++	++	++	+++	+
35	6	0.50	+++	+++	++	++	+++	+++	++	++	++	+++	+++	+++	++	+++	+++	+++	+++	+++	+++	+++
20	7	0.64	++	++	+	+	++	+	+	++	+	++	++	++	-	+	+	+	+	++	++	+
17	8	0.75	+++	+++	++	++	+++	++	++	++	++	+++	+++	+++	++	++	++	++	++	++	++	++
11	9	0.80	+	+	-	-	+	-	-	-	-	+	+	+	-	-	-	-	-	+	+	-
9	10	0.89	+++	+++	-	-	++	-	-	-	-	++	++	++	-	++	+	+	+	++	++	+

⁺ Low intensity of the band, ++Medium intensity of the band

Where,

F1S= F₁S crosses, F2s= F₂s segregants

F1S1- NDRK 5037 x NarendraUsarDhan 3, F1S2- NDRK 5062 x IR 28, F1S3- Sarjoo 52 x NarendraUsarDhan 3, F1S4- Narendra 2064 x NarendraUsarDhan 3, F1S5- NDRK 5062 x CSR 23, F1S6- NDRK 5004 x NarendraUsarDhan 3, F1S7- NDRK 5037 x CSR 23, F1S8- NDRK 5040 x NarendraUsarDhan 3, F1S9- NDRK 5093 x NarendraUsarDhan 3, F1S10-Narendra 2064 x CSR 23, F2S1- NDRK 5004 x NarendraUsarDhan 3, F2S2- Sarjoo 52 x NarendraUsarDhan 3, F2S3- NDRK 5040 x NarendraUsarDhan 3, F2S4- NDRK 5062 x IR 28, F2S5- CSR 10 x NarendraUsarDhan 3, F2S6- NDRK 5062 x CSR 23, F2S7-Narendra 2064 xNarendraUsarDhan 3, F2S8- NDRK 5039 x NarendraUsarDhan 3, F2S9- NDRK 5059 x IR 28, F2S10- NDRK 5037 x NarendraUsarDhan 3

Table 6: Relative mobility at 12% SDS-PAGE of highest depressed crosses and top transgressive segregants in rice

kDa	Length of	R.M.					Cross	es				
value	gel (cm)	Value	HID5	HID4	HID3	HID2	HID1	TS5	TS4	TS3	TS2	TS1
	0											
135	1	0.08	++	+	-	++	+	+	++	++	+	++
100	2	0.09	+++	++	-	+++	+	++	++	++	++	++
75	3	0.11	+++	+++	-	+++	++	+++	+++	+++	+++	+++
74	4	0.12	++	++	-	++	+	++	++	++	+	++
63	5	0.19	++	++	-	++	++	++	++	++	++	++
57	6	0.22	++	++	-	++	++	++	++	++	++	++
35	7	0.40	+++	++	-	++	+	+++	+++	+++	++	+++
25	8	0.55	+	+	++	++	+	++	++	++	+	++
20	9	0.65	+++	+++	+	+++	++	+++	+++	+++	+++	+++
11	10	0.85	+++	+	+	+	+	+	+	++	+	+++

⁺ Low intensity of the band, ++Medium intensity of the band; +++ High intensity of the band, -No band was found.

Where,

TS= Transgressive segregants; HID= Highly inbreeding depressed

TS1- NDRK 5004 x NarendraUsarDhan 3, TS2- Sarjoo 52 x NarendraUsarDhan, TS3- NDRK 5040 x NarendraUsarDhan 3, TS4-), NDRK 5062 x IR 28, TS5- CSR 10 x NarendraUsarDhan 3, HID1-NDRK 5037 x CSR 23, HID2- NDRK 5047 x NarendraUsarDhan 3, HID3- Sundri x IR 28, HID4- NDRK 5044 x IR 28, HID5- NDRK 5025 x CSR 23

Tolerant and susceptible breeding populations showed similar banding pattern but possesses wide variations between tolerant and susceptible. At 35 kDa the medium to dark bands were present in parents, F_1 s, F_2 s, transgressive segregants and checks while in highly inbreeding depressed cross combinations, variable range of the bands were observed *viz.*,

absence of bands, light, medium and dark bands. Such a banding pattern reflects that similar banding pattern on 35 kDa may certainly have salt tolerant QTLs in elite parents, which have inherited successfully in F_1 s and transgressive segregants. Hence, emphasis should be given to elute these desired QTLs inorder to incorporate these salt tolerant traits of interest in

⁺⁺⁺ High intensity of the band, -No band was found.



local cultivar and / or widely adopted genotype for sustainability under salt affected soils. The similar reports have been reported by Tripathy *et al.*, (2015).

Authors' Contributions

The idea of this study was developed by Shiv Prakash Shrivastav and O.P. Verma who also assisted in its design and interpretation of the data. Dan Singh Jakhar helped to draft the manuscript. All authors read and approved the final manuscript.

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Availability of data and material

The experimental data used for analysis and further writing of this article are available with the corresponding author on reasonable request.

Declaration

Conflict of interest

We declare that there is no conflict of interest in connection with the work submitted by us.

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RESEARCH ARTICLE

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Starch Kinetics, Cooking Quality and Phytochemical Composition of Geographical Indication (GI) Tagged Rice of Kerala

Chitra Pillai¹, Faseela KV² and Harikumaran Thampi^{1*}

¹Department of Life Sciences, University of Calicut-673635, Kerala, India.

²Department of Plant Breeding and Genetics, Regional Agricultural Research Station (Central zone), Kerala Agricultural University, Mele Pattambi-679306, Kerala

*Corresponding author Email: drhari@uoc.ac.in

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Abstract

Geographical Indication (GI) tagged traditional rice of Kerala namely the red pigmented Palakkadan Matta and non-pigmented aromatic rice - Wayanadan Gandhakasala and Wayanadan Jeerakasala are well known for their cooking and eating quality, but are underutilized for their nutritional significance. The objective of this study was to classify these varieties based on their starch digestibility, biochemical composition and cooking characteristics. Results indicated significant (p <0.05) variation among rice varieties for all the parameters evaluated. *In vitro* starch digestibility studies found significantly low glycemic index (< 68) but higher RS content for Wayanadan Gandhakasala and Chettadi. Aromatic varieties had lower cooking time (< 25 min) than Palakkadan Matta varieties. High water uptake ratio and elongation ratio was also noted for Wayanadan Gandhakasala. Total phenolic content correlated positively to total flavonoid content. Over all, Palakkadan Matta varieties had significantly higher free phenolic, free flavonoid and total flavonoid content than non-pigmented varieties. The study indicates that these GI tagged traditional rice varieties can thus be utilised in the functional food industry based on their intermediate glycemic index, desirable cooking qualities and the presence of beneficial bioactive components.

Key words: Geographical indication, Palakkadan Matta, Aromatic varieties, Glycemic Index, Cooking Properties, Total phenolic content

Introduction

Rice (*Oryza sativa* L.) is one of the most studied cereal crops of the world and forms the central part of the Indian diet. Rice gained popularity from the fact that it can be grown under diverse climatic conditions and can be cooked in many different ways. Therefore, quality of rice is a topic for which there can be different answers. Different countries or regions within a country prefer eating rice in different ways and therefore the definition of rice quality differs accordingly (Bhattacharya, 2009). The most commonly used quality traits include

physical appearance, nutrition, milling and cooking characteristics. Numerous scientific studies suggest that both genetic and environmental factors can affect quality traits in rice grain (Chen et al., 2012; Custodio et al., 2019; Krishnamrutha et al., 2023). Some rice varieties are well known for their unique qualities because of the particular region in which they are grown. These special varieties found reference in historical scripts and were assigned with a unique status or tag known as geographical indication (GI) under the Geographical Indications of Goods



(Registration and Protection) Act, 1999 (Blakeney et al., 2020). GI status gives them a unique status among other varieties and provides more market and export value.

Among the agriculture commodities, 12 GI tags are recorded for rice in India, six of which are cultivated in the state of Kerala. These include Palakkadan Matta, Kaipad rice, Navara rice, Pokkali rice, Wayanadan Gandhakasala and Wayanadan Jeerakasala. Among these varieties, Navara is the most popular and has the highest market income, which could be due to its medicinal properties (Radhika *et al.*, 2018). Majority of scientific literature available on traditional rice in Kerala has focused on Navara. Palakkadan Matta find reference in Tamil classic 'Thirukkural' and was consumed by the royal families of Chola and Chera dynasty. Under the GI registry, Palakkadan Matta include ten different varieties. However, only four

varieties are most commonly cultivated in Palakkad, which include Thavalakannan, Chenkayama, Chettadi and Chitteni. They have bold grain with red pericarp and is known for their unique earthy flavour which is supposed to be due to the heavy soil they are grown in, which is rich in clay and silt. These varieties are mostly consumed in parboiled form and used in the preparation of a wide range of rice based dishes and snacks.

Wayanadan Gandhakasala and Wayanadan Jeerakasala are non-pigmented fine aromatic rice cultivated organically in high altitudes of Wayanad by the Kurichiya tribe, an agricultural based community in Kerala. Owing to their distinct aroma when cooked, they are known as 'Kerala Basmathi' and fetch higher price in the market than other varieties (Krishnankutty *et al.*, 2021; Soam, 2005). Agronomical features of the varieties taken for this study are listed in **Table 1.**

Table 1: Agronomical features of rice varieties

Sl. No.	Popular name	Crop maturation (days)	Agronomical features
Pala	kkadan Matta V	arieties	
1	Chenkazhama	110- 120	Tall indica variety.
2	Chettadi	130 -160	Photosensitive and drought resistant.
3	Chitteni	130	Highly tolerant to Bacterial leaf blight and pests like Brown plant
			hopper, Gall fly and green leaf hopper.
4	Thavalakannan	130	Less prone to lodging, good straw yield and tolerant to Gall fly
			and Foot rot. Highly adaptive to adverse soil conditions and
			shows strong resistance to green leaf hopper.
Aror	natic varieties		
5	Wayanadan	160 - 180	Aromatic, highly tolerant to pest and disease, High priced, suitable
	Gandhakasala		as fodder (Blakeney 2020)
6	Wayanadan	160 - 180	Aromatic, high priced
	Jeerakasala		

Even though these varieties are known for their unique properties, very few scientific studies are available on the nutritional and cooking properties of these GI rice varieties. The selected varieties are not explored for their phytochemical composition

as most studies have focused on their agro morphological characteristics. The objective of this study is to evaluate and classify selected GI tagged rice varieties for their nutritional, phytochemical and cooking characteristics.



Materials and Methods

Rice samples

Six rice samples including four most commonly cultivated Palakkadan Matta rice varieties namely Chenkazhama, Chettadi, Chitteni and Thavalakannan and two aromatic varieties namely Wayanadan Gandhakasala and Wayanadan Jeerakasala were collected from Regional Agricultural Research Stations (Mele Pattambi and Ambalavayal), Kerala and Abhayam, Pattambi, Kerala. Varieties Wayanadan Jeerakasala and Wayanadan Gandhakasala were non-pigmented aromatic rice and all others were red pigmented varieties.

The dried paddy samples were kept at 4 °C in airtight plastic containers until used. The samples were dehulled manually and homogenised into fine rice flour (60 mesh sieve) freshly before analysis. For the determination of glycemic index and starch fractions, 50 mg rice flour was homogenised in 5 ml distilled water and cooked for 30 minutes.

Chemicals and Reagents

Glucose oxidase/peroxidase reagent, amyloglucosidase solution (from *Aspergillus niger*), heat stable α-amylase (from *Bacillus licheniformis*), standards of gallic acid and (+) - catechin were purchased from Sigma-Aldrich, USA. HPLC grade chemicals namely methanol, acetonitrile and pepsin from porcine stomach mucosa were purchased from HiMedia. All other chemicals used were of analytical grade.

Determination of Total Starch (TS) and its fractions

The TS content was determined following the method reported by Goñi *et al.*, (1997)to calculate the glycemic index (GI). Briefly, 50 mg brown rice flour was added to 2M KOH (6 ml) and energetically shaken for 30 minutes at room temperature. To the mixture, added 0.4 M Sodium acetate buffer, pH-4.75 (3 ml) and amyloglucosidase (60 µl) and incubated for 45 minutes in a shaking water bath at 60 °C.

Starch content was measured as glucose using glucose oxidase-peroxidase reagent and a factor of 0.9 was used for the conversion of glucose to starch.

Resistant starch (RS) content was measured following the enzymatic method described by Goñi et al., (1997)to calculate the glycemic index (GI. Briefly, to the brown rice flour (100 mg), added KCl-HCl buffer (10 ml), pH 1.5 and homogenized in 50 ml centrifuge tube. For the removal of proteins, the mixture was treated with freshly prepared pepsin solution containing 0.02 g pepsin in 0.2 ml KCl-HCl buffer at 40 °C for 60 minutes. Then added 9 ml Tris-Maleate buffer, pH 6.9 and the suspension was hydrolysed with solution containing α-amylase (40 mg) at 37 °C for 16 hours. The suspension was centrifuged and the residue was then incubated with 80 µl amyloglucosidase solution for 45 minutes at 60 °C with constant shaking. The hydrolysate was centrifuged and the supernatant was collected for the estimation of RS content, as described earlier. For the estimation of digestible starch (DS), the difference between TS and RS was calculated.

In vitro starch hydrolysis and determination of glycemic index

The rate of rice starch hydrolysis was studied following the *in vitro* method suggested by Goñi *et al.*, (1997) to calculate the glycemic index (GI. Briefly, 50 mg brown rice flour was added to 10 ml of KCl- HCl buffer and pH adjusted to 1.5. To this suspension, 0.2 ml of freshly prepared Pepsin solution was added and kept for 1 hour at 40 °C in a shaking water bath. The volume was completed to 25 ml with Tris maleate buffer. The solution was then hydrolysed with α - amylase solution (2.6 units in Tris-Maleate buffer) and kept in a shaking water bath at 37 °C. One ml hydrolysate solution was drawn every 30 minutes for a total of 180 minutes.

The aliquots were then heated in boiling water bath followed by rigorous shaking in a vortex mixer to inactivate their enzyme activity. To these aliquots,



added 0.4 M sodium acetate buffer, pH 4.75 (3 ml) and amyloglucosidase (60 μ l), mixed and incubated at 60 °C for 45 minutes in shaking water bath. The glucose content was determined by glucose oxidase/peroxidase reagent and the amount of starch was calculated. The results were then expressed as the per centage of TS hydrolysed at a given time interval.

The data obtained was used for plotting starch hydrolysis curve and the area under curve (AUC) was calculated. A first order non-linear reaction equation followed for the starch hydrolysis as provide by Goñi *et al.*, (1997)to calculate the glycemic index (GIto calculate the glycemic index (GI: C=C_x(1-e^{-kt}),

where C - Concentration at time t, $C\infty$ - Equilibrium concentration, k - kinetic constant and t - chosen time.

Parameters $C\infty$, k and AUC were determined from the experimental data using software SYSTAT (Sigma plot 14), MS office version. The hydrolysis index (HI) was expressed as per centage and calculated using the formula:

Glycemic Index was determined by the following equation:

Glycemic Index = 39.71 + 0.549 HI

Determination of cooking characteristics

Cooking characteristics of rice varieties were determined following the method suggested by Singh *et al.*, (2005).

Minimum cooking time

Brown rice kernels (1g) were added to 10 ml distilled water and heated in a boiling water bath. The rice grains were removed every 2 minutes during cooking and pressed between two glass slides. The time (min) at which no white residue was left in the glass slides was taken as the minimum cooking time.

Water uptake ratio

After cooking rice kernels for their minimum cooking time as described above, the water was drained and the cooked samples were weighed after pressing in filter paper. The difference in weight of cooked sample was calculated as the water uptake ratio.

Elongation ratio

Elongation ratio was obtained by dividing the average length of cooked rice kernels by the average length of uncooked rice kernels (n=10).

Cooked length-breadth ratio

The average length of cooked rice kernels was divided by the average breadth of cooked kernels and termed as 1/b ratio (n=10).

Extraction of free form phenolic compounds

Free form phenolic compounds were extracted from brown rice flour using the method of Gong *et al.*, (2017). Briefly, 500 mg of whole grain flour was blended with 80% chilled Ethanol (5 ml) for 15 minutes. After centrifuging the mixture (5000 rpm) for 10 minutes, the supernatant was pooled and the residue was extracted twice. The pooled extract was then subjected to evaporation at 45 °C until the extract was reduced to 3 ml. Reconstituted the extract with distilled water to 6 ml and kept at -40 °C until use.

Extraction of bound form phenolic compounds

The residue obtained after extracting free form phenolics, was treated with 2M NaOH solution (10 ml) at room temperature for 1 hour under nitrogen gas. The mixture was then acidified with 2 M HCl solution (10 ml) until pH 2 was obtained. The acidic solution was then extracted with hexane (10 ml). The final solution was then extracted with Ethyl acetate (10 ml) thrice. The extracts were pooled, dried and reconstituted in 5 ml of distilled water. The extract was then stored at -40 °C until use (Gong *et al.*, 2017).

Determination of phenolic content

The total phenolics content was estimated by the colorimetric method reported by Sompong et al., (2011). Briefly, extract solution (120 µl) was treated



with freshly prepared Folin-Ciocalteu reagent diluted 10-folds (600 μ l) and incubated for 2 minutes. To the mixture, added 960 μ l NA₂CO₃ solution (75 g/l) and kept at 50°C for another 2 minutes. The blue colour developed was then read at 760 nm. Gallic acid was taken as the standard and the results were expressed as mg gallic acid equivalent (GAE) per 100 g brown rice.

Determination of flavonoid content

The total flavonoid content was determined following the modified colorimetric method of Dewanto *et al.*, (2002). The extract solution (300 µl) was initially diluted with 1.5 ml of distilled water in a test tube. Then added 5% NaNO₂ solution (90 µl) and kept at room temperature for 6 minutes followed by the addition of 10% AlCl₃.6H₂O solution (180 µl) and incubated for 5 minutes. To the mixture, finally added 1 M NaOH (600 µl) and the volume was made up to 3 ml with distilled water. The colour developed was measured at a wavelength of 510 nm and compared with the standard + (-) Catechin solution. Total flavonoid contents were expressed in terms of mg + (-) catechin equivalents (CE) per 100 g of brown rice.

All the analytical assays were performed thrice (n-3) and reported as mean \pm standard deviation on fresh weight basis. The results were analysed by Analysis of Variance (ANOVA) SPSS version 20 using Duncan's Multiple Range Test (DMRT) to compare means at p < 0.05 significance level. Pearson's correlation analysis was performed for calculating the relationship between different variables.

Results and Discussions

TS and its fractions

TS, RS and DS contents of rice samples are shown in **Table 2.** The amount of TS in rice samples ranged from 72.99% in Wayanadan Jeerakasala to 83.68% in Chenkazhama. All the varieties were subjected to same cooking method for TS determination. Higher TS content in rice could result from leaching of starchy fragments during cooking because of different degree of damage to the grain structure (Ahmed and Urooj, 2003). RS and DS content were in the range of 0.56 - 0.69% and 72.35- 76.01% respectively. Wayanadan Jeerakasala had significantly lower TS and DS content among all varieties, which is a desired parameter for managing blood glucose response. The values obtained were comparable to other studies by Deepa *et al.*, (2010) and Hu *et al.*, (2004).

Statistical analysis

Table 2: Total starch, resistant starch and digestible starch content of rice varieties (%)

Sample	Total Starch	Resistant Starch	Digestible Starch
Chenkazhama	83.68 ±2.06 ^a	0.64 ± 0.01^{b}	83.03 ±2.07 ^a
Chettadi	77.75 ±2.33 ^b	0.69 ± 0.02^{a}	77.10 ±2.33 ^b
Chitteni	76.58 ± 1.83^{bc}	0.56 ± 0.03^{d}	76.01 ±1.82°
Thavalakannan	73.86 ±1.96°	$0.63 \pm 0.02^{\circ}$	73.22 ± 1.96^{d}
Wayanadan Gandhakasala	74.60 ± 0.36^{bc}	0.64 ± 0.0^{bc}	73.96 ± 0.36^{d}
Wayanadan Jeerakasala	72.99 ±2.5°	0.63 ±0.03°	72.35 ±2.53°
Mean	76.57	0.63	75.94
CV	2.76	0.38	2.77
CD (0.05)	3.85	0.00	3.08

Values with the same letters in a column are not significantly different (p < 0.05). TS, total starch; RS, resistant starch; DS, digestible starch

Rice mainly comprises Type 1 RS, which is the physically inaccessible starch normally found in whole grains or Type 5, which is formed by amylose-lipid

complexes (Sajilata *et al.*, 2006). Amylose content is predicted as a major factor affecting the RS content. Retrogradation of amylose was found to be the



primary mechanism for the formation of RS (Deepa et al., 2010; Berry, 1986). Lehmann and Robin, (2007) and Sajilata et al., (2006) reported a positive correlation between RS and amylose content in rice. In contrast, some of the intermediate amylose varieties found in the study had higher RS than high amylose varieties (reported elsewhere) (Pillai et al., 2020). In a similar study by Hu et al., (2004) rice cultivars with similar amylose content had different RS content due to difference in planting seasons and varietal factors. His study also characterized highamylose varieties with high RS content to be associated with low RVA (Rapid visco analyser) parameters like peak viscosity, hot plate viscosity and cool paste viscosity.

Cooking characteristics

Significant differences were observed for cooking characteristics of rice varieties as shown in **Table**3. Minimum cooking time ranged between 23.83

to 35.50 min. Wayanadan Gandhakasala and Wayanadan Jeerakasala had significantly lower cooking time than Matta varieties. Highest cooking time was observed in Thavalakannan, which also had significantly higher protein content (11.4 g/100 g) among the varieties analysed (Pillai et al., 2020). This was similar to the observation made by Juliano et al., (1965) that high protein rice varieties require longer cooking time. However, no positive correlation was found between protein content and cooking time of the rice varieties. Protein content of Chenkazhama, Chettadi, Chitteni, Gandhakasala and Jeerakasala were 6.75, 8.01, 7.77, 10.67 and 8.16 g/100g respectively (reported elsewhere) (Pillai et al., 2020). Longer cooking times can lead to low acceptability of brown rice among consumers (Adebamowo et al., 2017).

Table 3: Cooking properties of rice varieties

Rice varieties	Minimum cooking time (min)	Water uptake ratio	Elongation ratio	Cooked l/b ratio
Chenkazhama	26.46±0.41 ^d	1.76±0.01 ^d	1.15±0.00°	1.82±0.02 ^f
Chettadi	33.5±0.50 ^b	2.03±0.00 ^b	1.10±0.01 ^d	2.44±0.04e
Chitteni	32.4±0.36°	2.01±0.02°	1.18±0.02 ^b	3.04±0.03°
Thavalakannan	35.5±0.50°	2.15±0.01ª	1.16±0.03°	3.35±0.04 ^a
Wayanadan Gandhakasala	24.4±0.36°	2.16±0.01a	1.29 ±0.01ª	2.84±0.04 ^d
Wayanadan Jeerakasala	23.83±0.28 ^e	2.01±0.01°	1.11±0.02 ^d	3.18±0.02 ^b
Mean	29.35	2.02	1.17	2.78
CV	1.21	0.51	1.07	1.10
CD (0.05)	0.65	0.02	0.02	0.05

Values with the same letters in a column are not significantly different (p < 0.05)

Water uptake ratio is a measure of volume expansion of rice during cooking and was significantly higher in Gandhakasala (2.16) and Thavalakannan (2.15). ER refers to the length wise elongation of rice after cooking and is the most desirable cooking traits especially in varieties like Basmathi. Significantly

higher ER was observed in Wayanadan Gandhakasala (1.29) followed by Chitteni and Thavalakannan whereas Wayanadan Jeerakasala had significantly lower ER value. Wayanadan Gandhakasala had better price and demand than Wayanadan Jeerakasala in the market (Radhika *et al.*, 2018), which could



also be attributed to its better cooking qualities. Results obtained for ER and water uptake ratio were comparable to the observations made by Rajesh *et al.*, (2018) and Nirmala Devi *et al.*, (2015). Cooked 1/b ratio ranged from 1.82 to 3.35 and was higher for Thavalakannan which could be due to its higher water uptake ratio.

In vitro starch digestion

More than 50% of the TS was digested within the first 30 minutes of hydrolysis except for Wayanadan Gandhakasala and Chettadi. When the rate of starch hydrolysis was plotted against time, the curve obtained reached a plateau after 60 minutes of digestion as shown in **Figure 1.** According to Goñi

et al., (1997), rate of starch hydrolysis after 90 minutes (H 90 Experimental) was found to be the best hydrolysis value for determining the in vivo glycemic response. H 90 values were also calculated theoretically (H 90 Theory) as shown in **Table 4**. There was good agreement between H 90 Exp and H 90 Theory values. Rate of starch hydrolysis after 90 min was highest for Wayanadan Jeerakasala (62.56%) followed by Chitteni (59.74%) which suggests that they get digested more rapidly than other varieties and could elicit a high glycemic response. Significantly lower H 90 value was observed in Chettadi (51.41%) and Wayanadan Gandhakasala (52.16%). From 120 to 150 min, rate of starch hydrolysis was slow.

Table 4: Rate of starch hydrolysis (%) at 90 min

Sl. No.	Rice variety	H ₉₀ Exp*	H ₉₀ Theory**
1.	Chenkazhama	56.35±0.23°	56.78 ± 0.13^{d}
2.	Chettadi	51.41 ±0.22 ^d	53.02 ±0.34°
3.	Chitteni	59.74 ±0.71 ^b	60.20 ±0.61 ^b
4.	Thavalakannan	56.46 ±3.56°	59.09 ±1.68°
5.	Gandhakasala	52.16 ±1.07 ^d	53.14 ±0.85°
6.	Jeerakasala	62.56 ±0.43 ^a	64.91 ±0.32 ^a

^{*}based on experimental results; **based on the equation, $C = C\infty$ (1-e-kt)

Glycemic index

There were significant variations in $C \infty$ values of rice varieties which refers to the amount of starch hydrolysed after a prolonged time (180 minutes) as depicted in **Table 5**. $C \infty$ values of all varieties suggests that starch hydrolysis terminated before 180 min and significantly

lower values were observed for Chettadi and Wayanadan Gandhakasala. The k value ranged between 0.05-0.10. It determines the rate of starch digestion and absorption in the body. The C ∞ and k values are good predictors of glycemic index (Edwards *et al.*, 2019).

Table 5: Starch kinetics parameter of rice varieties

Rice variety	k	C _∞	AUC*	HI	GI
Chenkazhama	0.10	56.79±0.13°	9293 ±17°	$55.93 \pm 0.10^{\circ}$	70.41±0.06°
Chettadi	0.06	53.16±0.44 ^d	8505 ±٣٦ ^d	51.19 ±0.21 ^d	67.81±0.12 ^d
Chitteni	0.07	60.32±0.49b	9717±173 ^b	58.48±1.04 ^b	71.81±0.57 ^b
Thavalakannan	0.05	59.80±1.68 ^b	9365±230°	56.36±1.39°	70.65±0.76°
Wayanadan Gandhakasala	0.07	53.30±0.93 ^d	8525±116 ^d	51.30±0.70 ^d	67.87±0.38 ^d
Wayanadan Jeerakasala	0.06	65.22±0.32a	10373±44a	62.43±0.27 ^a	73.98±0.15 ^a

Values with the same letters in a column are not significantly different (p \leq 0.05)

AUC of glucose (reference food) was calculated as 16,616



The HI values ranged between 51.19- 62.43%. HI refers to the proportion of starch that is theoretically digestible and is a predictor of glycemic index. Glycemic Index values ranged from 67.81 to 73.98. Brand-Miller *et al.*, (2009) classified food based on their glycemic index values as low (55 or less), intermediate (56 to 69) or high (70 or more). Accordingly, Wayanadan Jeerakasala, Chitteni, Thavalakannan and Chenkazhama had high glycemic index whereas Chettadi and Wayanadan Gandhakasala were varieties with intermediate glycemic index.

Some of the physico-chemical factors that can affect the starch digestibility in rice include amylose and amylopectin ratio, size of starch granules, presence of fiber, natural amylase inhibitors, starch- protein interactions and formation of lipid-amylose complexes (Panlasigui et al., 1991 and Sagum and Arcot, 2000).

Total phenolic content

Table 6 provides the total phenolic content of brown rice flour along with its fractions namely free form and bound form phenolics. The free form phenolic content ranged from 34.45 mg GAE/100 g in Wayanadan Gandhakasala to 132.82 mg GAE/100 g in Chettadi. A range of 49.76 to 103.77 mg GAE/100g was found for bound form phenolics. Palakkadan Matta varieties had significantly higher free phenolic content than non-pigmented varieties. However, the highest amount of bound phenolics was recorded in Wayanadan Jeerakasala followed by Chitteni, Wayanadan Gandhakasala, Chenkazhama, Chettadi and Thavalakannan.

Table 6: Phenolics and flavonoid content of brown rice flour

Vowiety	Phenolics	content (mg G	AE / 100 g)	Flavonoid	content (mg	CE/ 100 g)
Variety	Free form	Bound form	Total	Free form	Bound form	Total
Chenkazhama	109.98±0.71°	74.30±0.87°	184.29±0.66°	272.50±4.44b	9.74±0.64 ^d	282.24±4.90b
Chettadi	132.82±4.07ª	66.36±2.47 ^d	199.19±5.38 ^b	388.50±9.5ª	3.37±0.21 ^f	391.87±9.73ª
Chitteni	116.77±4.30 ^b	94.43±0.58b	211.20±4.88a	264.66±8.5b	14.37±0.37 ^a	279.04±8.81 ^b
Thavalakannan	57.14±2.64 ^d	49.76±1.26e	106.90±2.95°	101.00±1.5°	6.49±0.12e	107.49±1.43°
Wayanadan	34.45±1.28e	72.89±0.47°	107.34±1.23°	81.33±3.01 ^d	11.49±0.12°	92.83±2.94d
Gandhakasala						
Wayanadan	36.94±1.03°	103.77±0.95a	140.71±0.30 ^d	85.66±2.51 ^d	13.33±0.06 ^b	98.99±2.58 ^{cd}
Jeerakasala						
Mean	81.35	76.91	158.27	198.94	9.80	208.74
CD (0.05)	5.41	2.18	6.31	11.20	0.61	11.68
CV	3.66	1.56	2.19	3.09	3.45	3.07

Values with the same letters in a column are not significantly different (p < 0.05).

The free phenolic content of the Palakkadan Matta varieties was greater than their respective bound form phenolics. This was consistent with the observation made by Sumczynski *et al.*, (2016) for red and black rice varieties. Non-pigmented rice varieties on the other hand, had higher content of bound form phenolics

than their free form phenolic content, as also observed by Goufo and Trindade (2014). Therefore, pigmented and non-pigmented rice varieties can be good sources of free and bound form phenolics respectively and their distribution in rice kernel might be related to their bran colour.



Free and bound form phenolics perform different physiological functions in the body. Free form phenolics are readily absorbed in the small intestine and exhibit inhibitory action against LDL cholesterol oxidation whereas bound form phenolics are released by enzymatic or microbial fermentation in the colon, have anti-inflammatory properties and provide protection against colon cancer (Chandrasekara and Shahidi, 2011; Shao and Bao, 2015).

Total phenolic content ranged between 106.90 mg GAE/100g in Thavalakannan to 211.20 mg GAE/100g in Chitteni. The values obtained falls within the range of 79.18 to 691.37 mg GAE/100g reported for red and black rice varieties by Sompong et al., (2011). Three out of four Palakkadan Matta varieties namely Chettadi, Chitteni and Chenkazhzma had higher total phenolic content than non-pigmented varieties. However, another red rice variety Thavalakannan had significantly less total phenolic content than nonpigmented variety Wayanadan Jeerakasala. This could be explained by the observation made by Sumczynski et al., (2016) that total phenol content is more of a cultivar specific property rather than a colour dependent trait. Their study further suggested that phenolic compounds are secondary metabolites, significantly affected by stress conditions like wounding, extreme temperatures as well as environmental factors like cultivation techniques, altitude and use of fertilizers.

Total flavonoid content

Flavanoids are phenolic compounds with wide range of biological activities. They are potent antioxidants, antimicrobial and anti-inflammatory compounds. They are primarily known for their antioxidant or radical scavenging activities (Pietta, 2000). Free form flavonoid content of rice varieties exhibited significant variations with high CD value of 11.20 (p <0.05) as shown in **Table 6**. The range obtained was 81.33 in Wayanadan Gandhakasala to 388.50 mg CE/100g in

Chettadi with a mean value of 198.94 mg CE/100g. Free form flavonoids content was significantly higher in Palakkadan Matta varieties than non-pigmented aromatic varieties, as also observed for free form phenolic content.

Bound form flavonoids ranged from 3.37mg CE/100g (Chettadi) to 14.37 mg CE/100g (Chitteni) and constituted a maximum of 5.15% towards total flavonoid content. This suggests that the flavonoids found in free form are the major contributor towards total flavonoid content (> 95%). This was in line with the observation made by Sumczynski *et al.*, (2016) for red and black rice varieties.

Total flavonoid content was significantly higher Palakkadan Matta varieties (Chettadi> Chenkazhama > Chitteni > Thavalakannan) than Wayanadan Jeerakasala and Wayanadan Gandhakasala. The width of variation between the lowest (92.83) and highest value (391.87) of total flavonoid content was high (299.04 mg CE/100g). High CD value was observed for total flavonoid content, suggesting high varietal variation among rice varieties. The results obtained for total flavonoid content and total phenolic content in the present study was comparable to the data reported by Goufo and Trindade (2014). Total flavonoid content and total phenolic content of rice (r- 0.76, p-0.001) were found to be positively correlated, as also reported in a study done by Zhang et al., (2010) (Table 7). Their study also found significant correlation between total phenolic content and total flavonoid content of black rice bran with its total antioxidant activity. Therefore, rice varieties with elevated levels of total phenolic content and total flavonoid content can be used for their antioxidant properties in developing rice based functional foods.



Table 7: Correlation among different characteristics studied in rice varieties

	Total phenolic content	Total flavonoid content	Total starch	Resistant starch	Glycemic index
Total phenolic content	1				
Total flavonoid content	0.760**	1			
Total starch	0.301	0.484	1		
Resistant starch	0.126	0.348	-0.042	1	
Glycemic index	0.210	-0.363	-0.311	-0.329	1

^{**} Correlation is significant at 0.01 probability level

Conclusion

GI tagged rice varieties of Kerala were evaluated for various grain quality parameters and significant differences (p & lt; 0.05) were observed for every parameter studied. All varieties had high TS content (& gt; 70%) however, their rate of starch digestibility was different. For all varieties except Wayanadan Gandhakasala and Chetttadi, more than 50% of the TS was digested within 30 min and the hydrolysis reaction terminated before 180 min. Wayanadan Gandhakasala was also noted for significantly lower cooking time but higher water uptake and elongation ratio. Palakkadan Matta varieties had comparatively higher free phenolic, free flavonoid and total flavonoid content than the nonpigmented varieties. Pearson's correlation analysis revealed significant positive correlation between total phenolic and total flavonoid content. The width of variation for lowest and highest flavonoid content was high amongst varieties suggesting the necessity of screening rice varieties for their phytochemical composition. The study suggests that GI tagged varieties, which are known for their eating and cooking qualities, can also prove to be a potent ingredient for the development of functional food.

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RESEARCH ARTICLE

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Impact of High Temperature Stress on Morpho-Physiological Components and Yield Traits in Rice

Veronica N*, Venkata Ramana Rao P, Suneetha Y and Vasantha Ch

Regional Agricultural Research Station, Maruteru, ANGRAU - 534 122, India

*Corresponding author Email: n.veronica@angrau.ac.in

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Abstract

Rice production is being threatened by increase in temperature around the globe. This is affecting the production and productivity of the crop. Changing climate scenario demands the need to identify thermotolerant genotypes to be used as donor in future breeding programmes. The effect of high temperature from panicle initiation to maturity was imposed in twenty-six rice genotypes. The difference in mean monthly maximum and minimum temperature was 3.5 °C and 1.4 °C, respectively between the polyhouse and ambient control. Results revealed a reduction in important physiological traits *viz.*, membrane thermostability as well as chlorophyll content. Of the genotypes tested, MTU 1290, MTU 1153, MTU 1156, JB 683-1, N22 and IL 19211 maintained a higher stability of membranes and lesser reduction in chlorophyll content when exposed to high temperature stress. High temperature resulted in reduction of grain yield as well as yield attributes in rice genotypes. The above genotypes also had a higher spikelet fertility and lesser yield penalty when exposed to high temperature. The results indicated that these genotypes could be selected as thermotolerant and could serve as potential donors for future breeding programmes.

Key words: Chlorophyll content, Grain yield, Membrane thermostability, Rice, Spikelet fertility

Introduction

Rice production is the backbone of Indian agriculture. Majority of the population depends on this cereal crop to meet either their economic needs or for their caloric intake. Rice production is hindered by abiotic as well as biotic stresses. Keeping the climate change scenario in view, abiotic stresses play a major role in decreasing the crop production and productivity resulting in need of a sustainable crop development and resistance to abiotic stress (Randive *et al.*, 2019). Of this, high temperature is one of the major constraints in rice production.

The fifth assessment report of the Inter-governmental Panel on Climate Change (IPCC, 2014) stated that the increase in global average temperature from 1880 to 2012 was 0.85 °C and by 2100 the projected increase of global mean surface temperature is 3-5 °C. This rise in temperature affects the growth and development resulting in morphological, physiological and biochemical changes in plants (Ashraf and Hafeez, 2004).

High temperature affects rice at every growth stage beginning from the inhibition of germination, seedling stage, tillering, reproductive and also at grain filling stage (Dubey *et al.*, 2018). Of all the stages, booting and flowering are the most sensitive stages to high temperature stress which leads to disruption of cell membranes as well as breaking of cell compartmentalization (Larkindale and Huang,



2004). High temperature at flowering stage results in inhibition of anther dehiscence and loss of stigma receptivity resulting in increase of spikelet sterility and lesser number of grains in the panicle. It also reduces the photosynthetic efficiency and ultimately resulting in yield reduction (Narayanan *et al.*, 2016). High temperature results in damage to the thylakoid membrane as well as inhibition of key enzymes in photosynthetic pathway (Bita and Gerats, 2013). In chlorophyll, the thylakoid granum and membranes are degraded to increase in temperature resulting in reduced photosynthesis (Xu *et al.*, 2021).

Imposition of high temperature at reproductive stage leads to reduction in panicle weight, number of spikelets and decreased grain weight (Wang *et al.*, 2019). Lyman *et al.*, (2013) reported that for every 1°C increase in average temperature during rice growing season the paddy yields reduced by 6.2%. It was also reported that in rice spikelet sterility was increased by 80% when high temperatures coincided the reproductive stage of the crop (Xu *et al.*, 2020).

Rice production plays a prominent role in ensuring food security in the country and at this juncture there is a need to identify and breed for thermo tolerant rice varieties that can withstand the future temperature elevations. With this aim, twenty-six genotypes of rice were screened and impact of high temperature was studied in these genotypes to identify thermo tolerant genotypes.

Materials and Methods

Plant material: Twenty-six rice genotypes were screened and tested for high temperature tolerance by conducting a field experiment in *kharif* 2023 at Regional Agricultural Research Station, Maruteru. The experiment was laid out in split plot design with three replications. A spacing of 20 cm between rows and 15cm between hills was maintained. Recommended dose of Nitrogen (N), Phosphorus (P) and Potassium

(K) was applied. Water management as well as crop protection measures were taken up time to time. All the other package of practices recommended for irrigated transplanted rice were followed.

Imposition of treatments: One set of genotype was grown in ambient conditions and another set was subjected to high temperature stress by enclosing a polythene sheet supported by bamboo poles and iron frame at panicle initiation (PI) stage. Temperature inside the polyhouse was monitored continuously using data logger up to physiological maturity.

Morpho-phenological traits: Observations were recorded for various morpho-phenological from the sampled plants tagged in each treatment and genotype. Plant height on tagged plants was recorded at reproductive stage by measuring the height from base of the plant to the tip of the terminal leaf or panicle on main stem and was expressed in centimetres (cm). The number of days taken for 50 per cent of plants to flower in each genotype and each treatment was noted as days to 50% flowering and was expressed in days. The number of days taken from sowing to physiological maturity was recorded and was expressed as days to maturity.

Membrane thermostability: Membrane thermostability (MTS) was measured following the procedure described by Haque et al., (2009). Mature leaves at reproductive stage were collected and first 2 to 4 cm was clipped off. The next 5 cm of leaf was washed three times with deionised water, cut into small pieces and placed in tubes with 10 ml deionized water. Two sets of each sample were prepared, one set designated as control was maintained at 28 °C while, other set was treated in water bath at 52 °C for one hour. Three replications were maintained for both the sets. After the treatment, control and treated tubes were kept at room temperature for 24 h. The initial conductance was measured using conductivity



meter. Thereafter, all the tubes were autoclaved at 121 °C at 15 lb for 20 mins and the next day final conductance was measured. This ensured complete electrolyte leakage from the plant tissue. The MTS was calculated using the following equation (Blum and Ebercon, 1981).

MTS (%) =
$$(1-(T1/T2)) / (1-(C1/C2) \times 100)$$

Where, C1= initial conductance of control sample; C2=final conductance after autoclaving of control sample; T1= initial conductance of high temperature sample (after water bath treatment); T2= final conductance after autoclaving of high temperature sample.

Chlorophyll content: It was estimated in flag leaf at reproductive stage (1 week after anthesis). For this flag leaf was cut into small pieces and 25 mg of leaf sample was weighed. Chlorophyll was extracted by placing the sample in 80% acetone solution as per the methodology described by Porra *et al.*, (1989). The absorbance was measured using a UV-VIS spectrophotometer. Chlorophyll a and chlorophyll b were measured at 663.2 nm and 646.8 nm, respectively and the chlorophyll content was expressed in mg g⁻¹ fresh weight (mg g⁻¹ FW). Chlorophyll a content, chlorophyll b content and the total chlorophyll content was calculated according to Lichtenthaler and Wellburn, (1983).

Yield and yield attributes: Yield and yield attributes such as grain number per panicle, 1000 grain weight, grain yield and total dry matter were recorded. At physiological maturity, panicles were threshed from a demarcated area of one square meter in all the genotypes under control and stress conditions. The number of panicles in one meter square area were threshed, cleaned and the weight of grains was recorded and expressed as grain yield in gm⁻². Five panicles were selected at random in every genotype and all the spikelets were separated from the panicle and filled grains were further separated and expressed

as grain number per panicle. A sample of 1000 seeds at random were taken from every genotype under both the conditions and weighed in gm and expressed as test weight. After harvest the shoot was dried and shoot biomass was recorded. Spikelet fertility was calculated as the number of filled spikelets/ total number of spikelets x 100 and expressed in per cent. Harvest index was calculated as ratio of economic yield to biological yield and expressed in per cent.

Statistical analysis: Two-way analysis of variance (ANOVA) was performed using Statistix 8.1 package. Statistical significance of the parameter means was determined by performing Fisher's LSD test to test the statistical significance.

Results and Discussions

High temperature was imposed at panicle initiation and with the help of data loggers the temperature inside the polyhouse was monitored on daily basis. The mean maximum temperature in the ambient condition was 34.9 °C and the average mean minimum temperature was 25.6 °C. Whereas, under polyhouse the mean monthly maximum temperature was 31.5 °C and the average of monthly minimum temperature was 24.2 °C. The difference in mean monthly maximum and minimum temperature was 3.5 °C and 1.4 °C, respectively (**Figure 1**).

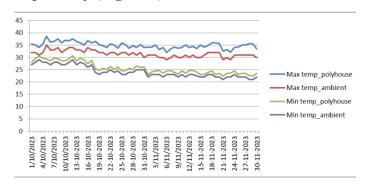


Figure 1: Temperature readings under polyhouse (heat stress) and ambient conditions (control)

Morpho-phenological traits:

The impact of high temperature was not significant on days to 50% flowering but the mean days to maturity



reduced by 3 days. At flowering, plant height reduced under high temperature among the genotypes. When compared to control maximum reduction was of 6 cm was noted in IL 19024 (**Table 1**). High temperature causes slower growth of the plants resulting in

reduction of plant height and 50% flowering (Pooja and Ajit, 2022). It has been stated that high temperature influenced negatively the growth parameters of rice plants by Korres *et al.*, (2017). In this study reduction of plant height as well as phonological traits was apparent.

Table 1: Impact of high temperature on morpho-phenological characters in rice genotypes

		ays to 5		Days to maturity		Plar	t height	(cm)		ophyll con		
Entry		flowerin	ıg				a	t flowerii	ng	(m	ng /g FW)	
	C	Ht	M	C	Ht	M	C	Ht	M	C	Ht	M
IET 29859	90	88	89	122	115	119	105	104	104	2.26	1.92	2.09
IL 19019	93	91	92	124	119	122	100	99	99	2.43	2.13	2.28
IL 19020	94	93	93	122	119	121	106	102	104	2.38	1.80	2.09
IL 19021	96	94	95	126	123	124	107	105	106	3.24	2.52	2.88
IL 19022	91	90	91	121	118	120	103	102	103	2.52	2.07	2.30
IL 19024	93	91	92	123	119	121	112	106	109	3.08	2.72	2.90
IL 19185	93	92	93	123	120	122	106	105	106	2.45	2.04	2.25
IL 19198	88	86	87	117	114	116	88	84	86	3.26	2.83	3.05
IL 19202	91	90	91	120	118	119	88	87	87	2.31	1.97	2.14
IL 19211	97	96	97	127	123	125	109	109	109	3.34	3.13	3.24
IL 19247	92	91	92	121	119	120	107	102	105	2.45	2.08	2.26
IL 19396	91	90	91	122	118	120	117	114	116	3.04	2.60	2.82
JB 680-2	90	89	90	121	119	120	112	107	109	2.41	2.00	2.21
JB 683-1	91	90	91	121	118	120	107	105	106	2.65	2.42	2.53
JB 687-3	91	90	90	121	119	120	115	113	114	3.07	2.63	2.85
JB 689-1	96	94	95	124	121	123	112	111	111	2.39	2.00	2.20
JBC 159-11	91	90	90	121	119	120	110	108	109	2.35	1.97	2.16
Krishnahamsa	91	90	90	121	118	120	104	101	102	3.26	2.85	3.06
MTU 1153	94	93	94	124	122	123	110	106	108	3.00	2.70	2.85
MTU 1156	92	91	91	121	119	120	104	102	103	3.20	2.73	2.97
MTU 1273	94	91	93	123	121	122	113	109	111	3.26	2.50	2.88
MTU 1290	92	91	92	122	119	121	112	110	111	2.79	2.61	2.70
MTU 1293	92	91	92	122	119	121	106	103	105	3.01	2.46	2.74
N-22	97	96	96	126	123	125	115	111	113	2.81	2.37	2.59
NLR 3776	97	95	96	127	123	125	110	105	108	2.62	2.13	2.38
NLR 3778	95	94	94	126	123	124	122	118	120	2.59	2.16	2.37
Mean	93	92		123	120		108	105		2.78	2.36	
LSD (T)		NS			0.53			1.04			0.026	
LSD (V)		1.62			1.93			3.76			0.095	
LSD (TxV)		2.29			2.73			5.32			0.135	
CV (%)		1.54			1.39			3.1			3.26	



Physiological traits:

Membrane thermostability: **Imposition** of high temperature led to reduction in the cell membrane thermostability in all the genotypes. Among the genotypes, MTU 1290 (77.9%), JB 683-1 (75.4%), MTU 1156 (73.9%), Krishnahamsa (73.3%), MTU 1153 (72.9%) and N22 (72.4%) had higher thermostability under heat stress treatment. JBC 159-11 (25.3%), IL 19247 (27.8%) and JB 680-2 (29.4%) had lowest thermostability. Maximum reduction in thermostability when compared to control (more than 50%) was in JB 687-3, JB 680-2, IL 19247 and JBC 159-11. The reduction in thermostability was minimum (less than 12%) in MTU 1290, MTU 1153, MTU 1156, JB 683-1 and N22 (Figure 2). The cell membrane is one of the most sensitive structures which is affected by variations in temperature (Hu et al., 2018). In this study it was noted that when subjected to high temperature stress the cell membranes were disrupted leading to a higher electrolyte leakage. Prasertthai et al., (2022) treated 28 days old seedlings of rice at 42 °C for 7 days and noted an increase in electrolytic leakage however N22 showed the highest heat tolerance displaying the lowest increase in electrolyte leakage. In this study also MTU 1153, MTU 1156, JB 683-1, N22 and MTU 1290 recorded lesser reduction in electrolytic leakage that could confer tolerance to increase in temperature.

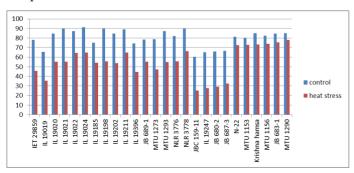


Figure 2: Impact of high temperature on Membrane thermo stability (%) in rice genotypes

Chlorophyll content: The mean chlorophyll content reduced from 2.78 in control to 2.36 under heat stress.

Highest chlorophyll content under high temperature stress was noted in IL 19211 (3.13 mg g⁻¹ FW) followed by Krishnahamsa (2.85 mg g⁻¹ FW), IL 19198 (2.83 mg g⁻¹ FW) and MTU 1156 (2.73 mg g⁻¹ FW). Lowest was in IL 19020 (1.8 mg g⁻¹ FW) followed by IET 29859 (1.92 mg g⁻¹ FW) and JBC 159-11 and IL 19202 (1.97 mg g⁻¹ FW). The per cent reduction of chlorophyll content compared to control was less than 10% in MTU 1153, JB 683-1, MTU 1290 and IL 19211. Whereas more than 20% reduction was observed in IL 19021, MTU 1273 and IL 19020 (**Table 1**). Chloroplast is a vital organelle for photosynthesis and contains photosynthetic pigments. The integrity of the organelle as well as biosynthetic pathways for chlorophyll formation is impacted by temperature. In a study conducted by Sánchez-Reinoso et al., (2014), high temperature at 35 and 40 °C resulted in reduction of chlorophyll a, chlorophyll b as well as total chlorophyll content in rice seedlings. Similar chlorophyll degradation was noted in KDML 105 rice seedlings when imposed to heat stress (Taratima et al., 2022). In the present study also there was a reduction in chlorophyll content in the tested entries. However, the reduction of chlorophyll content compared to control was less in MTU 1153, JB 683-1, MTU 1290 and IL 19211 indicating their tolerant nature when compared to other genotypes.

Grain yield and yield attributes:

Spikelet fertility (%): The mean spikelet fertility dropped from 92.0% in control to 75.7% under high temperature stress. Among the genotypes tested, higher spikelet fertility under stress conditions was observed in IL 19211 (87.2%) followed by MTU 1290 (85.4%), JB 683-1 (82.0%). Less than 65% spikelet fertility was noted in JB 680-2 (63.4%) and IL 19247 (64.8%) (Table 2).



Table 2: Impact of high temperature on yield attributes characters in rice genotypes

Entry	Spike	let fertility	/ (%)	Grain r	number per	panicle	1000	grain weigh	it (g)
Entry	C	Ht	M	С	Ht	M	С	Ht	M
IET 29859	93.0	76.0	84.5	146	114	130	20.5	18.2	19.
IL 19019	92.2	73.3	82.8	118	88	103	22.2	20.7	21.
IL 19020	88.5	68.1	78.3	85	62	73	22.4	21.2	21.
IL 19021	88.7	71.4	80.0	86	65	75	21.5	20.2	20.
IL 19022	86.2	65.9	76.1	81	60	71	20.3	18.7	19.
IL 19024	90.2	75.9	83.1	111	88	100	23.3	22.1	22.
IL 19185	91.4	73.2	82.3	106	82	94	24.1	23.2	23.
IL 19198	89.8	72.5	81.2	97	74	86	21.0	19.9	20.
IL 19202	91.9	81.7	86.8	124	107	115	20.9	19.5	20.
IL 19211	94.6	87.2	90.9	210	177	193	18.4	17.5	18.
IL 19247	91.1	64.8	78.0	92	59	76	23.1	21.2	22.
IL 19396	92.1	74.8	83.4	116	89	103	24.0	22.5	23.
JB 680-2	92.7	63.4	78.1	139	92	115	22.2	20.6	21.
JB 683-1	92.6	82.0	87.3	125	109	117	22.1	21.3	21.
JB 687-3	93.8	78.9	86.4	166	131	149	24.2	22.7	23.
JB 689-1	91.7	77.3	84.5	132	109	120	21.2	19.3	20.
JBC 159-11	91.8	78.7	85.3	123	100	112	17.2	15.7	16.
Krishnahamsa	92.0	78.6	85.3	104	88	96	19.2	17.9	18.
MTU 1153	94.5	75.6	85.1	155	118	136	22.1	20.6	21.
MTU 1156	93.3	77.2	85.3	140	112	126	22.2	20.6	21.
MTU 1273	92.6	75.7	84.1	150	115	132	24.4	22.8	23.
MTU 1290	94.6	85.4	90.0	140	123	131	24.7	23.3	24.
MTU 1293	92.5	73.4	82.9	135	102	119	21.2	20.0	20.
N-22	95.6	78.7	87.2	241	177	209	17.2	15.1	16.
NLR 3776	93.3	79.0	86.2	166	132	149	22.0	20.1	21.
NLR 3778	92.6	79.3	86.0	138	111	125	21.3	19.4	20.
Mean	92.0	75.7		132	103		21.6	20.2	
LSD (T)		5.6			2.86			0.10	
LSD (V)		1.5			10.3			0.37	
LSD (TxV)		2.8			14.5			0.52	
CV (%)		5.1			7.68			1.56	

Grain number per panicle: The average number of grains per panicle dropped from 132 to 103. N22 (177), IL 19211 (177), NLR 3776 (132) and JB 687-3 (131) had a higher grain number per panicle under high temperature stress and IL 19247 (59), IL 19022 (60) and IL 19020 (62) recorded lower number of grains per panicle (**Table 2**).

1000 grain weight (g): The average test weight reduced from 21.6g in control to 20.2g under high temperature conditions. The test weight was highest

in MTU 1290 (23.3g) followed by IL 19185 (23.2g). Lowest test weight was noted in N22 (15.1 g) and JBC 159-11 (15.7) (**Table 2**).

Total Dry matter (gm⁻²): The total dry matter reduced from 661 gm⁻² in control to 587 gm⁻² under high temperature conditions. Higher TDM under high temperature conditions were noted in JB 689-1 (680 gm⁻²) followed by Krishnahamsa (667 gm⁻²), IL 19185 (644 gm⁻²). Lowest TDM was in MTU 1293 (490 gm⁻²) (**Table 3**).



Table 3: Impact of high temperature on grain yield and yield attributes in rice genotypes

E 4	TDM	(gm ⁻²) at m	aturity	G	rain yield (gm- ²)	На	rvest inde	x (%)
Entry	С	Ht	Mean	С	Ht	Mean	С	Ht	Mean
IET 29859	743	583	663	535	345	440	41.9	37.1	39.5
IL 19019	590	522	556	501	351	426	46.0	40.1	43.0
IL 19020	691	597	644	523	387	455	43.1	39.1	41.1
IL 19021	633	596	615	493	408	451	43.7	40.5	42.1
IL 19022	626	588	607	511	396	454	44.8	40.3	42.6
IL 19024	592	537	565	429	348	389	41.9	39.2	40.6
IL 19185	688	644	666	529	388	458	43.4	37.5	40.5
IL 19198	622	577	600	469	386	427	43.0	40.1	41.5
IL 19202	681	614	648	514	403	459	42.9	39.6	41.3
IL 19211	621	582	602	468	416	442	42.9	41.5	42.2
IL 19247	640	566	603	462	364	413	41.9	38.7	40.3
IL 19396	689	585	637	516	407	462	42.8	41.1	41.9
JB 680-2	577	566	572	422	228	325	42.2	28.6	35.4
JB 683-1	679	596	637	515	443	479	43.2	42.8	43.0
JB 687-3	670	544	607	511	366	439	42.9	40.1	41.5
JB 689-1	677	680	679	507	388	448	42.8	36.0	39.4
JBC 159-11	640	574	607	483	364	424	42.9	38.7	40.8
Krishnahamsa	737	667	702	552	434	493	42.9	39.4	41.2
MTU 1153	664	629	647	501	420	460	42.8	40.0	41.4
MTU 1156	622	596	609	486	378	432	43.8	38.7	41.2
MTU 1273	607	630	619	472	345	408	43.3	35.4	39.4
MTU 1290	697	563	630	501	418	460	42.8	40.2	41.5
MTU 1293	711	490	600	551	345	448	43.6	41.4	42.5
N-22	767	598	683	577	392	484	42.9	39.7	41.3
NLR 3776	583	498	540	416	290	353	41.6	36.5	39.1
NLR 3778	751	627	689	571	434	503	43.2	40.9	42.0
Mean	661	587		501	379		43.1	39.0	
LSD (T)	<u></u>	19.6	<u> </u>	<u></u>	19.7			0.94	
LSD (V)		70.7			71.3			3.41	
LSD (TxV)		100.0			100.9			4.82	
CV (%)		9.9			14.1			7.27	

Grain yield (gm⁻²): Grain yield reduced from 501 gm⁻² in control to 379 gm⁻² under high temperature stress. Higher grain yield under high temperature was noted in JB 683-1 (443 gm⁻²) followed by NLR 3778 (434 gm⁻²), Krishnahamsa (434 gm⁻²), MTU 1153 (420 gm⁻²) and MTU 1290 (418 gm⁻²). Lower were in JB 680-2 (228 gm⁻²), NLR 3776 (290 gm⁻²) and IET 29859 (345 gm⁻²). More than 35% reduction was in IET 29859, MTU 1293 and JB 680-2 and less than 16% reduction was in MTU 1290, MTU 1153, JB 683-1 and IL 19211 (**Table 3**).

Harvest index (%): Harvest index reduced from 43.1% in control to 39.0% under high temperature conditions. Highest harvest index was noted in JB

683-1 (42.8%) followed by IL 19211 (41.5%). Lowest was in JB 680-2 (28.6%) (**Table 3**).

High temperature during the flowering period reduces the spikelet fertility (Zhang *et al.*, 2018). High temperature leads to poor anther dehiscence and less pollen grains on the stigma resulting in reduction of spikelet fertility (Zhao *et al.*, 2016). The number of spikelets reduced under high temperature majorly due to reduction in pollen production by preventing anther filling during the panicle initiation phase (Wang *et al.*, 2016). Hazra *et al.*, (2016), imposed high temperature at tillering, booting, panicle initiation as well as flowering stage in rice and reported that high temperature stress (36 °C) at PI stage resulted



in higher reduction of grain yield. Similarly high temperature exposure significantly decreased filled grains per panicle, 1000-grain weight and harvest index and increased the unfilled grains per panicle. Mahantashivayogayya *et al.*, (2016), screened forty rice accessions for heat tolerance and results revealed that EC792239, EC792285 and EC792185 which had heat tolerance, recorded more number of panicles and minimum per cent of chaffyness. In this study, reduction in grain yield and yield attributes were evident under high temperature stress and high yielding genotypes had higher spikelet fertility.

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Conclusion

High temperature has a negative impact on rice growth and development. It hampers the cell membranes leading to a higher electrolyte leakage as well as reduction in photosynthetic pigment. Impact of exposure at panicle initiation stage is evident from the reduction of yield and yield attributes of the genotypes tested, MTU 1290, MTU 1153, MTU 1156, JB 683-1, N22 and IL 19211 maintained higher membrane stability, lesser reduction in loss of chlorophyll content, high spikelet fertility and grain yield. These genotypes could thus be identified to be thermotolerant.

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RESEARCH ARTICLE

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Potential of Rice Straw as a Carbon Source in Organic Rice Farming and its Effect on Global Warming Potential

Surekha Kuchi*, Pragnya M, Kumar RM, Brajendra and Sundaram RM

ICAR-Indian Institute of Rice Research, Hyderabad, India.

*Corresponding author Email: surekhakuchi@gmail.com

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Abstract

The potential benefits of rice straw in organic farming in terms of rice productivity, soil organic carbon (SOC) stocks, carbon sequestration rate (CSR) and global warming potential (GWP) were studied for five years covering 10 wet (kharif) and dry (rabi) seasons with a wide seasonal temperature range (28-31 °C and 30-40 °C during wet and dry seasons, respectively). Field experiments were conducted at the ICAR-IIRR, Hyderabad, in a black clayey vertisol. The organic sources used were rice straw + green manure in kharif and rice straw + poultry manure in rabi. Organic system resulted in initial yield reduction by 15-20% than inorganic system and yields stabilized after two and five years in wet and dry seasons, respectively, with improved soil health parameters. The SOC stocks were higher with organics by 34-43% compared to inorganics after five years of study. The CSR was also positive with organics (0.97 and 0.57 t/ha/ year during wet and dry seasons, respectively) compared to inorganics (-021 and -0.33 t/ha/year during wet and dry seasons, respectively) which recorded negative C sequestration rate. Organics recorded favourable C sequestration even under dry situations which is more desirable to mitigate the adverse effects of global warming to certain extent by reducing CO, emissions. The global warming potential under organic system was 20% higher than in conventional system with increased CO, and CH₄ and reduced N₂O emissions. Since GWP was higher with rice straw, mixing of straw with other potential organics in proper proportions reduce the GWP in addition to carbon sequestration in organic rice cultivation.

Key words: Organic farming, rice straw, greenhouse gases, carbon sequestration, global warming potential

Introduction

One of the major challenges faced by humankind is to cope up with the changing climate and managing it for the sustenance of healthy life. IPCC, (2014) report reaffirms that human influence on the climate system is clear and recent anthropogenic emissions of greenhouse gases (GHGs) are the highest in history. According to Intergovernmental Panel on Climate Change (IPCC) (2014) data, it shows that 0.15 °C increase in temperature per decade, which is causing a drastic change in different farming systems and their productivity, is expected to increase by 1.7-4.8 °C by the end of the century.

Climate change and agriculture, being interrelated, do influence each other and the relationship between them is of high importance as the imbalance between world population and world food production is increasing. Agriculture is considered both as a contributor to climate change and a victim as well. Soil health degradation has emerged as a major factor responsible for the stagnation in agricultural production. Continuous use of inorganic fertilizers has not only brought about loss of vital soil fauna and flora but also resulted in loss of secondary and



micronutrients. Agriculture, responsible for 20-30% of global GHG emissions, contributes to climate change through practices such as excessive use of synthetic fertilizers, fossil fuel combustion during fertilizer production and intensive tillage operations. These practices release significant amounts of greenhouse gases such as carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O), which contribute to global warming. Global warming potential (GWP) is a metric that measures how much heat a greenhouse gas (GHG) traps in the atmosphere compared to carbon dioxide (CO₂) over a specific period of time.

Rice (Oryza sativa L.) is the major staple food crop in India, occupying around 45 million hectares (mha) of land and contributing about 135 million tons (mt) to the total food grain production (Agricultural Statistics at a Glance, 2023). Although rice production will be affected by climate change, rice farming also has the capacity to amplify the problem of GHG emission mediated global warming, which is a common phenomenon in rice growing countries especially in South Asia. Rapid mineralization of SOC compounds may lead to higher CO, flux to the atmosphere (Reicosky et al., 1995) thus leading to global warming. Soil C sequestration restores degraded soils, enhances biomass production and reduces the rate of enrichment of atmospheric CO₂ (Lal, 2004). Organic agriculture enables ecosystems to better adjust to the effects of climate change and improves carbon sequestration potential of the soil (Bhooshan and Prasad, 2011) and carbon sequestration is considered as a mitigation option to adverse effects of climate change. Keeping this in view, field experiments were conducted to study the influence of rice straw in organic farming on soil organic carbon, greenhouse gas emissions and global warming potential.

Materials and Methods

Field experiments were conducted over a spread of five years covering ten rice cropping seasons [five wet (WS) and five dry (DS)] on a deep black clayey vertisol (Typic pellustert) at the Indian Institute of Rice Research (IIRR) farm, Rajendranagar, Hyderabad. Organic and conventional farming systems were compared in terms of rice productivity, soil organic carbon (SOC) stocks, carbon sequestration rate (CSR) and global warming potential (GWP). The experimental soil characteristics were slightly alkaline (pH 8.2); non-saline (EC 0.7l dS/m); calcareous (free CaCO₃ 5.01%); with CEC 44.1 C mol (p+)/ kg soil and medium soil organic carbon (0.69%) content. Soil available N was low (228 kg/ha); available phosphorus was high (105 kg P₂O₅/ha); available potassium was high (530 kg K₂O/ha) and available zinc was also high (12.5 ppm). The temperature variation among the two seasons (wet and dry) was shown in **Table 1**.

Table 1: Mean Temperature (°C) range during crop growth period

Year	Wet se (August-D		Dry season (February-May)		
	Range Mean		Range	Mean	
1st year	28.5-30	29.3	32.9-38.1	35.5	
2 nd year	28.6-30.9	29.7	31.4-31.2	36.0	
3 rd year	29.5-30.6	30.0	30.5-39	34.8	
4 th year	29.3-31.4	30.0	33.9-40.3	37.3	
5 th year	28.5-31.4	30.3	33.0-40.3	37.5	
Overall range	28.5-31.4	-	30.5-40.3	-	

The four treatments consisted of: control (T1); 100% inorganic fertilisers (T2); 100% organics (T3) and 50% inorganics + 50% organics (T4, INM) with five replications. The organic sources used were green manure, dhaincha (*Sesbania aculeata*) + paddy straw during wet seasons (WS) and poultry manure + paddy straw during dry seasons (DS). Super fine rice varieties, BPT 5204 and Vasumati were tested in wet and dry seasons, respectively. The local recommended dose of inorganic fertilizers were given to conventional system @ 100-60-40 kg N, P₂O₅, K₂O/ha during both seasons through urea, single super phosphate, muriate of potash and 25 kg of Zinc sulphate, respectively. Nitrogen was applied in three equal splits at basal,



maximum tillering and panicle initiation stages while P, K and Zn were applied as basal doses only. Through organics, N dose was adjusted to recommended level based on their moisture content and 'N' concentration on dry weight basis. Organic fertilizers were incorporated one day before transplanting rice. Grain yield was recorded at the end of each season. At the end of 5 years, soil samples were collected from 15 cm depth and analysed for various parameters. Carbon stocks and C sequestration rate were computed using the following formulae (Lal *et al.*, 1998).

- 1) SOC storage (C stock t/ha) = $[\%C \times bulk \text{ density}]$ $\times \text{ soil depth (m)} \times 10000] / 100$
- SOC sequestered (t C/ha) = SOC (current) SOC (initial)

Integrated evaluation of GHG emissions expressed as GWP was computed for the current experiment by using the IPCC factors for calculating the combined GWPs for 100 years [GWP = 24.5 *CH₄+ CO₂+ 320 *N₂O kg CO₂-e ha⁻¹] from CH₄, N₂O and CO₂ efflux values under different treatment conditions (IPCC, 2007). The GHG emissions and GWP for inorganic fertilizers and organics sources used were calculated based on the studies of Snyder *et al.*, (2009); Bhattacharyya *et al.*, (2012); Wang *et al.*, (2019) and Fauzan *et al.*, (2021).

Results and Discussions

Grain yield trends

Grain yield trends showed that, during the wet season, yields in plots with fertilizer application and integrated nutrient management (INM) remained stable, ranging

from 5.2-5.5 t/ha and 4.7-5.2 t/ha, respectively. These yields were 15-20% higher than those in organic plots in the first two years, but organic yields improved over time, reaching comparable levels with inorganic treatments (4.8-5.2 t/ha) in later years. In the dry season, however, inorganics and INM consistently outperformed organics for four consecutive years, with organic yields matching those of inorganics and INM only in the fifth year (**Table 2**). Unfertilized control treatment recorded the lowest grain yields throughout the experiment. It was evident that all treatments with fertilization resulted in high yield increases, but yields were generally less in the DS.

Initially, organic farming led to a yield reduction of around 24% for rice, as reported by Mader *et al.*, (2002), but a gradual yield increase with organic methods over time was noted by Surekha *et al.*, (2010) and Urkurkar *et al.*, (2010). This may be due to a mismatch between the nutrient release from organic sources and crop demand, influenced by seasonal conditions in the initial years. Once soil fertility reached an adequate level, the organic system produced yields comparable to the conventional system. The slow, gradual release of nutrients from organic sources during the early years of conversion to organic farming did not immediately lead to higher yields. However, repeated applications of organic inputs over time built up sufficient soil fertility by enhancing soil biological activity (Surekha and Satishkumar, 2014).

Soil Organic Carbon (SOC) stocks and C sequestration rate (CSR)

The carbon stock and sequestration rate for each treatment were shown in **Table 3**. After five years, SOC stocks were significantly higher in organic

Table 2: Grain yield (t/ha) as influenced by nutrient sources

Sl. No	Year		Wet season				Dry season			
		Control	Inorganics	Organics	INM	Control	Inorganics	Organics	INM	
1	1st year	3.59°	5.52a	4.92 ^b	5.17 ^{ab}	1.92°	3.82ª	3.62 ^b	4.36a	
2	2 nd year	3.55°	5.63a	4.92 ^b	5.56a	2.33°	3.81a	3.11 ^b	3.81a	
3	3 rd year	3.32 ^b	5.50a	5.22ª	5.31a	2.77°	4.22ª	3.37 ^b	4.04ª	
4	4 th year	3.43 ^b	5.52a	5.60a	5.48a	1.98°	3.74ª	3.14 ^b	3.82a	
5	5 th year	3.34 ^b	5.29a	5.27a	5.02ª	2.13 ^b	4.21a	3.98ª	4.22ª	

Numbers followed by the same letter in each row are not statistically significantly



treatments, with values of 19.54 t/ha in the wet season and 17.55 t/ha in the dry season, compared to inorganic treatments, which recorded 13.63 t/ha and 13.05 t/ha, respectively. CSR were also positive for organic treatments, at 0.97 t/ha/year in the wet season and 0.57 t/ha/year in the dry season. In contrast, inorganic treatments showed negative CSRs of -0.21 t/ha/year and -0.33 t/ha/year for the wet and the dry seasons, respectively. The INM treatment showed intermediate values, while the lowest SOC stock levels were recorded in the control treatment.

Carbon sequestration rate was nearly half during dry season which recorded higher temperatures than that of CSR during wet season that favoured the loss of SOC stocks due to faster decomposition and mineralization. The treatment, 100% organics recorded 43% and 34% higher SOC stocks followed by INM treatment

that recorded 34% and 20% higher SOC stocks over 100% chemical fertilisers during wet and dry seasons, respectively. Thus, it was evident that organic rice farming can lead to better carbon accumulation under varied climatic conditions and increasing organic carbon in agricultural systems has been reported as an important mitigation option by IPCC (Muller, 2009). Bhattacharyya et al., (2012) also noticed that carbon storage rate was also found to be significantly higher (0.35 Mg C ha⁻¹ y⁻¹) in the application of rice straw. An increase in soil carbon by 15-28% in organic systems was also reported by Paul (2003). Similarly, higher values of SOC by $0.18 \pm 0.06\%$, for C stocks by 3.50 ± 1.08 Mg C ha⁻¹ and for sequestration rates by 0.45 ± 0.21 Mg C $ha^{-1}y^{-1}$ in organically farmed soils over non-organic management was reported by Gattinger et al., (2012).

Table 3: Soil Organic carbon (SOC) stocks and C sequestration rate after five years

0		,	1	•	
Treatments	Soil organic	Bulk density	Carbon stock	SOC sequestered	C seq. rate
Treatments	Carbon (%)	(g/cc)	(t/ha)	(t/ha)	(t/ha/yr)
		Wet	season		
Control	0.59	1.36	12.04	-2.66	-0.53
Inorganics	0.64	1.42	13.63	-1.07	-0.21
Organics	1.01	1.29	19.54	4.84	0.97
INM	0.91	1.34	18.29	3.59	0.71
		Dry s	season		
Control	0.54	1.38	11.18	-3.52	-0.70
Inorganics	0.60	1.45	13.05	-1.65	-0.33
Organics	0.90	1.30	17.55	2.85	0.57
INM	0.79	1.32	15.64	0.94	0.19
Initial value	0.69	1.42	14.7	-	_

Global warming potential (GWP)

The global warming potential along with the emission of greenhouse gasses such as CO₂, CH₄ and N₂O with usage of inorganic fertilizers and organic fertilizers were presented in **Table 4.** For inorganic fertilizers, urea contributed the most to emissions, particularly during application, where N₂O emissions were high compared to organics. Combined across both seasons, inorganic sources emitted a total GWP of 13,157 kg CO₂-e/ha. Organic applications resulted in higher CO₂ and CH₄ emissions but lower N₂O emissions compared to inorganics. The total emissions from organic sources across both seasons were with a GWP

of 15,909.6 kg CO₂-e/ha. Greenhouse gas emissions and GWP from organics were higher in *rabi* season than in *kharif* season. The application of organic fertilizers increased the CO₂ and CH₄ emissions by 15.5% and 33.2% and reduced N₂O emissions by 27.4% over inorganics though there was an increase of 20.9% in GWP with the application of organics. The data suggests that using organic fertilizers, such as rice straw and poultry manure, increased total greenhouse gas emissions compared to inorganic fertilizers. However, it also reduced N₂O emissions, a particularly potent greenhouse gas, by 27.4%



over inorganics. This trade-off highlights that while organic fertilizers may contribute more to CO₂ and CH₄ emissions in rice, they offer environmental benefits by lowering N₂O emissions, which could be advantageous for climate mitigation strategies focused on this gas. Wang *et al.*, (2018) also reported that rice straw increased CO₂ and CH₄ emissions but reduced N₂O emissions. According to He *et al.*, (2024) the straw return might facilitate soil respiration and oxygen depletion, leading to oxygen limitation and eventually spurring the reduction of N₂O to N₂ during denitrification, especially in oxygen-depleted paddy fields. Thus, it leads to a reduction in N₂O emissions.

GHG emissions when rice straw along with green manure (7764 kg CO₂-e/ha) applied were less compared to the rice straw and poultry manure (8145 kg CO₂-e/ha). The high C/N ratio in rice straw also contributes to increased GHG emissions from organic sources. Poultry manure and green manure have lower C/N ratios than rice straw, which contains abundant cellulose and hemicellulose (Li *et al.*, 2023). Gao *et al.*, (2023) reported that green manure increases

soil fertility and rice yields without increasing CH₄ emissions in green manure-rice system than fallowrice practice. Bayer et al., 2014 reported that the high C/N ratio of rice straw increased the metabolic C substrate available to methanogenic bacteria, thus promoting the production of CH₄ and lead to carbon loss by accelerating soil organic matter mineralization through the priming effect. Khosa et al., (2010) reported that rice straw compost reduced the GHG emissions compared to sole application of straw/ green manure, as direct straw use leads to increased GHGs. He et al., (2024) reported that GHG and GWP were influenced by a combination of straw size, straw return method and straw amount and recommended straw incorporation > 7.5t/ha with size ≥ 5 cm for reducing GHG emissions and GWP from straw return in paddy fields. Though rice straw has high potential as carbon source in organic farming, due to its contribution to global warming, its positive effects may not be visible. Hence, it is essential to find out suitable organic sources and their proportions to mix with straw so that GWP from straw can be reduced.

Table 4: GHG emissions and GWP from inorganic and organic sources (kg CO₂-e ha⁻¹ yr⁻¹)

		GHG Emissions		GWP
Inorganics	CO,	CH ₄	N,O	
Kharif	(kg CO,-e ha-1)	(kg CO,-e ha-1)	(kg CO,-e ha-1)	(kg CO,-e ha-1)
During Manufacture				
Urea 100 kg	310	7.77	0.93	320
Phosphate (SSP) 60 kg	60	2.268	0.372	60
Potash (KCl) 40 kg	28	0.084	0.124	28
During Application				
Urea-N 100 kg N/ ha	2412.83	3241.00	516.67	6170.50
Rabi				
During Manufacture				
Urea 100 kg	310	7.77	0.93	320
Phosphate (SSP) 60 kg	60	2.268	0.372	60
Potash (KCl) 40 kg	28	0.084	0.124	28
During Application				
Urea-N 100 kg N/ ha	2412.83	3241.00	516.67	6170.50
TOTAL (Kharif + Rabi)	5621.67	6502.24	1036.19	13157.00
ORGANICS				
Kharif				
During Application				
Rice straw + Green Manure @100 kg N	3097.50	4294.50	372.00	7764.00
Rabi				
During Application				
Rice Straw + Poultry Manure @100 kg N	3397.50	4368.00	380.14	8145.64
TOTAL (Kharif + Rabi)	6495.00	8662.50	752.14	15909.64
% Increase/decrease with organics	15.5%	33.2%	-27.4%	20.9%



Conclusion

Organic farming in rice production, though initially yields less, can match conventional yields over time by enhancing soil fertility through sustained nutrient release. Organic practices significantly improved soil organic carbon stocks and sequestration rates, supporting soil resilience and adaptation for climate change. However, organic treatments increased CO2 and CH₄ emissions but reduced N₂O emissions by 27.4%, indicating potential climate change mitigation benefits. Overall, organic farming enhances soil health and carbon storage supporting long-term sustainable productivity amidst climate change. Alternative ways of effective straw utilization like composting must be explored to reduce the GHG emissions and also to obtain fullest benefits from rice straw utilization in organic farming.

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RESEARCH ARTICLE

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Study of Skewness and Kurtosis for Spikelet Sterility and Yield Related Traits in Segregating Generations of a Rice (*Oryza sativa* L.) Cross to Identify Gene Action

Priyanka Kumari^{1,2}, Jenny Priya Ekka¹, Swapnil³, Krishna Prasad¹, Ekhlaque Ahmad^{1*} and Manigopa Chakraborty¹

¹Department of Genetics and Plant Breeding, Birsa Agricultural University, Ranchi, Jharkhand;

²Department of Plant Breeding and Genetics, Veer Kunwar Singh College of Agriculture,

Bihar Agricultural University, Sabour, Bhagalpur,

³Department of Genetics and Plant Breeding, Centurion University of Technology and Management,

Paralakhemundi, Odisha.

*Corresponding author Email: ekhlaque.bau@gmail.com

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Abstract

An investigation was performed in the F_6 and F_7 generation of the cross Pusa-1176 x BPT-5204. The phenotypic screening was done intensely for spikelet sterility in F_6 generation (*Kharif*, 2021) based on which some families were selected and grown as F_7 generation (*Kharif*, 2022) that were sown as panicle to progeny rows at two different dates as set I and set II with the interval of 15 days in order to study the influence of environment on the traits under study. The segregates were studied for skewness and kurtosis to identify the presence of gene interactions in 13 quantitative traits including grain yield per plant (Fisher *et al.*, 1932). The results displayed that various traits exhibited positive skewness along with platykurtic and leptokurtic frequency distribution in set I and set II. Similarly, a combination of negative skewness with platykurtic and leptokurtic frequency distribution was observed for diverse traits in set I and set II. Normal curve without skewness indicating symmetry of the trait and platykurtic frequency distribution was observed for L/B ratio in set I and for 100 seed weight in set II.

Keywords: Kurtosis, Gene Action, Skewness, Spikelet, Sterility

Introduction

Rice (*Oryza sativa* L.) belongs to Poaceae family and is one of the major staple foods for more than half of the world's population. It is also an essential commodity for food and nutritional security since it provides 20% of the regular calorie intake (Ricepedia, 2020). It is cultivated nearly on 162 million hectares of land in a wide range of environments with an annual production of about 756 million tonnes globally (FAO, 2022) that mostly does not reach its potentiality in terms of production and productivity due to various

factors affecting the yield of the crop such as biotic and abiotic stresses that are unpredictable in nature. Thus, a breeder should be capable of selecting the better-performing genotypes from a large population which requires intensive studies on various yield and yield-attributing traits. The extent of selection pressure given to the population depends on the study of a frequency distribution that is analyzed via skewness and kurtosis estimation (Choo and Reinbergs, 1932). The nature of gene action is



revealed by the estimates of skewness whereas the number of genes controlling the trait is determined by kurtosis (Savitha et al., (2015) and accordingly choice is made whether selection should be mild or intensive. Thus, to enhance the selection efficiency and breeding strategy, appropriate information on the gene interactions is essential which may lead to progressive research work thereby improving the performance of the population. The present investigation was thereby undertaken in the F₂ generation of the cross Pusa-1176 x BPT-5204. The main objective of the experiment was to assess the existence of variability among the characters under study, nature of gene action, number of genes controlling the trait and statistical analysis through frequency distribution patterns based on skewness and kurtosis (Roy, 2000, Robson, 1956).

Materials and Methods

The present investigation was carried out at the experimental plot of Birsa Agricultural University, Kanke under the rainfed conditions of Jharkhand, India during Kharif 2021. The phenotypic screening was done intensely for spikelet sterility in 23,083 F₆ plants (from 482 families) of the cross Pusa-1176 x BPT-5204 as large number of plants in a family showed sterile panicles as well as segregation among the traits under study. The parents of Pusa-1176 and BPT-5204 are [Bindli mutant 34 (BM34) x ARC line] and [GEB-24 x T(N)1 x Mahsuri], respectively. The parent, Pusa-1176 is an aromatic rice and the other parent BPT-5204 is resistant to blast. Also, both the parents were well suited to rainfed and shallow land. As the families of F₆ generation showed segregation among the traits under study, 15 families comprising of 158 sterile F₆ plants were selected based on the maximum number of sterile progeny plant with sterility % above 55% within the family. In the next generation (F₇ generation), it was sown as panicle to progeny rows

on the seed bed nursery and thereafter transplanted into the field. Sowing was done at different dates designated as set I and set II with the interval of 15 days in order to study the influence of the environment on the traits under study. The method of sowing adopted was panicle to progeny row. The transplanting was done by maintaining row-to-row distance as well as plant-to-plant distance of 20 cm. Other recommended agronomic practices were followed throughout the crop growth period. The data was recorded on individual plants representing each of the segregants due to segregation pattern observed among the traits under study. The traits for which observations were taken are days to flowering, number of tillers, plant height, panicle length, panicle number, number of filled spikelets, number of unfilled spikelets, spikelet fertility, grain length, grain width, L/B ratio, 100 seed weight and grain yield per plant. After that statistical analysis was done based on the estimation of skewness and kurtosis which was calculated using the frequency distribution of the characters under study (Kapur, 1981; Savitha et al., 2015).

Results and Discussions

The study of frequency distribution via skewness and kurtosis provides information about the nature of gene action and the number of genes controlling the traits respectively. The genes controlling the character with skewed frequency distribution tend to be predominantly dominant regardless of whether the effect on the trait has been enhanced or reduced. The results obtained have been presented in **Table 1.** The frequency distribution was found to be positively skewed and platykurtic for the number of panicles, number of unfilled spikelets, 100 seed weight and seed yield per plant in set I and for days to flowering, number of tillers, number of panicles and spikelet fertility in set II. Positive skewness in the segregants indicates its association



with complementary gene actions and platykurtic distribution shows that it is controlled by a large number of genes (Figure 1). Similar results were reported by Kiran et al., (2012) for number of tillers panicle length and grain yield per plant, Harshiya and Jagadeesh (2014) for plant height and test weight, Raghavendra and Hittalmani (2015) for number of filled grains and grain length, Savitha et al., (2015) for 1000-grain weight and single plant yield and Rani et al., (2016) for number of tillers per plant, panicle length and also for grain yield per plant. It is also supported by Sheshaiah et al., (2018) for panicle length and grain yield per plant. Sushma Lilly et al., (2018) reported for the number of filled grains and grain length whereas Prisca Seeli et al., (2021) reported for number of unfilled grains.

The positively skewed and leptokurtic frequency distribution was observed for days to flowering, number of tillers, plant height and number of filled spikelets in set I and for plant height, number of filled spikelets, number of unfilled spikelets and seed yield per plant in set II that exhibited the presence of complementary gene action by the estimation of skewness and the leptokurtic nature indicated that it is controlled by a few genes. The results were appropriately supported by Kiran et al., (2013) for panicle length, Harshiya and Jagadeesh (2014) for spikelet fertility, Manjappa et al., (2014) found positively skewed distribution for grain yield plant per plant and tiller number, Lestari et al., (2015) for panicle weight, Savitha et al., (2015) and Nikhitha et al., (2020) for the number of productive tillers per plant, panicle length and Govintharaj et al., (2017) for number of grains.

The presence of gene interactions is represented by a positive value of kurtosis while it is negative or close to zero in the absence of gene interaction (Kotch *et al.*, 1992). Negatively skewed and platykurtic frequency

distribution was observed for spikelet fertility and grain width in the set I and for panicle length, grain width and L/B ratio in set II. It is also supported by Kiran et al., (2013) who noticed duplicate interaction for panicle length, Harshiya and Jagadeesh (2014) for panicle length, grain yield per plant, grain length, grain breadth, length to breadth ratio, Manjappa et al., (2014) found negatively skewed distribution for plant height, days to flowering, days to maturity, panicle length, panicle exertion, test weight and spikelet fertility, Lestari et al., (2015) reported that the panicle length and weight. Savitha et al., (2015) found platykurtic with left-skewed distribution for days to 50 per cent flowering, plant height, number of productive tillers per plant, panicle length, hundred-grain weight and single plant yield. Vijaya and Shailaja (2016) reported platykurtic and negatively skewed distribution for plant height, Rani et al., (2016) recorded for test weight, grain length and grain breadth.

Negative skewness and leptokurtic frequency distribution were found for panicle length and grain length in set I and for grain length in set II. Similar results were supported by Kiran et al., (2013) for plant height, Savitha et al., (2015) for the number of productive tillers per plant, panicle length and Sushma Lilly et al., (2018) for L/B ratio. The frequency curve depicting negative skewness is associated with duplicate (additive x additive) gene interactions and it is controlled by many genes or by few genes indicating platykurtic or leptokurtic nature respectively. Normal curve without skewness indicating symmetry of the trait and platykurtic frequency distribution was observed for L/B ratio in set I and for 100 seed weight in set II. A similar result was reported by Kiran et al., (2013) for test weight indicating complete ambi-directional epistasis.



Table 1: Estimates of skewness and kurtosis for different characters under study in F_7 generation of the cross Pusa-1176 and BPT-5204 for both sets

Sl.	Characters	M	ean	Skev	vness	Kuı	tosis
No.	Characters	Set I	Set II	Set I	Set II	Set I	Set II
1	Days to Flowering	89.40	87.77	0.69	0.75	1.40	0.18
2	Number of Tillers	11.02	21.59	0.87	0.67	1.02	0.49
3	Plant Height	93.78	77.76	1.09	0.68	4.86	1.36
4	Panicle Length	23.66	22.01	-0.13	-0.20	1.18	0.54
5	Panicle Number	9.76	18.4	0.82	0.90	0.92	0.95
6	Number of Filled Spikelets	69.72	36.27	0.84	1.19	1.12	1.28
7	Number of Unfilled Spikelets	58.63	80.86	0.77	0.76	0.23	1.31
8	Spikelet Fertility	54.77	30.89	-0.13	0.56	-1.12	-0.92
9	Grain Length	0.90	0.89	-0.25	-0.92	1.19	3.11
10	Grain Width	0.28	0.28	-0.01	-0.12	-0.49	-0.14
11	L/B Ratio	3.23	3.14	0.00	-0.11	0.06	0.47
12	100 Seed Weight	1.74	1.82	0.24	0.02	0.66	-0.27
13	Grain Yield per plant	12.83	10.37	0.24	3.40	0.66	16.88

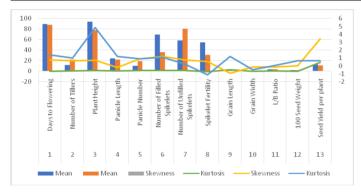


Figure 1: Graphical representation of the estimates of skewness and kurtosis

Conclusion

To increase the efficiency of selection in breeding programs among large populations, breeders should have adequate knowledge of the prevailing amount of gene interaction and the number of genes governing the trait. The results indicated that mild selection would be sufficient for improvement in the characters number of panicles, number of unfilled spikelets, 100 seed weight and seed yield per plant in the set I and for days to flowering, number of tillers, number of panicles and spikelet fertility in set II whereas stringent selection might be deployed for trait improvement in panicle length and grain length in set I and only for grain length in set II. Therefore, it is concluded from the study that progress in improving the performance of genotypes would be higher under complementary

gene interaction as compared to duplicate gene interaction.

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RESEARCH ARTICLE

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Economic Worth Assessment of Implementing Baler Technology for the Management of Paddy Straw

Sharma A¹, Mavi HK^{2*} and Brar AS³

¹Department of Farm Machinery and Power Engineering, Punjab Agricultural University, Ludhiana; ²Department of Economics and Sociology, Punjab Agricultural University, Ludhiana; and ³Krishi Vigyan Kendra, Moga, Punjab

*Corresponding author Email: mavihk05@pau.edu

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Abstract

Mechanical collection of paddy straw with the help of the baler technology from the combine harvested paddy fields is a prevalent paddy straw management technique in the country. Baler technology manages the paddy straw by collecting it with the rectangular baler, rake and stubble shaver machines and preventing the on-farm paddy straw from burning. Finding the financial constraints for creating a tailored recruiting system for technology implementation at the field level in paddy straw management was one of the main goals of this study. The economic worth assessment showed that the total operating cost for systems I and II was estimated as ₹ 4285.74 and 2374.34 per hour and the net benefit was ₹ 998.09 and 991.16 per hour. The benefit-cost ratio of the system I and II of baler technology was found to be 1.23 and 1.42 and the break-even usage of the baler technology was assessed at 154.66 and 109.65 hours per year of machine operation, which is less than the machine's useful life used under baler technology implementation. Hence, machine owners, local service providers (LSPs) and forward-thinking farmers can seize this opportunity as a highly lucrative endeavour. Steering clear of on-farm burning of paddy straw is a crucial step toward reducing environmental pollution, making it a worthwhile contribution to environmental conservation.

Keywords: Baler, Break-Even Point, Burning, Cost, Paddy, Payback Period.

Introduction

Globally, India has the largest area (44.6M ha) under paddy and is second only to China in paddy production. Paddy constitutes 52 per cent of food grain production and 55 per cent of total cereal production (Hira *et al.*, 2015). Paddy-wheat cultivation is a common farming exercise in northern Indian region. India produces 117 Mt paddy per year and such a vast amount generates 185-200 Mt paddy straw. Twenty-five per cent of Asia's total paddy straw production comes from India (Bhuvaneshwari *et al.*, 2019). The states of Haryana and Punjab are recognized as India's "food bowl" because they contain a highly

fertile paddy-wheat area on the Indo-Gangetic plain. Only in Punjab are paddy and wheat cultivated in more than three and 3.5 million hectares of land, with about 22 million tonnes of paddy residue per year (Mander, 2018).

Residues from paddy crops consist of biomass remaining in the field following grain harvesting and other valuable elements. As a byproduct of harvesting rice, paddy straw is created. Irrespective of whether it was harvested manually or by machinery, it is taken out during harvest with the rice grains and then stacked up or scattered across the field. As labour



costs rise and rice production intensifies in Asia, combine harvesters are becoming more common in rice fields during harvest season. However, the loose rice straw left behind by these machines complicates the collection and transportation process, resulting in increased expenses and time requirements. Asia annually generates between 600 to 800 million tons of rice straw, with global production nearing 1 billion tons each year (Sarkar and Aikat, 2013; McLaughlin et al., 2016). Farmers chose to burn rice straw as a fast fix to swiftly eliminate the biomass and get the field ready for the forthcoming crop since they are unaware of the straw's other uses. Burning rice straw in the field poses risks to both human health and the environment, leading to increased greenhouse gas emissions. For every kilogram of dry rice straw burned, emissions of 0.7 to 4.1 grams of CH_4 and 0.019 to 0.057 grams of N₂O occur, along with additional gaseous pollutants such as SO2 and NOx. Additionally, though to a lesser extent, the burning process also releases dioxins and furans (Oanh et al., 2011; Jenkins et al., 2003). Burning rice straw serves as a crucial origin of aerosol particles, impacting both local air quality and the Earth's radiation balance (Engling et al., 2009). These particles include small dust particles (PM_{2.5}) and coarse dusty particles (PM₁₀) (Chang et al., 2013). Furthermore, biomass loses its potential energy content (Tabil et al., 2011). Ozone levels in the lower atmosphere are also raised by burning agricultural crop residue (Kumar et al., 2015). Burning agricultural leftovers raised soil temperatures to 33.8-42.2 °C to a depth of 1 cm, as Gupta et al., (2004) reported. This has an impact on soil ecology. Thus, due to the increased soil temperature, the favourable microbial population in the soil decreases to a depth of 2.5 cm and around 23-73% of the nitrogen in different kinds is taken from the soil. The burning of residue raises the soil's temperature considerably, which causes the upper three inches of the soil's carbon-nitrogen (C-N) balance to shift quickly. Nitrogen transforms

intonitrate, while carbonise mitted into the environment as CO₂. Approximately 824 thousand metric tons of nutrients-nitrogen, potassium and phosphorus-are lost from the soil due to this process (Gupta *et al.*, 2004). Two more approaches to controlling paddy straw are baling the straw and integrating it into the field, in addition to infield burning (Singh *et al.*, 2005).

Rice straw management and collecting continue to be challenging tasks. In the past, several methods have been experimented with for collecting and managing rice straw. These include field cubers, stack wagons, buck rakes, standard three-wire balers, traditional large roll balers and high-flotation big roll balers, were documented by Dobie et al., (1977). Dobie concluded that, for a 16 km haul distance, the most economical system to be provided would be a 1.2 m comprehensive extended roll baler system. Though he had yet to test a big rectangular baler, Dobie (1980) also suggested that a substantial rectangular bale system would be more promising than the massive roll bale system. Rice straws should be gathered and used to provide electricity or be subject to alternative management alternatives for financial and environmental reasons. For individuals looking to develop a cost-effective structure using sustainable materials, straw bale construction may be one of the greatest options (Bhattarai et al., 2012).

A big rectangular baler for gathering rice straw was tested by Jenkins *et al.*, (1985). The performance of balers was contrasted with that of alternative handling methods and large roll balers. Economically speaking, large bale systems are preferable to tiny, rectangular bale systems. Big rectangular bales worked well for transportation, but they needed to be stored under cover to prevent spoiling. Large roll bales may be stored outside without losing much dry matter, although they were less popular for long-distance transportation. Straw delivery costs vary depending on the kind of packaging, distance



travelled, necessary processing steps and method of use. Timely use of the field for following planting, collecting and baling paddy straw in the combined harvested field is a suitable and financially feasible solution (Tathod et al., 2015). The economic and environmental performance of straw baler (Model 338 make: John Deere), for the collection of paddy straw generated after mechanical harvesting by combine harvester was determined by Pal et al., (2019). Straw baler facilitated the collection of paddy straw of 4.36 tons/ha at a cost of just ₹1650. Nghi et al., (2015) conducted experiments which revealed an internal rate of return of 38%, a payback period of 2.1 years and a baling cost of US\$ 19.0 per ton of rice straw. Along with the baling fee, the transportation cost ranges from US\$24 for a 100-kilometer journey to US\$32 for a 150-kilometer distance. In 2013, Shafie et al., undertook a logistic cost analysis focusing on rice-straw-based power generation in Malaysia. They developed mathematical logistic models to assess the collection, storage and transportation costs associated with this form of power generation. The ideal quantity of storage facilities and the location of the power plant were determined using the optimization approach. According to the results, the transportation expenses for conveying rice straws to collection centres primarily stemmed from the influence of truck capacity, constituting 89.9% of the total expenditure in transportation. Transportation costs were also the highest, accounting for 54% to 63% of the overall logistic costs. The number of storage facilities might be decreased to lower the cost of transportation.

The baler has been introduced in India to recover straw from the combined harvested paddy field. Baler machine owners have the option to utilize their equipment on their own land and also generate revenue by offering custom hiring services to fellow farmers. Sharma and Chandel (2016) assess a baler's performance on loose paddy straw (system A) after

the use of a stubble shaver (system B) and following the use of a rake in addition to the stubble shaver (system C). When the feed rate of paddy straw rose from 1.12 to 4.22 tonne per hour, the number of bales per hectare and density of bales also increased. The maximum feed rate was seen when the stubble shaver and rake were used before the baler (system C). Systems A, B and C had field capacities of 0.35, 0.40 and 0.53 ha per hour and ranged in the number of bales per ha from 266-292, 298-332 and 126-149, respectively. For systems A, B and C, the corresponding mean fuel consumption was 5.0, 10.0 and 12.0 litre per hour. System C had a more significant mean per centage increase in bale density, quantity of bales and baler productivity than did systems A and B. System C had the highest benefitcost ratio at 1.16:1, while systems B and A had the highest ratios at 1.06:1 and 0.85:1, respectively. With systems B and C, the net savings per hectare were Rs. 471.05 and 1537.59, respectively. The economic and practical effectiveness of a straw baler in a combine-harvested rice field is assessed by Singh et al., (2005). In the paddy field harvested by a combine, the baler's field capacity was 0.26 ha per hour; however, in the field where the stubble shaver was used before baling, it was 0.36 ha per hour. Bales ranged in size from 800×450×450 to 900×450×450 mm and correspondingly, their weight varied between 18 and 28 kg. In combine-harvested paddy fields, 205 bales were made; in stubbleshaved paddy fields, 425 bales were formed. The straw baler's economic analysis showed that baling in fields with stubble shaved fields costs Rs. 2276 per hectare, while transporting the bales costs Rs. 4400 per hectare. Baling in stubble-shaved fields costs Rs. 6676 per hectare, including bale transportation. The machine's extremely high shipping cost is the single factor keeping it from becoming more widely used. Straw sales brought in a total of Rs. 5865



per hectare. According to Mangaraj & Kulkarni's (2011) research, the cost of making a single twinetied bale for wheat and paddy was ≥ 5.00 and ≥ 2.75 , respectively. The net income from collecting and baling straw using the machine was ₹ 607.00 per hectare for rice straw and, ₹ 235.00 per hectare for wheat straw, assuming nominal prices of ₹ 0.25 kg⁻¹ for paddy straw and ₹ 0.75 kg⁻¹ for wheat straw. Not all of the villages in the area have had access to baler technology yet. Baler technology service providers, or new entrepreneurs, may get started and have a great potential to offer this service to end users nearly all year round. For this purpose, from the owner or custom operator's point of view, the economic worth assessment of baler technology is a dire need. Therefore, determining critical indicators related to the economic worth assessment of baler technology used by the service providers/progressive farmers to manage (collect) paddy straw is the need of the hour. So, the present study assessed the economic worth of baler technology implemented for managing paddy straw by its mechanical collection.

Materials and Methods

Data collection

The study was conducted in the district Moga of Punjab in India. Moga district is located in the central zone of the state, having a plain geographical area of 2230 sq. km., which comes to 4.42 per cent of Punjab state. It stretches between 75 degrees 15'E and 75 degrees 25'E longitude and 30 degrees 35'N and 31 degrees 15'N latitude. Farmers in the Moga district primarily rotate their crops using paddy wheat. The district of Moga is expected to have 195,237 hectares of total agricultural land, of which 176,000 hectares (91.27%) are under rice cultivation and 175,000 hectares (89.63%) are under wheat crop cultivation. In this study, secondary data were gathered from a variety of sources. Studies journals, published

studies, progressive farmers and machine owners/ operators served as the primary sources of data. Using an information panel, several crucial operational data were gathered from primary sources and research on baler technology.

Baler technology

The baling process, common automated a technique for gathering hay, straw and other fibrous materials through densification, uses baler technology. This process effectively facilitates the removal of materials from the field, simplifies the transportation and manipulation of bales, addresses shortages and provides flexibility in storage options. (Van-Hung et al., 2016; Guerrieri et al., 2019; Lemos et al., 2014). This mechanical process of collecting straw is performed with the help of tractor-operated machinery. It requires three tractor-driven machines: a stubble shaver for cutting, a rake for lining and gathering and a baler for making bales.

Stubble shaver

Paddy stubble was trimmed with a stubble shaver when it was standing. The machine operates in 2nd low gear, maintaining engine rpm between 1500 and 1700, which can vary depending on the load of paddy straw. It features two blades mounted on a vertical shaft, enclosed within a frame on the top and all four sides. Through a gearbox, a tractor's PTO (power take-off) shaft rotates the shaft.

Rake

Following the stubble shaver's operation, leftover straw can be gathered in a small breadth with a tractor-driven rake equipment that needs between 40 and 50 horsepower (hp) of power. The rotating rake has a working width of 3.5 meters and a transport width of 1.5 meters. It weighs between 350 and 450 kg. The rake's job is to gather loose and chopped paddy straw from the field and create a windrow in the smaller area so that the baler machine has thick straw to work with.



Adjusting to the load of paddy straw, this machine is frequently operated in third low gear, maintaining engine RPMs between 1500 and 1700.

Straw Baler

In the present study, a rectangular baler was considered which picks up a pre-cut crop from a windrow and feeds it into the bale chamber, where it is cut again, compacted, tied and discharged out the back of the machine. It has the ability to regulate the bale density and level of compaction. The machine also has a measuring system for changing the bale length. The hypoid gear, which had spiral interactions between the crown wheel and pinions, served as the baler's main drive (Figure 1). This had the benefit of having a larger gear tooth contact area than with conventional gear meshing, which increased longevity and consistent power flow (Sharma and Chandel, 2016). In order to ensure smooth operation of the baler, a broad flywheel was installed in front of the transmission system to absorb the stresses of the ram. Depending on the amount of paddy straw, the baler was run in 2nd low gear in the current investigation, with engine rpm ranging from 1500 to 1800.

Mechanical operation of bale formation

Initially, the stubble shaver machine is employed to harvest the stubbles, left in the field subsequent to combine harvesting of the paddy, from the base level. Subsequently, the lining operation is executed by the rake machine, followed by the gathering and formation of rectangular bales by the baler. It automatically picks up the straw from the field with the help of a reel and transfers it into the bale chamber with the help of a feeder and then the straw is compressed with the reciprocating ram. Straw baler made highly compressed, firm and perfectly shaped bales (**Figure 1**), reducing the storage space (Sharma *et al.*, 2014). All these machines are commercially available and can be operated by the 40-50 hp tractor.





Figure 1: Operational view of rake and baler machine

Economic assessment of baler technology for managing paddy straw

The economic assessment of agricultural systems is necessary to understand peasant practices and create and disseminate innovative systems. Agricultural machinery costs assessment is critical when considering structural or technological changes. Because the advantages and costs of machinery investments in agriculture accumulate over a number of years, it is more complicated than dealing with yearly monetary inputs like seed or fertilizer. A stream of cash inflows and outflows is connected to every machine action. In this study, the economic evaluation of baler technology for managing paddy straw was conducted from the perspective of the machine owner. At the field level, two systems of using baler technology were generally used: System I and System II. In system I, the machine owner is taking rent in Indian Rupees (₹) from farmers for operating stubble shaver, rake and baler machine on farmer's field for making bales and cleaning the field



by collecting them. After collection machine owner sold the bales to the biomass power generation plant (situated under a radius of 35 kilometers) @ ₹ 1300 per ton. On the other hand, in system II, the machine owner operates only the rake and baler machine at the farmer's field, as the farmers operate the stubble shaver (Figure 2). The farmer retains the bales made by the baler machine and gives the rental charges @ ₹ 6350 per ha to the machine owner. System I involve the twine and transportation cost (Figure 3) @ ₹ 185 per kg and ₹ 450 per tonne, but system II does not involve the transportation cost as the farmers retain the bales. The actual field capacity of the machines was calculated by recording the actual area covered by the machine in the total actual time taken. Fuel consumption was determined by measuring the amount of fuel that was added to the tank both before and after use (Malik et al., 2017).

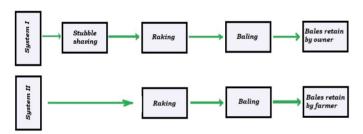


Figure 2: Flow diagram of use of baler technology in system I and II



Figure 3: View of transportation of bales formed by baler

There are many different definitions, each with its pros and cons, of the various economic criteria that must be considered. Therefore, it becomes essential to adopt precise definitions for the various economic terms used and clearly lay down the conventions of calculating them. The three assumptions used for the present study are:

- 1. The tractor is already available with the owner.
- 2. Throughout the project's duration, all of the equipment are paid for with cash and the operating technology is unchanging.
- 3. All inputs, outputs prices and conversion rates remain the same throughout the study period.

The complete cost of baler technology at the field level is comprised (Rahman *et al.*, 2013) of fixed and variable costs. Fixed costs include depreciation, interest on investment, shelter expenses, taxes, insurance and housing costs, among others. Variable costs encompass fuel expenses, lubricants, operator salaries, labour costs, repairs and maintenance.

Fixed Cost (FC)

A *fixed cost* pertains to a resource with a definite quantity that does not vary when the output level does. Since the average yearly cost of the equipment is frequently the only factor to be considered, the straight-line technique, which is the easiest to calculate depreciation, is commonly employed in budgeting (Barnard and Nix, 1980). Consequently, straight-line depreciation is presumed in the computation of fixed cost and the yearly depreciation was determined using the following formula:

$$D = \frac{P - S}{L}$$

where.

D: depreciation, ₹ per yr

P: machine purchase price, $\mathbf{\xi}$

S: machine salvage value, $\mathbf{\xi}$

L: machine life in years, yr.

The fixed cost estimate considers the interest in purchasing a baler technology. Since the investment funds cannot be utilized for other interest-paying businesses, a charge is applied even if they are not



borrowed. There is a 12% interest rate. The investment interest was computed using the following formula:

where, i: interest rate, decimal.

Variable cost (VC)

Variable costs are those that vary with changes in production level. Hourly labour, gasoline, oil, repair and maintenance expenses and hours needed for each field activity affect variable costs. The labour rate of ₹/h was used to compute the cost of labour for the operator. The consumption rate was used to determine the expenses of gasoline and oil, which were subsequently multiplied by the prices for each.

Operating cost (OC)

There were two categories of annual operating costs for the machinery employed in this study: fixed and variable. Following converting the computed fixed and variable costs into ₹/ha (Rs/h), the total of the fixed and variable costs was supplied OC in ₹/ha (₹/h). Following is the computation of the OC:

OC, ₹/ha = Fixed cost +Variable cost

Break Even Point (BEP)

Many farmers want assistance selecting or purchasing all the equipment needed for farming. This is frequently due to a need for more resources, labour, tiny land holdings, or other factors. These farmers get the necessary machinery services for their farms by employing bespoke services. Compare the fixed and variable costs of owning and running the machinery to the overall expenses of bespoke service to determine if owning or engaging a client operator is more cost-effective. The break-even point (Gutierrez and Dalsted, 2020) is found faster and more accurately with the following formula:

where,

F: Annual fixed costs

V: Variable costs per unit of operation

R: Custom hiring charge/rent per unit

Payback period

The time frame whereby revenues can offset the costs of the investment is referred to as the payback. Stated differently, it is the amount of time needed for the cash flow generated by an investment to match the initial outlay made. The desirability of an investment is directly related to its payback period. Shorter paybacks mean more attractive investments (Mohammad *et al.*, 2019). This can be computed by applying the following formula:

Benefit-cost ratio (BCR)

The present value of the benefit stream divided by the present value of the expense stream is known as the benefit-cost ratio. In theory, the benefit-cost analysis approach is straightforward. Equation following provides it and it follows the methodical process of choosing amongst economic investment options (Guttinger, 1994):

A BCR greater than one indicates a profitable investment. To avoid duplicate accounting, depreciation and investment interest are not included in the costs. Including the investment cost accounts for depreciation, while the discount factor accounts for the interest of investment.

Utility index

It is an exact representation of the hours of labour machine interaction. The operation cost and the non-operating hours reduce as the utility index rises. This ultimately leads to a net gain in the overall electricity that may be used for agricultural tasks. One may compute the utility index (K) as follows:

Results and Discussions

For an entrepreneur, custom operator, or progressive farmer, the business of collecting paddy straw using baler technology and providing custom hiring services is a cyclical venture that takes place during the combine harvesting of rice crops. The total cost



of baler technology operations at the farm level comprises variable and fixed costs. Depreciation of the machine was calculated using the straight-line method and taken as a fixed cost. The findings indicated that investing in baler technology proved to be profitable for entrepreneurs. The subsequent section outlines the significant cost and return elements associated with operating a baler technology business in custom hire entrepreneurship:

Economic analysis

The economic analysis of using the baler technology and various assumption made during the analysis for both systems is given in Table 1. The financial analysis was calculated from the perspective of the machine owner, who may be a progressive farmer or a custom hired operator. Based on field data, the baler technology total operating cost for the system I and II were estimated as ₹ 4285.74 per hour and ₹ 2374.34 per hour. The actual field capacity of the baler, rake and stubble shaver machine was 0.53, 0.79 and 0.51 ha per hour having fuel consumption of 6.6, 4.8 and 4.25 per hour respectively. Fixed and variable cost for the machine operation were calculated as ₹ 629.2 and 3656.54 per hour for system I and 609.4 and 1764.94 per hour for system II based on the average field data collected through personal interviews of custom-hire service providers. The amount of twine used was 6.35 kg per ha costing ₹ 622.62 per hour. In case of system I, the transportation cost including loading the bales on the trolley and transport to the buying point (biomass power plant) was 1514.48 ₹ per hour. In operating the baler machine for system I, the total per hour operating cost was estimated as ₹ 3313.39, which includes a total fixed cost of ₹ 484, total variable cost of ₹ 866.54 and twine cost of ₹ 622.62 and transportation cost of ₹ 1514.48 respectively. This showed that the bale transportation cost (from the collection point to the selling point) was the major contributor (45.70%) in the baling

system I, followed by the variable, twine and fixed costs. In system II, however, the bale transportation cost was not included as the farmers kept the bales. The total per-hour operating cost for operating the baler machine in system II was ₹ 1798.91, which includes the maximum contribution (38.48%) from variable cost followed by twine and fixed cost. The total per hour operating cost was computed as ₹ 575.43 for both the system and the major contributor of cost (78.21%) in operating the rake machine was variable cost followed by fixed cost. The total perhour operating cost of the stubble shaver machine (for system I only) was ₹ 396.93 and the significant contribution (95.01%) was from variable cost followed by the fixed cost. The various itemized cost per hour for both systems were analysed and presented (Figures 4 and 5).

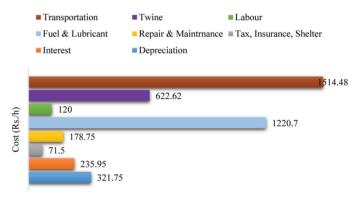


Figure. 4: Itemized cost per hour of operation for system I

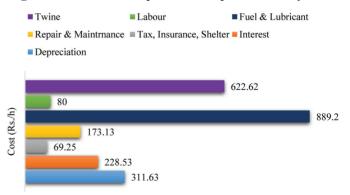


Figure 5: Itemized cost per hour of operation for system II



Table 1 Economic analysis of system I and II

System I: Operator retaining	ng the bales and sellin	g to power	plants	System II: Fa	rmer retainir	g the bales
Particulars	Assumption	Baler	Rake	Stubble shaver	Baler	Rake
Purchase price, ₹	P	1100000	285000	450000	1100000	285000
Salvage (S), ₹	10% of P	110000	28500	4500	110000	28500
Life, yr	-	10	10	10	10	10
Economic life, h	-	400	400	400	400	400
Depreciation, ₹/yr	-	99000	25650	4050	99000	25650
Interest cost, ₹/yr	Rate of interest, 12%	72600	18810	2970	72600	18810
Tax, insurance, shelter, ₹/yr	2% of P	22000	5700	900	22000	5700
Fixed Cost, ₹/yr (₹/h)	-	193600	50160	7920	193600	50160
Total fixed cost (FC), ₹/yr (₹/h)	-		251680 (62	29.2)	243760	(609.4)
Repair & maintenance, ₹/h	5% of P/Avg/yr	137.5	35.63	5.63	137.5	35.63
Fuel cost, ₹/h	@ ₹ 87/litre	574.2	399.18	369.75	574.2	399.18
Lubrication cost, ₹/h	20% of Fuel Cost	114.84	79.836	73.95	114.84	79.836
Labour Cost, ₹/h	-	40	40	40	40	40
Variable cost (VC), ₹/h	-	866.54	554.646	489.33	866.54	554.646
Twine cost, ₹/h	₹ 185/kg	622.62	-	-	622.62	-
(Twine used						
6.35 kg/ha)						
Transportation cost, ₹/h	₹ 4.5/ton	1514.48	-	-	-	-
Operating cost (OC), (₹/h)		3313.39	575.43	396.93	1798.91	575.43
Total operating cost, ₹/h	-		4285.7	4	2374	1.34

Economic worth assessment

The details of all economic parameters for assessing the economic worth of implementing baler technology were presented (**Table 2**). The rental charges for using baler technology by a customer operator is ₹ 1346.2 per hour for the system I and 3365.5 per hour for system II. The net benefit comes out to be ₹ 998.09 and 991.16 per hour (**Table 2**) for the system I and II. Considering 150 hours of annual use of baler technology according to the custom operators the net annual benefit or revenue was observed to be ₹ 399237 and 396463 for the system I and II.

After a detailed economic analysis of systems I and II, it has been observed that the net benefits per ha were ₹ 1883.19 and 1870.11, which showed that the net benefit was almost at par for both systems. Therefore, both systems followed by the machine owner or baling service provider gave at-par financial benefits. Therefore, if a new entrepreneur or progressive

farmer wants to start a new venture of collecting and selling paddy straw, they can choose systems I and II. Considering the capital cost involved in the purchase of baler technology for both the systems and the assumption made for economic analysis in this study, the break-even point comes out to be 81.97 ha or 154.66 ha per year and 80.72 ha 152.23 hours per year for the system I and System II (Figures 6 and 7). In contrast, the actual use of baler technology is approximately 400 hours per year by the custom operator. Considering the actual annual use of baler technology, the custom operator or progressive farmer shall be able to recover his cost in 3.58 and 3.49 years (payback period). Mangaraj and Kulkarni (2011) have reported a payback period of 5 years while studying the techno-economic perspectives of baler machines. Both systems are profitable from the viewpoint of custom operators. The payback period of both the systems is



less than the life (10 years) of the baler technology machinery and also, the benefit-cost ratio for both systems I and II are found to be 1.23 and 1.42, which is greater (>) than unity. Pal et al., (2019) reported a benefit-cost ratio 1.39 for collecting paddy straw with a baler machine after harvesting paddy crops with a combine harvester. So implementing baler technology for managing paddy straw by its collection, is an acceptable venture from the business point of view. The utility indexes obtained were 2.58 and 3.64 for system I and II which are greater than unity. This indicates efficient utilization of baler technology.

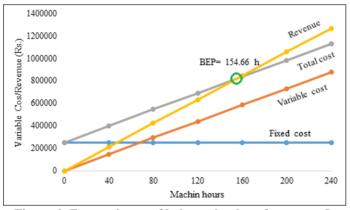


Figure 6: Economic use of baler technology for system I

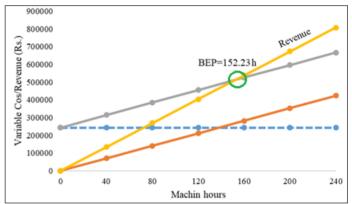


Figure 7: Economic use of baler technology for system II

Table 2: Baler technology economic worth assessment

System I: Operator retai	ning the bales ar	nd selling to power plants	System II: Fa	rmer retaining the bales
Items	Value	Remarks	Value	Remarks
Actual field capacity, ha/h	0.53	Field capacity of straw baler after operation of rake	0.53	Field capacity of straw baler after operation of rake
Rental revenue, ₹/ha (₹/h)	2540 (1346.2)	Rent for cleaning the field by making and collecting bales	6350 (3365.5)	Rent of making bales only
Straw collected, ton/ha	6.32	Average amount of straw collected from one ha	-	-
Revenue form straw sale, ₹/ha (₹/h)	8255 (4375.15)	Sold at the rate of ₹ 130 per quintal	-	-
Revenue after moisture cut, ₹/ha (₹/h)	7429.5 (3937.64)	10% moisture cut generally applied by buyer	-	-
Total revenue, ₹/ha (₹/h)	9969.5 (5283.84)	-	6350 (3365.5)	-
Net Benefit, ₹/ha (₹/h)	1883.19 (998.09)	-	1870.11 (991.16)	-
Annual Benefit (₹)	399237	-	396463	-
Actual Payback period (yr)	3.58	Less than economic life of machine (<6 yr)	3.49	Less than life of machine (<10 yr)
Break-even point ha/yr (h/yr))	81.97 (154.66)	More than the actual covered area	80.72 (152.23)	Less than the actual covered area
Benefit-cost ratio (BCR)	1.23	Greater than unity (>1)	1.42	Greater than unity (>1)
Utility index	2.58	Greater than unity (>1)	3.64	Greater than unity (>1)



It was observed that, although both the system of implementing baler technology is acceptable, system II is a little bit more beneficial as it is having benefit-tocost ratio and less payback period than system I. This is due to the absence of cost involved in the operation of the stubble shaver machine and the absence of capital cost required to purchase the stubble shaver machine. In system II, farmers retain the bales, which they can sell to some other buyer at a higher price to get additional benefits or can use them for another purpose, such as the production of bioenergy and use in the packaging as a mushroom growth medium and paper industry, as natural manure (worm-compost), etc. Implementing baler technology also generates employment for the rural youth by engaging the youth in loading and unloading, transporting the bales, etc.

Limitation in Implementing Baler Technology

This study's budgeting process only permits the consideration of variables that can be measured and for which accurate estimates are available. Therefore, when employing the baler technology, the potential restrictions of the situational elements are not considered. The profit or revenue generated from using baler technology largely depends upon the sale price of the straw. If the sale price of paddy straw goes down or machine operators do not get the appropriate price, then the implementation of baler technology is considerably affected. There are also hidden costs, such as twine, labour and bales transportation costs, including using baler technology. The significant variation in these hidden costs may affect the implementation of the baler technology. The number of machinery and their high initial cost can also be hindrances in choosing the baler technology. The sole thing contributing to rising expenses per unit is the tardiness of field operations. On the other hand, management is responsible for operations scheduling. Only until a predetermined pattern of operations including the dates and orders of each field operation is defined can the budgeting approach be applied. Any disruption to this pattern results in untimeliness losses. But managing each circumstance to maximize gains or prevent losses is the manager's job. Depending on

the situation, an observant owner or management may alter this operation pattern. The output from the baler technology also depends upon the skill of the machine operator. While operating the straw collection machinery, an unskilled operator can cause damage to the machinery, thereby halting the collection process and ultimately resulting in revenue loss. Biomass power plant operators apply 10% of the moisture cut if the bales come to them, which is moister than the desired level, which further reduces the income from selling the straw bales. So, keeping the moisture level of paddy straw within the limit while selling to the buyer is another challenge to the machine operator/owner to avoid an unnecessary reduction in revenue.

Therefore, it is worth mentioning that before implementing baler technology, a machine owner/ progressive farmer/custom operator must ensure where they will sell the bales or their end-use. This exercise must be done before adopting the baler technology. Further, the help of government aid, such as providing subsidies on the machinery involved in the baler technology or developing more power plants or other practical industries which can utilize the paddy straw, can enhance the decision to use baler technology by the progressive farmer or service provider. Machine custom hiring centres or societies can be developed to enhance the availability of machinery to the individual or end user. The grouping of farmers can be done so that they can buy the costly baler technology machinery collectively and can implement this technology at the village or block level to earn and avoid straw burning issues. This will enhance the economic condition of farmers/entrepreneurs/service providers along with the avoidance of the infield burning of paddy straw problems, which will safeguard the overall ecosystem.

Conclusion

According to the study, implementing baler technology for paddy straws management by straws collection is profitable. The additional benefit is that paddy straw burning (on-farm) can be avoided, which is beneficial and much needed for the environment, agriculture and living beings. The economic worth assessment showed



that the total operating cost and net benefit for systems I and II were estimated as ₹ 4285.74, 2374.34, 998.09 and 991.16 per hour for systems I and II. The benefit-cost ratio of systems I and II of baler technology was found to be 1.23 and 1.42 (greater than unity), which makes it a profitable and acceptable venture for an entrepreneur. The break-even usage of the baler technology was appraised at 154.66 and 152.23 hours per year of machine operation for systems I and II. The utility indexes obtained in system I and II were 2.58 and 3.64 which are more than unity.

The economic indicators determined in the present study indicates the efficient and beneficial baler technology venture. The owners of baler technology or progressive farmers/service providers can embrace this profitable venture and get monetary benefits while contributing to the reduction of paddy straw burning and the associated environmental pollution.

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RESEARCH ARTICLE

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Status of Bacterial Leaf Blight in Major Rice Growing Districts of Gujarat

Ajay Chaudhary^{1*}, Gangwar RK² and Thorat SS²

¹Department of Plant Pathology, BACA, AAU, Anand (Gujarat) 388 110

²Main Rice Research Station, AAU, Nawagam, Kheda (Gujarat) 387 540

*Corresponding author Email: ajchaudhary02@gmail.com

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Abstract

Rice (*Oryza sativa* L.), a staple food for over half of the global population, plays a crucial role in meeting nutritional needs, particularly in Asia, often referred to as the world's "Rice Bowl." Bacterial leaf blight (BLB) caused by *Xanthomonas oryzae* pv. *oryzae* poses a significant threat to rice production. In response to the destructive nature of the pathogen, a survey was conducted during the *kharif* 2023 in Ahmedabad, Tapi, Anand, Kheda and Navsari districts of Gujarat. The survey aimed to assess the incidence of bacterial leaf blight, providing crucial insights into the current status of the disease. Among all the surveyed districts highest per centage incidence (49.54%) of bacterial leaf blight was observed in Kheda followed by Tapi (48.73%), Ahmedabad (46.53%), Navsari (12.74%) and the lowest incidence per centage was recorded in Anand (22.57%).

Keywords: Bacterial leaf blight, rice, survey, Xanthomonas oryzae pv. oryzae

Introduction

Rice (Oryza sativa L.) belonging to the Poaceae family, is the world's most widely grown food crop (Ezuka and Kaku, 2000). In terms of its nutritional content, rice is becoming of paramount importance to more than half of the world's population. Asia is considered as the world's "Rice Bowl". It is a nourishing food because it has 6.89 g of protein, 78.2 g of carbohydrates, 0.5 g of fat, 0.2 g of crude fibre, 0.6 g of mineral matter, 10 mg of calcium and 160 mg of phosphorus per 100 g (Mangalarai and Mauria, 1999). In tropical and subtropical rural and urban settings, people get between 40 to 70 per cent of their calories from rice (Hossain and Fischer, 1995). The cultivation of rice is a significant source of income and employment in the rural areas of Asian countries (Hossain, 1997). Rice is a tropical plant and cultivated in hot and humid climate. It is primarily grown in the regions which are rainfed and get good rain every year during kharif season. In India, rice occupies an area of 46.5 m ha with the production of 130.84 million tonnes and productivity of 2809 kg per ha (Anonymous, 2023). Whereas in Gujarat rice crop is cultivated on 0.89 million hectares with the production of 2.10 million tonnes and productivity of 2356 kg per ha (Anonymous, 2023). Rice is mainly grown in central and southern districts of Gujarat, which include Valsad, Dang, Kheda, Ahmedabad, Anand, Mahisagar, Panchmahal, Vadodara, Tapi and Navsari. The crop is cultivated in both irrigated as well as rainfed conditions during *Kharif* as well as summer season in Gujarat state. The productivity of rice in Gujarat is quite low due to several abiotic and biotic factors, which includes bad weather, diseases and insect-pests infestation.



Among the diseases bacterial leaf blight (BLB) or bacterial blight (BB) caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*) is a serious problem and threat to rice production in both tropical and temperate rice growing regions due to its high epidemic potential (Mew,1987). The disease broke out in the epiphytotic form in Shahabad district of Bihar (Srivastava and Rao, 1966). The disease has become endemic on rice due to intensive and repeated cultivation in many Asian countries, (Mew *et al.*, 1993). BLB has spread to many non-traditional areas in India, in addition to recurring incidence in the traditional areas under irrigated and rainfed shallow lands. The pathogen is highly variable in nature (Kumar *et al.*, 2016).

The disease causes up to 20 per cent of the yield losses when appear at the tillering stage (Thind and Bala 2002; Liu et al., 2006). The disease has the potential to cause 10-12 per cent yield losses under mild infection (Mew et al., 1993). Under severe conditions, bacterial blight of rice can cause significant yield losses. In Japan, yield losses have ranged from 25-35%, sometimes reaching up to 60% (Ou, 1985). In the Philippines and Indonesia, losses were also very high, with the Philippines recording losses of 24.50% in the moist season and 7.21% in the dry season for vulnerable crops (Exconde, 1973). In India and Bangladesh, heavy yield losses of 12-32% have been reported (Shah Jahan, 1992). Similarly, in East India, yield losses have ranged from 7- 62% and even up to 82% for the same genetic resources (Srivastava et al., 1967; Singh et al., 1980; Srivastava and Kapoor, 1982).

Laha et al., (2016) reported in production-oriented survey 9-52% intensity of bacterial leaf blight during 2001 to 2014 in different rice growing districts of Gujarat. Laha et al., (2020) reported that a production-oriented survey conducted in 2020 found the incidence of bacterial leaf blight to be low to moderate in different rice growing districts of Gujarat. The disease occurs

at the seedling, vegetative and reproductive stages but at the tillering stage it causes severe blighting of leaves resulting in yield loss. The pathogen, is seedborne, residing in glumes and occasionally within the endosperm. Seeds collected from heavily diseased fields carry the pathogen and seedlings grown from such seeds typically exhibit disease symptoms and succumb at an early stage (Srivastava and Rao, 1966). The ability of the bacteria to persist in seeds contributes to the perpetuation and dissemination of the disease, further emphasizing the need for effective management strategies to safeguard rice crops from this destructive threat (Mondal *et al.*, 2019).

The most effective approach to control these this disease is using resistant varieties. Development of disease resistant rice is one of the most important achievements rice breeders attempt to accomplish (Tejaswini *et al.*, 2016).

In view of the destructive nature of the pathogen, a roving survey on the incidence of bacterial leaf blight was carried out during *Kharif*, 2023 in major ricegrowing districts of Gujarat.

Symptomatology

Bacterial leaf blight is a typical vascular disease that can be divided in three distinct phases of symptoms i) Leaf blight phase, ii) Kresek phase and iii) Pale yellow leaf phase. The leaf blight phase is characterized by water-soaked lesions starting at the leaf tips, which then extend downward. Initially, these lesions are pale green but later turn yellow to straw-colored with wavy margins. Water soaking can be observed where the lesions meet healthy tissue. Lesions can begin at one or both edges of the leaves and sometimes appear along the midrib or elsewhere on the leaf blade. As the disease progresses, the lesions can cover the entire leaf blade, turning white and eventually gray or black due to saprophytic fungi growth. In humid areas, yellowish, turbid drops of bacterial ooze may be seen on young lesions in the early morning, which dry into



small, yellowish beads. The most destructive phase, known as the 'kresek' or wilt phase, occurs in the tropics due to early systemic infection in the nursery or from seed infection. This phase causes leaves to roll, droop, turn yellow or gray and ultimately wither. In severe cases, the entire affected plant may die. And the last is pale yellow leaf phase, which was reported from the Philippines, this phase causes some of the youngest leaves in a clump to turn pale yellow or whitish. These diseased leaves then wither, turn yellowish-brown and dry up. This phase has not been reported in other countries (Laha *et al.*, 2009).

Materials and Methods

The roving survey was conducted during the *kharif* season of 2023 to estimate the incidence of bacterial leaf blight in rice in five major rice growing districts of Gujarat *viz.*, Anand, Kheda, Navsari, Ahmedabad and Tapi. To conduct this survey two talukas were selected from each district and two villages were chosen from each taluka, whereas total five fields were selected randomly from each village to ensure a broad and representative sample of the rice crops in these regions. The survey was conducted between panicle initiation to booting stage of the crop.

During the field surveys, positive sampling was conducted and infected leaves of rice plants exhibiting typical bacterial blight symptoms (Figure 1) were collected.



Figure 1: Bacterial leaf blight infected plants

For accurately record observations of the Per centage Disease Index (PDI) and Disease Incidence (DI) for (BLB) of rice, several key parameters were recorded.

For calculating the actual incidence of disease in a field, plants were observed at ten points along a diagonal transect (IRRI, 1996). Five quadrates were randomly selected from each field and ten plants were then observed for the disease symptoms.

Number of bacterial blight
$$DI (\%) = \frac{\text{infected plants}}{\text{Total number of plants examined}} \times 100$$

Total number of plants examined within each quadrate, as well as the number of plants showing symptoms of BLB were counted and recorded. The per cent disease index (PDI) was assessed by evaluating the severity of the disease on individual plants, using a standardized scale (e.g., 1-9, where 1 indicates mild symptom and 9 indicates severe disease).

The PDI is calculated with the formula:

Observations recorded

Per centage disease index (PDI)

Disease incidence (DI)

Results and Discussions

A roving survey was conducted during September-October of *kharif* 2023 in major rice growing districts of Gujarat *i.e.*, Anand, Ahmedabad, Tapi, Kheda and Navsari which are known for their rice cultivation to assess the incidence of bacterial leaf blight of rice.

The data in **Table 1** revealed that the highest incidence (49.54%) of bacterial leaf blight was observed in Kheda followed by Tapi (48.73%), Ahmedabad (46.53%), Navsari (12.74%) and the lowest incidence per centage was recorded in Anand (10.56%).



Table 1: Per cent disease incidence and per cent disease index of bacterial leaf blight of rice in different rice growing districts of Gujarat

Sl.				Don	cent Inci	danaa	Per cen	t Disease	GPS	Cultivar/
No.	District	Taluka	Village	Per	cent inci	uence	in	dex		
NO.				Village	Taluka	District	Village	District	coordinates	Varieties
1.	Kheda	Kheda	Nawagam	87.30	84.4		47.30		22.797505°	GR11, Surya
									72.573497°	Moti,
2.			Lali	81.50			38.50		22.861302°	Punjab S, Moti
						49.54		31.31	72.633431°	Gold
3.		Matar	Radhvanaj	13.17			20.17		22.70970°	GAR13
					14.69				72.72390°	
4.			Ratanpur	16.21			19.27		22.73660°	GAR13
									72.71477°	
5.	Ahmed-	Daskroi	Devdi	71.92			35.92		22.901808°	GR11, Punjab S
	abad				74.65				72.660803°	
6.			Nandej	77.38			37.55		22.908878°	Moti Gold, GR11,
						46.53		28.56	72.673322°	Gurjari
7.		Bavla	Kathwada	29.50			22.45		23.80799°	GAR 13, Moti
					18.41			_	72.55299°	Gold
8.			Bavla	7.33			18.33		22.842144°	Surya Moti,
									72.363074°	GAR13
9.	Anand	Anand	Lambhvel	21.45			22.50		22.577175°	GR21, Sonam,
					15.06				72.924656°	Moti gold
10.			Chikhodra	8.67			16.67		22.559579°	Masuri, Sonam
						10.56		17.50	73.00783°	
11.		Petlad	Morad	6.97			19.97		22.543975°	Moti Gold,
					6.07			_	72.873618	GAR13
12.			Ravipura	5.18			11.18		22.549936°	Sriram125,
									72.847512°	GAR13,
					0.1.10	10.70		25.11	-10-011	Mahisagar
13.	Tapi	Vyara	Magarkui	79.31	81.43	48.73	39.31	26.44	21.070414°	Versha, US-312
			D 11	00.55			40.20		73.394043°	TTG 010 150 10
14.			Panvadi	83.55			40.38		21.094584°	US-312, MC-13,
1.5		X 7 1 1	D 11 1	15.55	1602		10.55	_	73.400135°	Annapurna
15.		Valod	Borakhadi	15.55	16.03		12.55		21.094584°	US 2171, Masuri,
1.0			D - ::	16.50			12.50	_	73.400135°	GAR13
16.			Bajipura	16.52			13.50		21.10013°	Bayer 6444, GAR
17	NI	C1. '1-1.1'	C1. 11-1.11	0.10	10.17	12.74	11 10	16.62	73.291831°	13, US-312
17.	Navsari	Chikhli	Chikhli	9.18	18.17	12.74	11.18	16.63	20.76388°	GNR7, GAR13
10			Dathrani	27.17			20.70	-	73.06012°	Noth Down MC
18.			Rethvania	27.17			30.70		20.81083° 73.12391°	Nath Pawan, MC-13, US 312
19.		Navsari	Dhantej	7.13	7.32		14.13	-	20.92247°	US 2111, Gurjari
19.		mavsari	Duamej	/.13	1.32		14.13			OS 2111, Gurjari
20.			Sisodra	5.51			10.51	-	72.93124°	HC 2111 CD15
∠0.			Sisoura	3.31			10.31		20.93445°	US 2111, GR15,
									72.98385	GAR13

The incidence of bacterial blight varied across talukas in different districts. In Kheda district, the highest incidence was observed in Kheda taluka (84.4%), followed by Matar taluka (14.69%). In Tapi district,

Vyra taluka recorded 81.43%, while Valod taluka had 16.03%. Ahmadabad district reported 74.65% in Daskroi taluka and 18.41% in Bavla taluka. In Navsari district, the incidence rates were 7.32% in



Navsari taluka, 18.17% in Chikhli taluka and 16.03% in Valod taluka. Meanwhile, Anand district showed 15.06% in Anand taluka and 6.07% in Petlad taluka, reflecting considerable variation in disease incidence across these regions.

The data on village wise incidence of bacterial leaf blight showed that the highest incidence (87.30%) was recorded in Nawagam of Kheda district followed by Panvadi (83.55%) of Tapi district, Lali (81.50%) of Kheda district, Mangarkui (79.31%) of Tapi district, Nandej (77.38%), Devdi (71.92) of Ahmedabad district and Kathwada (29.50%) of Ahmedabad district, Rethvania (27.17%) of Navsari district, Lambhvel (21.45%) of Anand district, Bajipura (16.52%) of Tapi district, Ratanpur (16.21%) of Kheda district, Borkhadi (15.55%) of Tapi district, Radvanaj (13.17%) of Kheda district, Chikhli (9.18%) of Navsari district, Chikhodra (8.67%) of Anand district, Bavla (7.33%) of Ahmedabad district, Dhantej (7.13%) of Navsari district, Morad (6.97%) of Anand district and Sisodra (5.51%) of Navsari district whereas the village with the lowest incidence was Ravipura of Anand district with a disease incidence of (5.18%) (Table 1).

As for the Per cent Disease Index (PDI) the highest PDI (31.31%) of bacterial leaf blight was recorded in Kheda followed by Ahmedabad (28.56%), Tapi (26.44%), Anand (17.50%) and the lowest PDI was recorded in Navsari (16.63%).

The PDI of BLB was also recorded during the survey in major rice growing districts of Gujarat. The highest PDI was observed in village Nawagam of Kheda district with a PDI of 47.30%, followed by Panvadi in Tapi district with 40.38% and Magarkui in Tapi district with 39.31%. Nandej and Devdi in Ahmedabad district also exhibited a high PDI (37.55 and 35.92%, respectively). Other significant observations in PDI were from Lali (38.50%) in Kheda district and Lambhvel (22.50%) in Anand district. Kathwada in Ahmedabad showed a PDI of 22.45% and Ratanpur in Kheda had a PDI of 19.27%. Whereas in Radhvanaj village of Kheda, the PDI was 20.17% and in Bavla

of Ahmedabad it was 18.33%, showed moderate PDI. While the lower PDI was recorded from the village Chikhodra in Anand (16.67%), Borakhadi in Tapi (12.55%) and Morad in Anand (19.97%). The minimum PDI was recorded from the Ravipura village (11.18%) and Chikli village (11.18%) while the lowest PDI was recorded in village Sisodra (10.51%) of Navsari district. This comprehensive data highlights the varying severity of BLB across different regions and varieties in Gujarat.

The result found were in accordance to that recorded by Laha et al., (2016) who reported during productionoriented survey 09-52 per cent incidence of bacterial leaf blight during 2001 to 2014 in different rice growing districts of Gujarat. Laha et al., (2020) also reported that during production-oriented survey conducted of kharif 2020 that the incidence of bacterial leaf blight to be low to moderate in different rice growing districts of Gujarat. The results of this study align with the findings of Thimmegowda (2006), who observed during the kharif season of 2005 that Raichur had the highest disease incidence rate of 61.7 per cent, while Siruguppa had the lowest incidence 46.78 per cent. In the summer of 2006, the highest disease incidence was recorded in Sindhanur at 74.69 per cent, with Siruguppa again having the lowest at 46.78 per cent.

Our present study identified the hotspots of bacterial leaf blight (BLB) of rice in different ecosystems of Gujarat. This disease, caused by a dynamic pathogen that rapidly evolves new races, can be effectively managed through several strategies.

The widespread presence of bacterial leaf blight (BLB) across various regions of Gujarat suggests the critical need for improved management practices. In particular, high incidences in specific areas point to potential hot spots where targeted interventions could be most effective. The variability in disease incidence across different districts, talukas and villages suggests that local environmental conditions, agricultural practices and the choice of rice cultivars play significant roles in disease dynamics.



High incidence rates in districts such as Kheda (49.54%), Tapi (48.73%) and Ahmedabad (46.53%) indicate the urgent need for focused disease management strategies in these regions. Similarly, talukas with exceptionally high incidences, such as Kheda (84.4%), Vyara (81.43%) and Daskroi (74.65%), represent critical areas where targeted control measures could substantially reduce the disease burden. Village-level data, revealed the highest incidence in Nawagam (87.30%) and Panvadi (83.55%), further emphasize the importance of localized interventions.

Several factors contribute to the observed variability in BLB incidence, including climatic conditions, soil health, water management and the susceptibility of rice cultivars. For instance, areas with high humidity and frequent rainfall provide conducive environments for the spread of BLB. Poor soil health and improper water management practices can exacerbate disease conditions. Additionally, the use of susceptible rice cultivars in certain regions can lead to higher disease incidences. Research by Mew *et al.* (1993) and Gnanamanickam *et al.* (1999) highlights that high-yielding varieties and excessive nitrogen use are key factors in bacterial blight outbreaks.

To mitigate the impact of BLB and improve rice yields in Gujarat, region-specific integrated disease management strategies are essential. These strategies should combine both chemical and cultural practices. Chemical management could include the use of effective bactericides and antibiotics, while cultural practices might involve crop rotation, proper water management and the use of resistant rice varieties.

Studies have shown that integrated disease management, which combines various control methods, is more effective in managing BLB than relying on a single approach. For instance, Krishnakumar and Kumaravadivel (2018) reported that the incidence of BLB could be reduced through the adoption of integrated management practices that include timely application of bactericides, use

of resistant cultivars and implementation of good agricultural practices.

Conclusion

Bacterial leaf blight (BLB) is a major issue in Gujarat's traditional rice-growing areas, with disease incidence ranging from 10.56% to 49.54% and PDI from 16.63% to 31.31%. It is severe in moderate rainfall areas with high humidity, driven by factors like strong winds, rain splashes and heavy monsoon rains. The widespread cultivation of susceptible varieties such as GR11 and Moti Gold, monocropping and excessive nitrogen fertilization have further increased its prevalence.

The significant variability in BLB incidence across Gujarat highlights the need for targeted, region-specific management strategies. By addressing the unique conditions and practices in each region, it is possible to effectively control BLB, thereby safeguarding rice production and enhancing food security in the state. Effective control can be achieved through proper cultural practices, resistant cultivars and timely antibiotic applications. Identifying high-risk areas is essential for implementing targeted interventions to reduce disease impact and improve yield.

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RESEARCH ARTICLE

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Evaluation of Donor and Advanced Lines for Blast Resistance Across Agroclimatic Zones of Andhra Pradesh and Telangana

Eden Georgia Karedi^{1*}, Srinivas Prasad M², Jesudas GS², Bhuvaneswari V³, Madhusudhan P⁴, Udayababu P⁴, NVSR Ravi Kumar B⁵ and Manoj Kumar V⁶

¹Agricultural College, ANGRAU, Bapatla, AP,

²Indian Institute of Rice Research, Rajendranagar, Telangana,

³Regional Agricultural Research Station, ANGRAU, Maruteru, AP,

⁴ Agricultural Research Station, ANGRAU, Nellore, AP,

⁵Regional Agricultural Research Station, ANGRAU, Nandyal, AP,

⁶Regional Agricultural Research Station, ANGRAU, Guntur, AP

*Corresponding author Email: karedi.eden@gmail.com

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Abstract

Rice blast, caused by the fungal pathogen *Pyricularia oryzae*, is a major threat to rice production worldwide. As *P. oryzae* is a highly evolving and dynamic pathogen and adapting host plant resistance is the most significant way of managing the blast disease. The present study aims to assess the performance of rice genotypes across different locations to understand their adaptability and also to monitor the virulence patterns of the blast pathogen population. A multi-location trail was carried out with 39 cultivars consisting of near-isogenic lines, international differentials, donors and commercial cultivars possessing different genes for blast resistance by adopting Uniform Blast Nursery (UBN) at six locations in different agroclimatic zones of Andhra Pradesh and Telangana during *Kharif* 2023. There was significant variation in disease pressure among locations, with ARS, Nellore exhibiting the highest Location Severity Index (LSI) of 4.44 and ARS, Uttukur showing the lowest LSI of 2.76. The susceptibility Index (SI) 7.45 was recorded highest in the susceptible check (HR-12) while the resistant check Tetep showed the lowest SI of 2.16. Several genotypes carrying the *Pi-1*, *Pi-12*, *Pi-z+Pi-a+Pi-i*, *Pi-54*, *Pi-9* and *Pi-kh*+ genes demonstrated consistent resistance across different locations. The study highlights the importance of considering gene combinations for durable resistance and the need for site-specific adaptation in rice breeding programs to combat rice blast effectively.

Keywords: Agroclimatic zones, Blast disease, Differential lines, Rice, Telangana

Introduction

Rice is one of the most significant edible cereal grains. Rice is a rich source of carbohydrates, which is essential for providing energy. It also contains vitamins and minerals, contributing to a balanced diet and it serves as a primary staple food for more than two-thirds of the world's population (Fukagawa and

Ziska, 2019). Although rice output has expanded to meet global demand, productivity has not increased appreciably. One of the major constraints faced by the rice growers for low productivity is biotic stress. Among the biotic factors, diseases remain a persistent challenge in rice cultivation. The rice crop suffers



from diseases caused by fungi, bacteria, viruses, phytoplasma, nematodes and other non-parasitic disorders. Among the fungal diseases blast disease is caused by Pyricularia oryzae Cavara which is a significant threat to rice production worldwide (Anand Kumar et al., 2023). It can cause severe loss in yield to the extent of 70-80% in various rice ecosystems (Aruna et al., 2015). P. orvzae is constantly evolving and is regarded as a threat to global food security. Management of blast disease is carried out using various strategies, including crop rotation, fungicide application and planting of resistant varieties. Among the various methods for managing plant diseases, genetic resistance is widely regarded as the most effective and sustainable, providing both economic and environmental benefits (Bonman, 1992). Rice blast resistance is a classic gene-for-gene interaction, where a major resistance gene in the host plant effectively counters P. oryzae strains carrying the corresponding avirulence gene (Silue et al., 1992). Developing rice cultivars with durable resistance to blast is crucial for ensuring food security. This study aimed to evaluate the resistance of various rice genotypes containing of near-isogenic lines, international differentials, donors and commercial cultivars possessing different genes for blast resistance under different agro-climatic conditions in Andhra Pradesh and Telangana, India.

Materials and Methods

The present study was carried out to evaluate blast disease in different agro-climatic regions of Andhra Pradesh and Telangana during *kharif* 2023. These lines were evaluated in Uniform blast nurseries at six different locations *viz.*, IIRR Hyderabad, RARS Maruteru (A.P.), ARS Nellore (A.P.), ARS Ragolu (A.P.), RARS Nandyal (A.P.) and ARS Uttukur (A.P.). The details of the locations were presented in the **Table 1**. Thirty-nine cultivars consisting of near-isogenic lines, international differentials, donors and

commercial cultivars possessing different genes for blast resistance including appropriate susceptible (HR-12, Co-39) and resistance (Tetep, Rasi and IR 64) checks were evaluated for blast resistance. The fungus was isolated by tissue segmentation method (Bonman et al., 1987). Single spores were located and picked up microscopically and transferred to fresh sterilized Petri plates containing Oat Meal Agar (OMA) medium. The Petri plates were incubated at 28 °C for 7 days and the fungus was identified following mycological description (Ou, 1985). After 14 days of incubation at 28 °C, petri plates (90 mm) of P. oryzae isolate was washed with 20 ml of sterile distilled water to produce spore suspension. The concentration of the conidial suspension was adjusted to 1×10⁵ conidia ml⁻¹ using a haemocytometer.

Uniform Blast Nursery (UBN) was a 10x1 m bed. The soil was pretreated with FYM, NPK and commercial sulphuric acid before tilling to distribute the manure uniformly in the soil. Highly susceptible variety (HR 12) was sown as a border row on either side of the bed and between the test material rows. After every 10 rows of test materials HR 12 was planted which acts as spreader rows. Test material was sown in 50 cm rows perpendicular to the border row with 10 cm spacing between the rows (**Figure 1**).

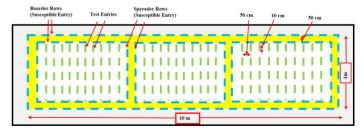


Figure 1: Layout of Uniform Blast Nursery

25 days after sowing these nursery beds were sprayed with spore suspension of local blast isolate using a hand-operated atomizer. Relative humidity was maintained by sprinkling water as mist with



30 min time interval depending on the intensity of temperature. The UBNs were covered with polythene sheets during night to maintain the humidity to build up more spore load and in turn to increase the disease pressure on the varieties. Scoring was done after 10-15 days of post infection depending on the severity of the infection on the susceptible check using SES

scale (IRRI, 1996) (**Table 2**). Scoring was done after 10-15 days of inoculation depending on the severity of the infection on the susceptible check using SES scale (IRRI, 1996). The data thus obtained from field experiments in a Randomized Block Design were analyzed statistically by Two-way ANOVA (Gomez and Gomez, 1984).

Table 1: Details of different locations of Andhra Pradesh and Telangana

Agro climatic zone	Location	Research Station/ Institute	State	Latitude	Longitude	Ecosystem
Godavari Zone	Maruteru	Regional Agricultural Research Station (RARS)	Andhra Pradesh	16.6299° N	81.7457° E	Irrigated, Lowland
Southern Zone	Nellore	Agricultural Research Station (ARS)	Andhra Pradesh	14.4303° N	79.9987° E	Irrigated, Lowland
North coastal Zone	Ragolu	Agricultural Research Station (ARS)	Andhra Pradesh	18.16412°N	83.5010° E	Irrigated, Lowland
Scarce rainfall Zone	Nandyal	Regional Agricultural Research Station (RARS)	Andhra Pradesh	15.4786° N	78.4831° E	Rainfed, Upland
Southern Zone	Uttukur	Agricultural Research Station (ARS)	Andhra Pradesh	14.4373° N	78.8050° E	Rainfed, Upland
Southern Telangana Zone	Hyderabad	Indian Institute of Rice Research (ICAR-IIRR)	Telangana	17.3871° N	78.4916° E	Irrigated, Lowland

Results and Discussions

The results revealed that the intensity of rice blast disease incidence varied at different test locations. Among the locations, the Location Severity Index (LSI) was recorded high at ARS, Nellore (4.44) located in the Southern region of the agroclimatic zone of Andhra Pradesh followed by RARS, Maruteru (3.70) located in the Godavari zone and the least disease incidence was recorded at ARS, Uttukur (2.76) located in the Southern region of the agroclimatic zone of Andhra Pradesh (**Figure 2**). HR-12 showed susceptibility to blast in all the regions (**Plate 1a** and **1b**). HR-12 showed highest susceptibility index (7.31) and resistant check Tetep recorded lowest susceptibility index (2.06) while,

Rasi and IR-64 recorded an average score of 2.95 and 3.10 respectively (**Table 2**).

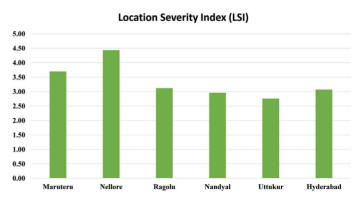


Figure 2: Location Severity Index (LSI) of different test locations

The differential lines RP Biopatho-3 (*Pi-2*), RP Biopatho-4(*Pi-54*) and PRS-59 (*Pi-9*) showed resistant





RARS, Maruteru ARS, Nellore ARS, Ragolu
Plate 1a: Multilocation evaluation of differential lines carrying different blast resistant genes



RARS, Nandyal ARS, Uttukur IIRR, Hyderabad Plate 1b: Multilocation evaluation of differential lines carrying different blast resistant genes



reaction (<3) to blast disease in all the locations. PRS-17 (*Pi-9+Pi-54*) exhibited resistant reaction (<3) in four locations, whereas at IIRR, Hyderabad and ARS, Uttukur it showed moderately resistant reaction of 3.15 and 3.52, respectively. Differentials with combinations of genes BL-122 (*Pi-1+Pi-2*) and A57 (*Pi-1+Pi-2+Pi-4*) showed resistant reaction in all the locations except at RARS, Maruteru and ARS, Nellore. Differential line Zenith (*Pi-z+Pi-a+Pi-i*) showed resistant reaction in all the locations except in ARS, Nellore. C104 PKT (*Pi-3*) recorded susceptible reaction in RARS, Maruteru, ARS, Nellore and ARS, Ragolu. Tadukan (*Pi-ta*) showed susceptible reaction only in ARS, Nellore and it showed resistant reaction in all other locations.

The susceptible check (HR-12) succumbed to blast

in all the locations except at ARS, Uttukur it showed moderately resistant reaction (5.04). The resistant check Tetep ($Pi-k^{h+}$) showed resistant reaction (<3) to blast disease in all the test locations. Resistant check IR 64 was found resistant (<3) only at RARS, Nandyal, while it showed moderately resistant reaction in the remaining locations. Another resistant check Rasi showed resistant reaction (<3) in four locations and at ARS, Nellore and IIRR, Hyderabad it exhibited moderately resistant reaction. The susceptible check Co-39 showed resistant reaction (<3) at ARS, Ragolu and ARS, Uttukur while it exhibited moderately resistant reaction at RARS, Nandyal and IIRR, Hyderabad. It exhibited a moderately resistant reaction at RARS, Maruteru and susceptible reaction at ARS, Nellore.

Table 2: Details and performance of the differential lines to *Pyricularia oryzae* under different agro climatic regions during *kharif* -2023

S.	Desig-	E (N	Resistance				Dis	ease reac	tion to	blast 0-9 S	cale (RRI, 1996	o)*			
No.	nation	Entry No.	Gene	Maruteru	R/S	Nellore	R/S	Ragolu	R/S	Nandyal	R/S	Uttukur	R/S	Hyderabad	R/S	SI**
1	BL1	C101 LAC	Pi-1	2.80	R	5.53	MS	3.43	MR	2.43	R	3.22	MR	3.45	MR	3.48
2	BL2	C101 A51	Pi-2	3.69	MR	5.61	MS	3.18	MR	3.50	MR	3.01	MR	3.88	MR	3.81
3	BL3	C104 PKT	Pi-3	5.37	MS	4.87	MS	5.10	MS	3.17	MR	3.21	MR	3.58	MR	4.22
4	BL4	C101 TTP	Pi-4b	4.91	MS	5.62	MS	2.35	R	2.25	R	2.35	R	2.78	R	3.38
5	BL5	RIL - 10	Pi-12	2.74	R	5.48	MS	2.14	R	3.50	MR	3.11	MR	2.65	R	3.27
6	BL6	RIL - 29	Pi-7	5.31	MS	3.61	MR	3.54	MR	3.63	MR	2.28	R	3.60	MR	3.66
7	BL7	O. minuta	Pi-9	3.63	MR	4.82	MS	2.30	R	3.77	MR	2.10	R	2.27	R	3.15
8	BL8	BL-122	Pi-1 + Pi-2	4.07	MS	4.12	MS	2.69	R	3.12	MR	3.36	MR	2.26	R	3.27
9	BL9	BL-245	Pi-2 + Pi-4	4.25	MS	3.62	MR	2.27	R	3.21	MR	3.04	MR	3.07	MR	3.24
10	BL10	A 57	Pi-1 + Pi-2 + Pi-4	4.60	MS	5.22	MS	2.17	R	2.57	R	2.15	R	2.53	R	3.21
11	BL11	C101 PKT	Pi-4a	3.87	MR	4.56	MS	3.24	MR	3.22	MR	3.23	MR	2.07	MR	3.36
12	BL12	Raminad-STR-3	-	3.32	MR	5.68	MS	3.47	MR	2.53	R	3.02	MR	2.76	R	3.46
13	BL13	Zenith	Pi-z + Pi-a + Pi-i	2.75	R	4.43	MS	2.67	R	2.16	R	2.22	R	2.42	R	2.77
14	BL14	NP - 125	-	2.28	R	5.91	MS	3.48	MR	3.32	MR	3.02	MR	3.20	MR	3.54
15	BL15	USEN	Pi-a ⁺	3.47	MR	3.97	MR	3.57	MR	3.15	MR	3.10	MR	4.89	MS	3.69
16	BL16	Dular	Pi-k ^{a+}	3.28	MR	3.88	MR	2.88	R	2.22	R	3.31	MR	2.63	R	3.03
17	BL17	Kanto - 51	Pi-k	5.21	MS	4.64	MS	2.18	R	2.80	R	3.14	MR	2.79	R	3.46
18	BL18	Shi-tia-tao	Pi-ks	5.44	MS	3.92	MR	2.42	R	2.25	R	2.30	R	5.30	MS	3.60
19	BL19	Calaro	Pi-k ^s	5.22	MS	3.78	MR	2.48	R	3.23	MR	2.37	R	3.17	MR	3.37
20	BL20	Tadukan	Pi-ta	3.69	MR	5.66	MS	2.82	R	2.20	R	2.20	R	2.77	R	3.22
21	BL21	IR - 64	Resistant	3.85	MR	3.89	MR	3.05	MR	2.22	R	3.12	MR	3.05	MR	3.20
22	BL22	Tetep	$Pi-k^{h+}$	2.47	R	2.97	R	2.71	R	1.97	HR	1.83	HR	1.02	HR	2.16



23	BL23	HR - 12	Susceptible	7.98	S	8.69	HS	7.44	S	6.54	S	5.04	MS	9.00	HS	7.45
24	BL24	Rasi	Resistant	2.87	R	3.95	MR	2.78	R	2.14	R	2.22	R	3.67	MR	2.94
25	BL25	Co - 39	Susceptible	4.25	MS	6.92	S	2.35	R	3.12	MR	2.43	R	3.18	MR	3.71
26	BL26	RP Patho-1	Pi-1	2.70	R	4.73	MS	2.64	R	2.37	R	3.10	MR	2.33	R	2.98
27	BL27	RP Patho-2	Pi-2	3.90	MR	4.96	MS	3.46	MR	3.05	MR	2.07	R	2.69	R	3.36
28	BL28	RP Patho-3	Pi-54	2.54	R	3.80	MR	2.32	R	3.12	MR	3.05	MR	3.66	MR	3.08
29	BL29	RP Patho-7	Pi-1	2.70	R	3.60	MR	3.48	MR	3.57	MR	3.01	MR	2.45	R	3.14
30	BL30	RP Patho-8	Pi-2	3.60	MR	3.78	MR	3.78	MR	2.48	R	2.35	R	2.53	R	3.09
31	BL31	RP Patho-9	Pi-54	3.73	MR	5.67	MS	3.50	MR	3.73	MR	3.07	MR	2.22	R	3.65
32	BL32	RP Biopatho-1	Pi-2	3.96	MR	4.69	MS	4.58	MS	2.67	R	2.14	R	2.13	R	3.36
33	BL33	RP Biopatho-2	Pi-54	2.59	R	3.86	MR	2.82	R	3.11	MR	3.15	MR	2.66	R	3.03
34	BL34	RP Biopatho-3	Pi-2	2.62	R	2.90	R	2.88	R	2.28	R	2.13	R	2.71	R	2.59
35	BL35	RP Biopatho-4	Pi-54	2.77	R	2.47	R	2.85	R	2.42	R	2.08	R	2.56	R	2.52
36	BL36	PRS-17	(Pi-9 + Pi- 54)	2.49	R	2.65	R	2.50	R	2.98	R	3.52	MR	3.15	MR	2.88
37	BL37	PRS-50	Pi-54	3.74	MR	2.80	R	3.69	MR	3.44	MR	2.15	R	3.67	MR	3.25
38	BL38	PRS-58	Pi-9	2.71	R	2.75	R	3.44	MR	3.87	MR	3.22	MR	2.47	R	3.08
39	BL39	PRS-59	Pi-9	2.93	R	3.02	R	2.85	R	2.12	R	2.12	R	2.59	R	2.60
	Lo	cation Severity In	ndex (LSI)	3.70		4.44		3.12		2.96		2.76		3.07		

^{*}Blast scale (IRRI, 1996), 0- HR, 1-R, 2 to 3-MR, 4 to 5-MS, 6 to 7-S, 8 to 9-HS; R/S- Resistant/Susceptible, HR-Highly Resistance, R-Resistance, MR-Moderate resistant, MS- Moderate Susceptible, S- Susceptible, HS- Highly Susceptible, ** SI-Severity Index

Statistical analysis was carried out using Two way Anova (RBD), the results obtained are presented in the **Table 3**.

Table 3: LSD of test lines and locations

LSD	Differential lines	Locations
C.D.	0.81	0.32
SE(m)	0.29	0.11
SE(d)	0.41	0.16
C.V.	10.07	10.07

The difference in disease reaction scores of susceptible and resistant checks reveals the shift in the pathogen population. The results also reveal that the disease severity and the performance of genes varied in different agroclimatic zones. Apex and minimal disease severity were recorded in the southern zone alone due to varied climatic conditions. Similar trial was also conducted by Muralidharan *et al.*, (2004) and evaluated rice genotypes carrying resistance genes to blast disease in multi-environment tests (METs) Tadukan carrying resistance gene *Pi-ta* showed small lesions infecting <2% leaf area indicating a very

high level of durable resistance to blast disease. The METs clearly demonstrated the expression of a high degree of resistance in A57 carrying three resistance genes (*Pi-1*, *Pi-2* and *Pi-4*). A 57 was identified as the best line that exhibited resistance to blast across the country in all rice growing environments irrespective of ecosystems.

Similar results were observed by Jahaar *et al.*, (2018) screened 23 Near Isogenic Lines (NILs) at four different agroclimatic locations and reported that NILs with combination of resistant genes *i.e.*, BPT5204×C101LAC×C101A5×Tetep (*Pi-1*, *Pi-2* and *Pi-54*), Swarna×C101LAC×Tetep (*Pi-1* and *Pi-54*) and Swarna×C101LAC×C101A5×Tetep (*Pi-1*, *Pi-2* and *Pi-54*) showed complete resistance to blast disease in all the locations.

In the same way, Abamu *et al.*, (1998) studied effects and Multiplicative Interaction Models which are widely used for analyzing main-effects and genotype by-environment (G×E) interactions in multilocation variety trials to gain insight into G×E in rice blast and



identify genotypes with high and stable resistance to the disease. Divya *et al.*, (2013) also reported that lines with gene combinations Pi-1+Pi-2+Pi-33+Pi-54 and Pi-1+Pi-2+Pi-33 were highly resistant to blast disease than those with single genes indicating that these non-allelic genes have a complementary effect.

The present study offers valuable insights into the genetic factors underlying blast resistance in rice. The Southern Zone saw the highest and lowest LSI values at ARS, Nellore and ARS, Uttukur, respectively, reflecting changes in pathogen virulence and environmental factors. The resistant genes *Pi-1*, *Pi-12*, *Pi-z* + *Pi-a* + *Pi-i*, *Pi-54* and *Pi-9* and the demonstrated effectiveness of combining these genes provide promising avenues for breeding programs to develop cultivars that are more resilient to this devastating disease.

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RESEARCH ARTICLE

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A Sustainable Approach to Rice Cultivation in Punjab-Direct Seeding of Rice (DSR) Sangeet Ranguwal^{1*}, Raj Kumar² and Gurpreet Singh³

¹Agricultural Economist, Department of Economics and Sociology, Punjab Agricultural University, Ludhiana, Punjab; ²Principal Extension Scientist (Agricultural Economics), Department of Economics and Sociology, Punjab Agricultural University, Ludhiana, Punjab;

³Senior Research Fellow, Crop Residue Management, Agricultural Technology Application Research Institute (ATARI) ICAR, Zone I, Ludhiana

*Corresponding author Email: sangeet@pau.edu

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Abstract

Considering sustainable agriculture as the keystone of social and economic prosperity, promotion of Direct Seeded Rice (DSR) has been one of the pioneer steps in the Punjab Government's 2023-24 Budget. In the present study based on field survey of it was found that use of all the inputs was lower on DSR farms as compared to conventional Puddled Transplanted Rice (PTR) farms except seed, plant protection chemicals and some micro-nutrients. Though the yield in DSR (2769 kg per acre) was marginally lower than PTR (2801.3 kg per acre), the net returns over variable cost were higher by about 13 per cent in DSR (Rs 31482.14 per acre) than PTR (Rs 27788.41 per acre) because of lower variable costs involved in DSR. The cost in production of one kg grain using DSR was found to be lower (Rs 8.43 per kg) by about 15 per cent than in PTR (Rs 9.88 per kg) and the input energy involved in the same was Rs 7.84 MJ as compared to 8.86 MJ indicating that DSR has the potential to increase farmer's income and save scarce resources. DSR technology is a viable alternative to overcome the problems of rising cost of cultivation, labour and water shortages for sustainable rice production yet it has not been adopted at a very large scale. There is need for more research in development of high yielding rice cultivars suitable for DSR along with ensured and timely availability of agro-inputs and machinery. Also, there is a need to ³generate more awareness of recommended DSR production practices among the farmers for its speedy adoption and thus achieving sustainable production.

Keywords: DSR, social and economic prosperity, farmers income, high yield rice, sustainable production

Introduction

Rice is one of the most important food crops of India. About 70 per cent Indians use rice as their primary food source and it occupies 40-45 per cent of all the land under cereal crops in India. The growing demand of rice has to be met by producing more rice with less agricultural input usages. Different challenges like lowering of water table, labour shortage during peak times and declining soil quality necessitate an alternative establishment approach

to maintain rice production as well as natural resources. When the future of rice production is in jeopardy due to worldwide water constraint, Direct Seeded Rice (DSR) presents a desirable alternative. The conventional Puddled Transplanting of Rice (PTR) is water, capital, energy and labour-intensive practice. There is an urgent need to switch from the traditional PTR to DSR because it is not only cost, input, energy and time saving but is also environment



friendly (Bandumula et al., 2018; Jat et al., 2022; Singh et al., 2023a).

Punjab state has been playing a leading role in the agricultural transformation of the country. A sustainable production of rice in the state is crucial for the food security of India. The state has contributed about 25-30 per cent rice and 35-40 per cent wheat to the central pool during the last one decade (PAU, 2022). Water guzzling paddy is a dominant crop in the cropping pattern of the Punjab state and is putting the groundwater resources in a jeopardy situation. Out of 153 water blocks of Punjab, only 17 are safe and the remaining 136 (89%) are in alarming condition (Anonymous, 2022). Further, electricity demand is increasing for irrigating the paddy crop which undermines the viability of the power sector as power for agricultural use is fully subsidized in the state. In addition, yield stability and assured marketing of paddy makes it the most remunerative rainy (kharif) season crop.

During the COVID-19 lockdown, the return of migrant labour who were working in Punjab, to their native places, created a severe shortage of labour during the kharif season of 2020 in Punjab. During that time the DSR, which was being promoted in the State for a long time as a more water and labour efficient alternative of paddy cultivation, seemed more attractive to the farmers. They perceived that the economic losses of shifting to DSR were significantly less than shifting to alternative crops due to larger market risk and disruption of supply chains for alternative crops. Further, the Government of Punjab encouraged DSR by distributing about 4000 DSR machines at subsidized rates along with largescale efforts on extension activities to promote this technology. Reportedly, about 5 lakh hectares (ha) area under paddy was sown through DSR during that time (Vatta et al., 2021). But the results were not encouraging during the subsequent years as the

paddy area cultivated with DSR technology was much lesser than the targets. During 2022 kharif season despite announcing incentives for farmers, the Punjab government missed its DSR target by a huge margin. Against the target of 30 lakh acres (12 lakh hectares), the government managed to bring only 1.68 lakh acres (5.6% of target) under DSR (HT 2022). Various studies (Kaur and Kaur 2017, Kaur and Singh 2017, Kaur et al., 2017, Sidana et al., 2020, Vatta et al., 2021; Singh et al., 2023b) listed poor initial germination, poor crop look, high weed infestation, problem of rodents, lesser yield, high risk to crop in DSR and non-availability of DSR drill as the reasons for shifting back to traditional technique of PTR cultivation. Further, there were considerable variations in the practices of cultivation followed by the farmers from the recommended practices (Kamboj et al., 2022). Due to this they faced plenty of technical issues regarding the establishment of the crop and ploughed back the crop. Keeping all this in view, the present study was carried out in Punjab state with the following objectives.

Objectives

- 1. To examine the extent of DSR adoption in Punjab
- 2. To compare input use and energy consumption pattern in DSR and PTR
- 3. To study the reasons for non-adoption of DSR in the state.

Materials and Methods

To study the extent of adoption of DSR in the Punjab state, secondary data for the year 2023-24 was gathered from the Department of Agriculture, Punjab. Further, to accomplish the objectives of the study, primary data was collected by using multi-stage random sampling technique. At the first stage, one district namely Sri Mukatsar Sahib having the highest area under the DSR technology for paddy cultivation was identified through consultation with officials of the Punjab



State Department of Agriculture. Keeping in view the concentration of DSR technology, two blocks namely Gidderbaha and Mukatsar were selected at the second stage (Figure 1). From each selected block, two villages were selected for the study, as shown in Figure 1.

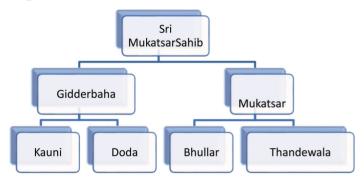


Figure 1: Distribution of survey sample

20 DSR farmers were chosen from each selected village for the study using simple random sampling technique. In order to undertake impact assessment of the DSR technology, ten PTR farmers from the same vicinity were also taken as a control group in the analysis. Thus, the total sample for the study comprised of 120 farmers (80 DSR and 40 PTR farmers) spreading over different farm size groups based on operational holding i.e. small (up to 5 acres), medium (>5 to 15 acres) and large (more than 15 acres).

The primary data pertaining to the two systems of rice cultivation i.e. DSR and PTR were collected from the sample farmers for the agricultural year 2021-22 through personal interview method. Requisite information relevant to various inputs used in paddy cultivation such as seed, diesel fuel (consumed for various farm operations viz. seed bed preparation, inter-culture operations, harvesting, transport on farm etc.), fertilizers, Farm Yard Manure (FYM), chemicals (insecticides, fungicides, herbicides), total working hours of labour (men and women hours) as well as draught power used for different farm operations along with total working hours of agri-machinery were recorded. The information on capacity of the pumps used by the farmer for irrigating in terms of horse

power (Hp) was also collected from the respondents. Data on paddy grain yield was used for the estimation of straw yield using crop to residue ratio method (Chauhan, 2012).

The data on inputs used in paddy cultivation and output (grain and straw) were converted to energy units using embodied energy equivalents for each input and output energy type and expressed in Mega Joules (MJ) using specific energy coefficients taken from the Research Digest on Energy Requirement in Agriculture Sector, Department of Farm Power and Machinery, PAU (Singh and Singh, 2002) as mentioned in **Table 1**.

Table 1: Energy coefficients for energy calculation in paddy cultivation

Sr. No.	Energy source	Energy coefficient (MJ/unit)
1	Human labour (h)	1.96
2	Animal labour (h)	14.05
3	Fertilizer (kg)	
	N	60.6
	P_2O_5	11.1
	K ₂ O	6.7
4	Farmyard manure (FYM) in kg	0.3
5	Chemicals (kg and litre respectively)	120 and 102 respectively
6	Machinery (h)	62.7
7	Diesel (litre)	56.31
8	Seed/Grain (kg)	14.57
9	Straw (kg)	12.5
10	Electricity (KWh)	11.93

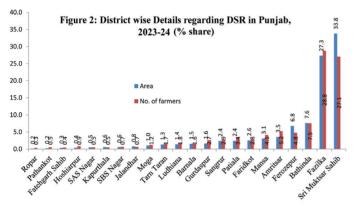
Source: Singh and Singh, 2002

Status of adoption of DSR in Punjab

Considering sustainable agriculture as the keystone of Punjab's social and economic prosperity, promotion of DSR remained one of the pioneer steps in the Punjab Government's 2023-24 Budget.



About 17026 thousand famers have been provided with an incentive Rs1500 per acre for adopting the practice of DSR, for which Rs 19.92 crore has been paid during 2023-24. During 2023-24, the area under DSR was 53 thousand hectares forming only about 2 per cent of the total area under paddy. District wise analysis of the data revealed that the highest proportion of area under DSR was in Shri Mukatsar Sahib (33.8%) followed by Fazilka (27.3%), Bathinda (7.6%), Firozpur (6.86%) and Mansa (3.1%) while in other districts it was below 3 per cent as shown in **Figure 2.**



Source: Department of Agriculture, Punjab

In terms of number of farmers availing the subsidy, the maximum belonged to Fazilka (28.8%) followed by Shri Mukatsar Sahib (27.1%), Bathinda (7.5%), Amritsar (5.2%), Ferozepur (4.8%) and Mansa (4.1%).

Thus, DSR paddy adoption rate was higher in the south western districts of the state. One major reason behind this is that ground water is not fit for irrigation in most of the villages in this area. According to an earlier study, the adoption of DSR was higher amongst the farmers with relatively lower access to irrigation (Vatta *et al*, 2021).

According to estimates of Department of Economics and Sociology PAU, it was observed that maximum area under DSR during 2023-24 was under recommended non-basmati paddy variety PR126, requiring 120

days to mature followed by un-recommended Pusa 44 (15%) which is a long duration variety of paddy needing 155-160 days to mature (**Figure 3**). Among basmati varieties, Pusa Basmati 1718 occupied about 10 per cent of the DSR area followed by unrecommended Pusa Basmati 1847 (5%) and Pusa Basmati 1401 (4%) with rest being on less than three per cent of the DSR area.

2023-24 (% share) Others (20%) Pusa Basmati 1885 , (3%) Pusa Basmati 1121 , (3%) Pusa 44. (15% Pusa Basmati 1401, (4%) Pusa Basmati 1847, (5%) PR 131, (5%) Pusa Basmati PR 114, (6%) 1718 . (10%) CR 212, (6%)

Figure 3: Variety wise area under DSR in Punjab,

Source: Department of Economics and Sociology, PAU

Comparison of DSR to PTR in terms of input use energy consumption

The resource use in paddy cultivation on different farm sizes under DSR method of cultivation selected for the present study is given in **Table 2**. Analysis of the data revealed that use of human labour worked out to be 101.03 hours per acre on an average and machine labour (use of machinery for various cultural operations comprising mainly land preparation, irrigation, harvesting and on farm post-harvest operations) ranged between 7.85 - 8.67 hours per acre and it was 8.3 hours per acre on an average.



Table 2: Input use pattern, energy consumption and returns for paddy cultivation by DSR in Punjab

(Per acre)

Sr. No.	Input/Farm category	Small	Medium	Large	Overall (DSR)	PTR	DSR wit	fference for h traditional cultivation
					(DSIL)		Input use	Energy consumption
1	Human Labour (h)	100.05	101.01	102.09	101.03	169.90	-68.87**	-122.38**
2	Animal Labour (h)	1.11	0.75	0.40	0.75	1.00	-0.25	-3.51
3	Machine Labour (h)	7.85	8.38	8.67	8.3	9.50	-1.20**	-18.73**
4	Diesel (litre) #	39.8	42.45	46	42.75	46.50	-3.75*	-211.15**
5	Seed (kg)	7.48	7.9	8.29	7.89	5.30	2.59**	38.07**
6	Fertilizers, micro nutrients and FYM							
a	Urea (kg)	143.0	141.0	147.5	143.80	170.62	-26.82**	-1696.63**
b	Phosphatic (kg)	6.21	6.53	6.90	6.54	6.80	-0.26	-2.89
c	Muriate of Potash (kg)	5.62	6.30	6.50	6.14	6.80	-0.66	-4.42
d	Zinc (kg)	4.50	5.20	5.70	5.13	6.50	-1.37*	-28.63*
e	Iron Sulphate (kg)	4.90	5.90	6.66	5.82	4.60	1.22	12.23
f	Others (kg) ##	2.50	3.20	3.60	3.18	2.70	0.48	4.80
g	FYM (Ton)	5.30	5.82	6.10	5.74	5.90	-0.16	-78
7	Plant Protection Chemicals							
a	Rodenticide (kg)	1.20	1.62	1.80	1.54	0.50	1.04**	125.98**
b	Insecticide (litre andkg)	2.30	2.60	3.20	2.70	2.23	0.47	47.98
c	Weedicide (litre)	2.80	3.30	3.55	3.21	1.20	2.01**	205.22**
8	Electricity for irrigation (KWh)	603.03	609.01	618.16	610.05	725.40	-115.35**	-1376.12**
9	Output							
A	Grain (Qtls)	27.20	28.01	27.88	27.69	28.01	-32.30	-1.15
В	Straw (Qtls)	36.72	37.81	37.63	37.38	37.81	-43.61	-1.15

Non-significant differences were observed among farm categories

#includes use of tractor for land preparation, irrigation, transport on farm and harvester combine

##includes seed treatment chemicals and growth regulators; ** and * significant at one and five per cent level of significance

Source: Field Survey

Thus, with farm size the use of human as well as machine labour increased. Consequently, the diesel fuel used in prime movers and oil engines/generators for running pumps on small farms (39.80 litre per acre) was lesser than on large farms (46.00 litre per acre) with average figure being 42.75 litre per acre. On the contrary, the animal labour use for on farm transportation showed inverse relationship with the

farm size. The use of animal labour was reported to vary between 0.40 hours on large to 1.11 hours per acre on small farms and average figure worked out to be 0.75 hours per acre. In a similar kind of study for Punjab, maximum value for mechanization index was observed in the case of large farmers and maximum animal labour index was observed in the case of marginal farmers (Kaur *et al*, 2017).



Further analysis revealed that the seed rate increased with rise in the farm size. On an average, 7.89 kg/acre seed rate was followed by DSR adopters as against recommended seed rate of 8 kg/acre and it was the highest on large farms (8.29 kg /acre), followed by medium (7.90 kg/acre) and small farms (7.48 kg/acre). On the other hand, dose of urea applied was higher for small farmers than the medium category farmers. Two main reasons behind this pattern are lack of knowledge among farmers about the recommended package of practices and existing nutrient based subsidies on these chemical fertilizers. The DSR adopters were found to be using much higher dose of urea than recommended by the PAU (130 kg per acre). High magnitude of subsidies for nitrogen fertilizer extended by the government indirectly encouraged the farmers to apply larger quantities of nitrogen fertilizer for paddy crop. The average figures for the use of different chemical fertilizers namely urea, phosphatic fertilizers, muriate of potash and micro nutrients zinc, Iron sulphate and others (including seed treatment chemicals and growth regulators) were estimated to the tune of 143.80, 6.54, 6.14, 5.13, 5.82 and 3.18 kg per acre respectively on an average and their use was also found to increase with the farm size. Almost similar results were reported by Saha et al., 2020.

The use of farm yard manure (FYM) was the highest on large farms (6.10 ton per acre) and the least on small farms (5.30 ton per acre) and this happened due to high availability of FYM from large livestock with the large farmers. As regards the use of plant protection chemicals (PPC) is concerned, the average use of rodenticides, insecticides (both liquid and granular) and weedicides turned out to be 1.54 kg, 2.70 (litre and kg) and 3.21 litres per acre respectively and their use was the highest by the large farm category. Similarly, the use of electricity for the irrigating one acre of DSR paddy turned out to be the highest on large farms (618.16 KW) as compared to small (603.03 KW) and

medium farms (609.01 KW) though it was freely available to all the farm categories. The pumping of irrigation water from deeper layers of underground water through submersible electric pumps and electric motors has led to the high electricity consumption in the state. Further, on account of free of cost supply of electric power to agricultural sector in Punjab state, farmers had no incentive in saving electricity. The output from paddy cultivation in terms of grain and straw production of paddy was to 27.69 and 37.38 quintals on an average and it was the marginally high for the medium farm category (28.01 Qtls per acre) than small (27.2 Qtls per acre) and large farms (27.88 Qtls per acre). The analysis revealed that the input use in DSR paddy cultivation increased with the farm size, except use of animal labour. However the differences existed among the different farm categories in input use was non-significant.

The results for comparative input use pattern under DSR and PTR method revealed that human labour use was found to be about 41 per cent higher for PTR (169.9 hours) than for DSR (101.03 hours) as the human labour requirements in DSR were reduced due to no need for transplanting the paddy seedlings. Machine labour use was also higher by about 13 per cent for PTR (9.50 hours) than DSR (8.30 hours) and consequently about 8 per cent higher diesel use existed in PTR (46.5 litre) than DSR (42.75 litre). Compared to the average seed rate used by DSR adopters (7.89 kg), the PTR followers used only 5.30 kg of seed for sowing one acre of paddy because of self-confidence in their farming practices. Among different chemical fertilizers, the use of urea, phosphatic fertilisers, muriate of potash and micro nutrients-zinc and Iron sulphate, was higher for PTR than DSR except Iron sulphate (lower for PTR by 1.22%) and seed treatment chemicals and growth regulators (by 0.48%). On the contrary, the use of PPC was much higher by the DSR adopters. Due to huge weed infestation, almost double amount of weedicide



application per acre (3.21 litre) was observed for DSR than PTR (1.20 litre). Further, use of rodenticides to avoid rodent attack was three times higher side in DSR (1.54 kg) than PTR (0.50 kg). Insecticide application was also higher in DSR (2.70 kg) than PTR (2.23 kg) though the difference was statistically non-significant. The use of electricity for the irrigation was higher on PTR (725.40 KW) than the DSR farms (610.05 KW) by about 16 per cent because of lesser number of irrigations and water application in DSR.

The analysis revealed that use of all the inputs was lower on DSR farms as compared to PTR farms

micro nutrient iron sulphate. The mean difference of major inputs such as human labour, machine labour, diesel fuel, seed rate, urea, rodenticides, weedicides, electricity differ significantly between DSR and PTR method of paddy cultivation. Accordingly, energy use was also lower in DSR system.

except seed, plant protection chemicals (PPC) and

In terms of important economic parameters such as yield, total variable cost, gross returns, net income of DSR over PTR method of paddy cultivation the results are presented in **Table 3**.

Table 3: Economic benefits of DSR vs PTR in Punjab

(per acre)

Sr. No.	Particulars	DSR	PTR	Advantage in DSR (%)
1	Yield (kg)	2769.00	2801.30	-1.15
2	Total variable cost (Rs)	23344.06	27677.33	-15.66
3	Gross returns (Rs)	54826.20	55465.75	-1.15
4	Net returns over variable cost (Rs)	31482.14	27788.42	13.29
5	Cost of Grain production (Rs per kg)	8.43	9.88	-14.68
6	Energy (MJ per kg)	7.84	8.86	-11.51
7	Total Energy Input (MJ/Acre)	21708.61	24816.79	-12.52
8	Total Energy Output (MJ/Acre)	87431.18	88451.05	-1.15

Source: Field Survey

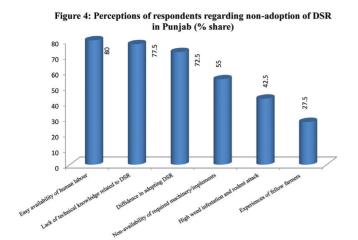
The results revealed that though the yield in DSR (2769 kg per acre) was marginally lower than PTR (2801.3 kg per acre), the net returns over variable cost were higher by about 13 per cent in DSR (Rs 31482.14 per acre) than PTR (Rs 27788.41 per acre). This is because of lower variable costs involved in DSR. The cost in production of one kg grain using DSR was found to be lower (Rs 8.43 per kg) by about 15 per cent than in PTR (Rs 9.88 per kg) and the input energy involved in the same was Rs 7.84 MJ as compared to 8.86 MJ.

Hence, the results of the present study indicated that DSR is an input saving as well as cost saving technology for paddy cultivation in comparison to traditional PTR method.

Perceptions of the respondents regarding non-adoption of DSR

Some genuine reasons and perceptions of the respondents for 'not adopting' the DSR has been presented in **Figure 4.** The results revealed that easy availability of labour for transplanting paddy emerged to be the major reason for not following the DSR by 80 per cent respondents. Other perceptions like lack of technical knowledge of DSR (77.5%), diffidence in adopting DSR (72.50%) and non-availability of required machinery/implements (55%) for sowing were the major reasons of not adopting the DSR.





Source: Field Survey

Other reasons such as high risk of weed infestation and rodent attack and experiences of fellow farmers were reported by about 28 and 43 per cent, respectively. In a study by Kaur and Singh, (2017) at Punjab Agricultural University Ludhiana several constraints associated with shift from PTR to DSR included, high weed infestation, evolution of weedy rice, increase in soil borne pathogens (nematodes), nutrient disorders, poor crop establishment, lodging, incidence of blast, brown leaf spot etc.

Conclusions and Suggestions

Currently, DSR is emerging as an option for sustainable rice production, owing to limited water availability, shortage of labour and rising cost of cultivation. Though the method is economically advantageous and also farmer-friendly, yet it needs technological advancement to reap the full benefits and speedy adoption. It points towards need for more research in development of high yielding rice cultivars suitable for DSR in various agro-climatic situations along with ensured and timely availability of agroinputs and machinery at affordable prices. There is a need to generate more awareness of recommended DSR production practices among the farmers along with the benefits of such practices. Embracing of such standard practices especially judicious use of inputs like fertilizers, underground water and plant

protection chemicals will not only optimize the energy use but also will minimize the cost of cultivation. There is a need to focus more on capacity building by educating/training the young farmers for promotion of DSR. A campaign with the combined efforts of various stakeholders such as government agencies and non-government organisations as change agents will help in fast pacing the adoption process of DSR.

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SHORT COMMUNICATION

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BPT 2846 (IET 28737), A High Yielding Slender Grain Rice Variety Suitable for Krishna Zone of Andhra Pradesh

Krishna Veni B¹, Tushara M^{2*}, Satyanarayana PV³, Ramana JV⁴, Suneetha Y⁵, Roja V⁵, Srinivas T⁵ and Anny Mrudhula K⁶

¹Agricultural Research Station, Bapatla, ²Agricultural College, Bapatla, ³Director of Research, ANGRAU, ⁴Planning and Monitoring cell, ANGRAU, ⁵Regional Agricultural Research Station, Maruteru, ⁶Agronomy, SWS, Bapatla

*Corresponding author Email: m.thushara@angrau.ac.in

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Abstract

BPT 2846, developed through pedigree breeding at the Agricultural Research Station, Bapatla, is a result of the cross between MTU 1061 and IR78585-64-2-4-3-1. During the *kharif* seasons of 2015-2017, it underwent station yield trials, demonstrating an average grain yield of 6352 kg/ha. This marked a substantial 18.6% improvement over the reference variety BPT 5204. In the Multi-location Trial-Slender grain of kharif 2019, BPT 2846 showcased a noteworthy average grain yield of 6355 kg/ha, exhibiting a 23.0% improvement over the common check BPT 5204. Nominated in the AICRIP trial under the Initial Variety Trial-Medium Slender category as IET 28737 in 2019, BPT 2846 displayed adaptability across various locations in the country, recording a mean grain yield of 4420 kg/ha. The rice variety was further subjected to adaptive minikit testing in farmers' fields by DAATT Centres for three years, resulting in an average grain yield of 6513 kg/ha. This reflected an 11.8% improvement over BPT 5204. Similarly, minikit testing conducted by DAOs recorded an average grain yield of 6497 kg/ha for BPT 2846, demonstrating a 12% yield improvement over BPT 5204. Belonging to the late duration group with a 145-150 days duration in the kharif season, BPT 2846 is wellsuited for irrigated wetlands in Andhra Pradesh. It possesses characteristics such as non-lodging, a semi-dwarf and erect plant type, dark green foliage, two weeks of seed dormancy, low shattering and high grain yield. Additionally, BPT 2846 exhibits moderate resistance to leaf blast, neck blast and leaf folder. It boasts excellent cooking quality, intermediate values of alkali spreading and amylose content, high head rice recovery (65.2%) with translucent grains and a Kernel Length to Breadth ratio of 2.85. These attributes make it highly desirable for obtaining a premium market price.

Keywords: Rice variety, medium slender grains, high yielding, late duration

Introduction

One of the main objective of the Agricultural Research Station, Bapatla is to create a high-yielding rice variety tailored for the *kharif* season, featuring a duration of 145-150 days. Even though, BPT 5204 possess good yield potential and excellent grain and cooking quality, it is susceptible to major pests/diseases due to which

often farmers suffer heavy yield losses. Hence, there is a demand from the farming community for stable rice varieties with sustained production even under adverse climatic conditions with reduced cost of cultivation. Efforts are underway at Agricultural Research Station, Bapatla for the development of high yielding rice



varieties with 145-150 days duration (suitable for double cropped areas), non-lodging, low shattering of grains along with resistance to leaf blast, neck blast, plant hoppers coupled with good grain quality suitable for raw rice. Many varieties possessing high yield potential with tolerance to blast were released for replacement of Samba Mahsuri. But, many of them were not accepted by farmers due to their poor cooking quality traits. After years of dedicated efforts starting in 2009, BPT 2846, a rice variety with favorable features capable of replacing BPT 5204, was finally released by SVRC in 2023.

BPT 2846 possess excellent grain and cooking quality parameters similar to Samba Mahsuri. The cultivar BPT 2846 has the following desirable features.

- Duration: 145-150 days
- Erect plant type
- Strong culm, Non-lodging
- One-week seed dormancy
- Moderately resistant to leaf blast, neck blast and leaf folder
- Low shattering of grains

- Suitable for raw rice
- Medium slender grain with high market price and good consumer acceptability

The proposed cultivar, BPT 2846 is a high yielding, non-lodging, semi-dwarf, profuse tillering culture, possess medium slender grains with straw glume with 145-150 days duration suitable for double cropped areas also. It has two weeks seed dormancy. It is very much suitable for cultivation during *kharif* season in Krishna zone of Andhra Pradesh in realizing higher yields. Due to its non-lodging nature, the proposed culture is suitable for cultivation under direct sown conditions and for mechanical harvesting also.

Biotic stress resistance:

The screening trials data revealed that the suggested variety demonstrates moderate resistance to leaf blast, neck blast and leaf folder. This finding suggests that farmers may reduce pesticide sprayings, resulting in lower cultivation expenses and promoting sustainable yields (Tables 1 and 2).

Table 1: Reaction of BPT 2846 to major diseases

Variate	National Screen (AICRIP) testin	ning Nursery-II ng during 2019)	Variotes	ARS, Nellore during, 2020-21					
Variety	Leaf Blast	Neck Blast	- Variety	Leaf Blast	Neck Blast	Sheath Rot	BLB		
BPT 2846	5.2	3.0	BPT 2846	5.0	5.0	3.0	5.0		
Check (BPT5204)	6.3	7.0	Check (BPT5204)	9.0	9.0	7.0	9.0		
Resistant Check (Tetep)	3.0	2.0	Resistant Check	-	-	-	-		
Susceptible Check (Pusa 44)	5.3	8.0	Susceptible Check (NLR34242)	9.0	9.0	9.0	9.0		

Table 2: Reaction of BPT 2846 to major insect pests

Variety	National Screening Nursery - II (AICRIP) testing during 2019)						
variety	Leaf Folder Score						
BPT 2846	0.8						
Check (BPT5204)	2.46						
Resistant Check (W1263)	1.85						
Susceptible Check (TN1)	3.36						



BPT 2846 grain quality:

BPT 2846 exhibits outstanding cooking quality, along with intermediate and desirable values of alkali spreading value and amylose content. Additionally, it boasts high head rice recovery (65.2%) characterized by translucent grains and a Kernel Length to Breadth ratio of 2.85, making it highly sought after for obtaining a premium market price (**Table 3**).

Table 3: Physico-chemical and Biochemical quality characteristics of unpolished BPT 2846 and BPT 5204

S. No	Trait/ Character	Description of BPT 2846	Description of BPT 5204
1	Hulling (%)	76.6	75.8
2	Milling (%)	69.3	68.4
3	Head rice recovery	65.2	62.8
4	Kernel length (mm)	5.42	5.63
5	Kernel breadth (mm)	1.90	2.14
6	L/B ratio	2.85	2.63
7	1000 grain weight (g)	13.5-14.0	14.5-15.0
8	Gelatinization	Intermediate	Intermediate
	temperature		
9	Keeping quality	Good	Excellent
10	Cooking quality	Excellent	Excellent
11	Alkali spreading value	4.5	5.0
12	Amylose content (%)	22.7	22.65
13	Protein content in	8.2	8.0
	unpoli-shed rice (%)		
14	Fe content in	6.2	8.7
	unpolished rice (ppm)		
15	Zn content in	21.8	16.8
	unpolished rice (ppm)		

The organoleptic evaluation of BPT 2846 along with other rice varieties revealed that it recorded on par score with the popular BPT 5204 for overall acceptability and possess 8.2% protein content in brown rice. The unpolished rice of BPT 2846 possessed, 21.8 ppm Zn and 6.2 ppm iron content.

Performance in Multi-Location Trials and Adaptive minikit tests:

BPT 2846 was evaluated in station yield trials in *kharif* 2015-2017 and recorded an average grain yield of 6352 kg/ha in station trials with 18.6% yield improvement

over the check variety BPT 5204. In Multi-Location Trial-Slender Grain conducted during *kharif* 2019, BPT 2846 recorded 6355 kg/ha and showed 23.0% yield improvement over common check BPT 5204. BPT 2846 (IET 28737) was nominated in AICRIP trial (Initial Variety Trial- Medium Slender) during 2019 and recorded a mean grain yield of 4420 kg/ha in different locations tested across the country.

The entry BPT 2846 was evaluated in adaptive minikit testing conducted by DAATT Centres in farmer's fields of erstwhile Krishna, East Godavari, Guntur, Kurnool, Darsi, Srikakulam, Vizianagaram, Visakhapatnam, Nellore, Ongole and Peddapuram districts in *kharif* season during the years 2019-20, 2020-21 and 2021-22. Across all the locations over three years, it recorded an average grain yield of 6513 kg/ha and exhibited 10.6% yield superiority overchecks *viz.*, BPT 5204/BPT 2782 and exhibited 11.8% improvement over BPT 5204.

Minikit testing conducted by the JDA's of erstwhile Guntur, Nellore, Krishna, East Godavari, Prakasam, Visakhapatnam and Vizianagaram districts of Andhra Pradesh, BPT 2846 recorded an average grain yield of 6497 kg/ha and 11.3% yield improvement over checks *viz.*, BPT 5204/BPT 2782. When compared with BPT 5204 it recorded 6545 kg/ha and 12% yield improvement.

During 2021-22, BPT 2846 was tested against BPT 2782 in Guntur, Nellore, Krishna, Visakhapatnam and Prakasam districts and it recorded 6563 kg/ha grain yield and exhibited 8.6% yield superiority over the recently released variety *i.e.*, BPT 2782.

Agronomy trials were conducted for BPT 2846 at Agricultural Research Station, Bapatla. The results revealed that BPT 2846 recorded significantly superior grain and straw yield at 160 kg nitrogen/ha.



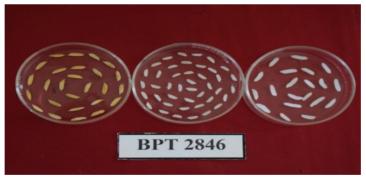
Conclusion

Farmers, industry (millers) and consumers alike have embraced BPT 2846. Over 50,000 acres in Krishna, Guntur, Nellore and Prakasam Districts were dedicated to planting BPT 2846 during the *Kharif* seasons of 2021 and 2022. Millers express great satisfaction due

to its impressive milling capabilities and high head rice recovery, accompanied by minimal breakage (2-3%). Consequently, millers are offering a premium price for BPT 2846. Consumers are content with the cooking and eating quality parameters of BPT 2846.



Field View of BPT 2846



Paddy and Rice of BPT 2846



Panicles of BPT 2846



Paddy and Rice of BPT 2846



SHORT COMMUNICATION

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DRR Dhan 74 (IET 30252) a Climate Resilient High Yielding Rice Variety with Low Soil Phosphorous Tolerance

Divya Balakrishnan*, Sarla Neelamraju, Sundaram RM, Swamy AVSR, Jyothi Badri, Padmavathi G, Anantha MS, Senguttuvel P, Aravind Kumar J, Sai Prasad SV, Neeraja CN, Kalyani MB, Suneetha K, Prasad MS, Ladhalakshmi D, Laha GS, Gireesh C, Jhansi Lakshmi V, Padmavathi Ch, Padmakumari AP, Latha PC, Mahendrakumar R, Sreedevi B, Tuti M, Mangrauthia SK, Brajendra P, Manasa V, Sanjeeva Rao D, Muthuraman P, Pranay G, Rao YV, Krishnamraju A, Kavitha B, Vijay Kumar M and Tahseen M

ICAR-Indian Institute of Rice Research (ICAR-IIRR), Rajendranagar, Hyderabad-500030 *Corresponding author Email: dbiirr23@gmail.com

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Abstract

Changing climate conditions along with nutrient deficient soils adversely affect the environment and reduce rice production. Wild introgression lines derived from KMR 3 and *O. rufipogon* were evaluated in various environments to identify high yielding stress tolerant genotypes. DRR Dhan 74 (IET 30252) is a variety developed by IIRR, using parental lines from an Indian accession of *Oryza rufipogon* (IC22015/WR120) collected from Kerala, India, which was maintained at IIRR (erstwhile DRR) and was used as a donor parent. KMR 3, a restorer line for popular high yielding hybrid KRH 2 was used as recurrent parent and the variety was developed through advanced back cross breeding and selection. This short bold grain type variety is suitable for both normal irrigated and low soil phosphorous conditions in *kharif* and *rabi* seasons. An average yield of 7 t/ha under normal conditions with recommended dosage of fertilizers and 4.5 t/ ha under low Phosphorus condition was reported under AICRPR multi location trials. Variety possesses good cooking quality with short bold grain type and higher yield and moderately resistant reaction to leaf blast, neck blast, sheath rot and plant hoppers. It was released for cultivation in Karnataka, Maharashtra, Telangana and Jharkhand through Central Sub-committee on Crop Standards, Notification and Release of Varieties for Agricultural Crops vide S.O. 4027(E) dated 8th Oct 2024, CG-DL-E-14102024-257835.

Keywords: Rice variety, Low P tolerance, Climate resilience, Wild introgression

Introduction

Wild species are known for their stress tolerance and growth in resource-limited conditions. Several genes which contribute to yield and stress tolerance were lost during domestication and are not available in the existing gene pool of cultivars. Prebreeding programme at IIRR was initiated with the objective of bringing the lost valuable genes to cultivar background

and enhance the allelic diversity for climate resilience. Phosphorus is the second most important key nutrient, vital for plant growth and development at all stages. Phosphorous deficiency is one of the factors limiting rice yields and farmer's profitability so it is necessary to identify genotypes with stable yield and tolerance to P deficiency. DRR Dhan 74 (IET 30252) is a



climate resilient rice variety developed by ICAR-IIRR using an interspecific cross (RP Bio 4919) between KMR 3 and *Oryza rufipogon* (IC22015/WR120). It is high yielding under both low soil phosphorus and normal irrigated condition. An advanced backcross strategy with repeated selections was followed to develop the variety as it is having a wild species in the parentage.

The high yielding lines in the genetic background of KMR 3 were characterized for 3 years and were further advanced based on single panicle selection up to BC₃F₁₀ and the seed was multiplied. RP Bio 4919-NSR 5 is selected from a back cross introgression line derived from this cross, the genotype was tested rigorously in normal and low p conditions for several seasons and only tolerant line was advanced further. Interspecific introgression of *O. rufipogon* into *O. sativa* background helped to improve yield and tolerance to low Phosphorus in soil.

DRR Dhan 74 recorded an yield of 7 t/ha (under normal conditions; 60 kg/ha of P i.e., recommended dose), 4.4 t/ ha (under low Phosphorus; 40 kg/ha of P) and 4.56 t/ha (under no Phosphorus; 0 kg/ha of P). It recorded average grain yield advantage of +14.43% and +2.96% (in terms of weighted average) over the positive check Swarna (late duration) under 0% and 50% application of phosphorous. It also showed a yield advantage of +79.56, +14.43%, +20.58 and +5.94 over Rasi, Swarna (Positive Check), Improved Samba Mahsuri (Negative check) and Qualifying check 1 (IET 30242) respectively considering weighted mean average of kharif 2021, 2022 and 2023 under no application of recommend dose of fertilizer (RDF) phosphorus. Similarly, at Low phosphorus condition it yielded with +14.44, +2.96, +14.78 and +8.13% advantage over Rasi, Swarna, Improved Samba Mahsuri and Qualifying check 1 (IET30242) respectively considering weighted mean average of kharif 2021, 2022 and 2023 under application 50% of recommend dose of fertilizer (RDF) phosphorus. Based on the performance under both 0% and 50% P of RDF. IET 30252 was found promising in zone 7, zone 3, zone 6 and also based on overall mean for all three testing years of 2021, 2022 and 2023 and promoted in AICRPR testing. In addition, entry was found superior to best check during 2021 in Karnataka and overall, it yielded 4.94 t/ha with 2nd rank and 16% yield advantage over best check. During 2022, recorded 1st, 4th, 6th and 9th rank in the states of Bihar, Telangana, Karnataka and Maharashtra. During 2023, recorded 2nd, 3rd and 5th rank in the states of Telangana, Jharkhand and Karnataka.

Variety possesses short bold grain type with high HRR (62.2%) and acceptable grain quality parameters of amylose content (23.7%), gel consistency (22) and alkali spreading value (7.0) and cooking quality parameters. Based on molecular marker analysis, it has shown > 95% recovery of recurrent parent genome. Considering better performance of the variety for yield in P deficit as well as under normal conditions and stress tolerance, grain and cooking quality traits IET 30252 was released in the states of Karnataka, Telangana andhra Pradesh and Jharkhand as DRR Dhan 74. Hon'ble Prime Minister of India dedicated DRR Dhan 74, a climate resilient variety to the farmers of the Nation on 11 August, 2024. This variety has potential to replace medium to late duration short bold varieties in low-input areas of the country with reduced cost of cultivation and making rice cultivation more economical.





Field view DRR dhan 74



Pot view DRR dhan 74



Normal and low P of field view DRR dhan 74



Grains



Decorticated grains



Milled grains



GENETIC STOCKS

Rice Germplasm Registered during July to December 2024 at ICAR-National Bureau of Plant Genetic Resources, New Delhi

S. No.	Crop Name	Botanical Name	National Identity	Pedigree	INGR No.	Novel Unique Features
1	Rice	Oryza	IC76013	IET-9854/ (Swarnadan/	24054	Possesses resistance to brown
		sativa		veluthacheera)		plant hopper at reproductive stage
						(damage score of 2.2)
2	Rice	Oryza	IC75975	RP2068-18-2-9	24055	Novel donor for resistance to
		sativa		(Swarnadhan/		Brown plant hopper (damage
				Vellathachera)		score 2.3)
3	Rice	Oryza	IR 75870-	IR 64 x O. glaberrima//IR	24056	Novel donor for resistance to
		sativa	5-8-5-B-5-	64 in BC ₁ F ₁₁ generation		Brown plant hopper (damage
			B-HWR-15			score is 4.2) Present in the
						background of IR64
4	Rice	Oryza	IR73382-	IR 64 x O. rufipogon acc.	24057	Novel donor for resistance to
		sativa	80-9-3-13-	106412//IR 64 in BC ₁ F ₁₁		Brown plant hopper (damage
			2-2-1-3-B-HWR-16	generation		score <3) Present in the
						background of IR64
5	Rice	Oryza	RP 6837- RMS- ISMA	Improved Samba Mahsuri	24058	Novel donor for resistance to
		sativa	13	(RP Bio 226)*4/RP2068-		Brown planthopper (damage
				18-3-5 in BC ₃ F ₈		score <3) Present in the
				generation		background of popular variety
						Improved Samba Mahsuri.

Source: Member Secretary, PGRC (Plant Germplasm Registration Committee), ICAR-NBPGR, New Delhi

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Scope: Journal of Rice Research is a channel for publication of full length papers covering results of original research, invited critical reviews or interpretative articles related to all areas of rice science, rice based crop systems and rice crop management. The journal also publishes short communications, book reviews and letters to the editor.

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Submission to the journal must be reports of original research of at least two crop seasons and must not be previously published or simultaneously submitted to any other scientific or technical journal. At least one of the authors (in case of joint authorship) should be member of the Society for Advancement of Rice Research (SARR) and not in arrears of subscription. Authors of invited articles are exempted from this.

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Title should give a clear idea what the articles is about. It should be brief and informative (12-15 words).

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Results and Discussion should be supported by sound scientifically analysed data along with explanatory text with relevant tables and figures.

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Research papers

- 1. Durvasula V. Seshu. 2017. Networking a Pivotal Strategy for Rice Genetic Improvement. Journal of Rice Research, 10: 1-8.
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Book

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Figures: Photographs and drawings for graphs and charts should be prepared with good contrast of dark and light. Figure caption should be brief specifying the crop or soil, major variables presented and year. Give careful attention to the width of lines and size, and clarity of type and symbols.

Tables: Tables are used for reporting extensive numerical data in an organized manner and statistically analyzed. They should be self explanatory. Prepare tables with the word-processing tables feature and tabs or graphics boxes should not be used. Table head should be brief but complete and self contained. Define all variables and spell out all the abbreviations. An exponential expression (eg. x 10³) in the unit's line is often needed to keep length of the data reasonably short, and referenced with an explanatory note. Unless otherwise required, two decimal place values are suggested.

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