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Challenges Imposed By Abiotic Stresses on The Plant and Productive Traits In Rice And Strategies For Their Mitigation

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ABSTRACT

In recent years, agricultural crops, especially cereals are facing immense pressure of feeding ever increasing world population and climate induced negative impacts of abiotic stresses on their productivity. Rice is cultivated as a staple food in various parts of the world and delivers food to 50% of the populace. Its productivity also depends on and is affected by climatic factors and investigations on rice plant response to different abiotic stresses including drought, heat and salinity are reported. Rice plant is highly sensitive in terms of yield towards lower salinity levels (NaCl 3 dSm^{-1}), threshold temperature 37° C and drought occurrence for a period of 7 days. Pre-flowering vegetative, flowering and post flowering reproductive responses of rice plants were studied under adverse impact of abiotic stresses on these specific stages. Rice plant yield is compromised under these harsh climatic conditions through germination losses, impaired seedling growth, yellowing of leaves, leaves curling and wilting, delayed or early flower initiation, loss of fertilization process resulting from poor performance of anther dehiscence, pollen number, viability, germination on stigma and pollen tube formation. Further, shortening of grain filling span with deformed floral parts, infertile spikelets, reduced grain size and weight with low assimilation movement from source to sink are the plants traits which are key indicators of abiotic stress onset during grain filling period in rice. New cultivars with improved and stress tolerant traits are needed of this global warming era to withstand harsh climatic for sustainable productivity in crops. Foreseeing the future threats to food security through the agricultural system requires our breeding programs to be aligned with advanced strategies and techniques to overcome these challenges. This review covers potential threats imposed by abiotic factors to productive traits of rice plant, their physiological basis and strategies to ameliorate these effects. This work will provide scientific basis to design and amend breeding lines for researchers to improve rice crop productivity under adverse abiotic conditions.

INTRODUCTION

Our food security is facing two alarming threats in the near future for agriculture system: (i) Climate change and its associated deleterious effects of abiotic factors on crops and (ii) the high demand of agricultural commodities under ever increasing pace of world's population. Metrological data of previous years showed that global temperature has increased 0.72° C since 1950. Prediction of climate models reveals that mean temperature will continue to increase by linear trend and witness an increase of 1-4° C at end of current century (Malhi et al., 2021). Considerable temperature rise suggests that climate change has become unequivocal, impacting water availability and crating droughts, reducing agriculture productivity and food system efficiency. A rise of temperature between 0.3 - 0.7° C is expected in next decades depending upon the greenhouse gaseous effect. Abiotic stresses are increasing at a substantial rate due to changes in climate. Global temperature is increased to alarming levels and further predicted to increase in a range of 2-3° C in next 30-50 years (Begna, 2020). Additionally, extreme temperature events like sudden heat waves are likely to become more frequent, more intense and long lasting in near future than recently observed in many parts of World. These temperature waves may have temperature 5° C above the normal temperature. This condition would be more dramatic during summer season and will lead to drastic effects on crop productivity (Deryng et al., 2014). On other side, Global population is predicted to approach 9 billion by reaching 2050 and scarcity of resources especially agricultural production due to losses induced by abiotic stress under rapid climate change scenario. Many countries of the world are facing drastic climatic changes including unpredictable swings in winter and summers, floods, prolonged droughts, increase in temperature and heavy rains.

Thus, production food for producing a higher amount of quality food for the ever-increasing population is a major concern today. It is expected that about 70% food is required to fulfill the world needs by end of 2050 (Jolánkai et al., 2019). Recent FAO reports are showing that cereal crops are declining for yield enhancement. The yield performance of these domesticated plant species depends entirely upon its genetics and the environment in which it grows. Every plant species maintains its production potential at a specific minimum, maximum and optimum level of temperature and water requirements. Any change in upper or lower temperature limits will definitely reduce the performance of that specific plant species. Crop plants standing on a place cannot move from harsh conditions to suitable conditions. Thus, their growth and development are lethally affected by abiotic factors. Among other agricultural crops, rice is the most affected crop for which severe yield losses up to total crop failure is documented in recent times through onset of heat, drought, salinity and submergence conditions. In this review, we will discuss negative effects posed by abiotic stress conditions on productive traits of rice plant and also strategies to cope these effects (Radha et al., 2023).

ONSET OF ABIOTIC STRESS AND PLANT RESPONSE

Rice is staple food predominantly in Asia and Africa it feeds a big portion of world population (about 3.5 billion people). Abiotic stresses have reduced rice production worldwide by impeding rice plant through morphological, physiological, biochemical and molecular changes which ultimately result in arresting of morphological and yield attributing traits. Plant responses to abiotic stresses are very complex, varying with the frequency, duration and intensity of stress. Furthermore, it also depends upon the plant and organ type. For example, wheat (*Triticum aestivum* L.) is sensitive to heat stress (threshold temperature = 26° C) than rice and maize (threshold temperature = 37° C and 38° C respectively) (Buti et al., 2019). Similarly, pollen grains of maize plant are more sensitive to ovules. At exposure to extreme stress conditions of heat and drought, cell death or cellular alterations may occur within few minutes, leading to disastrous collapse of cellular organization. However, at moderate levels of stress for long duration, only cellular injuries or cell death occurs. Abiotic stresses have substantial effects on all growth stages i.e. germination, developmental, reproductive and finally, grain yield (Bheemanahalli et al., 2022). Direct effects are denaturation of protein and plasma membranes and high fluidity of plasma lipids. Indirectly they inactivate enzymes, degrade structural and functional proteins, and inhibit protein synthesis and loss of membranes structure. Such detrimental effects cause production of toxic compounds, growth inhibition, hormonal imbalance, disturbance in ion exchange, starvation and reactive oxygen species (Somero, 2020). Therefore, exposure to higher stress intensities in crop plants is crucial for food security. Plants are endowed with different regulatory mechanisms and physiological adjustments i.e. expression and regulation of stress responsive genes, numerous physiological and biochemical adaptations (change in stomatal aperture, accumulation of compatible osmolytes, maintenance of membranes integrity, increased production of antioxidants and decreased ROS damage etc.), antioxidant defense and so on (Fahad et al., 2017). Immediately after the exposure to abiotic stresses, stress signals are assessed and production of certain chemicals like reactive oxygen species (ROS), Ca^{2+}, enzymatic up regulation of certain proteins (mitogen activate protein kinases MAPK and calcium dependent protein kinases CDPK) occur. Such stress signals then triggers a downstream signaling processes to alter the expression of genes and to activate responsive mechanisms for stress and to reestablishing homeostasis (Xiong & Zhu, 2001).

YIELD LOSSES IN RICE UNDER ABIOTIC STRESSES

Rice is facing a potential loss of 3.2 to 6.2 % decrease of yield with increase of every 1 °C in future. Rice area under warming regions is expected to be 16% by the end of 2030 and it would expand up to 27% by 2050 (P. Wang et al., 2014). The rice plant holds 50 to 90% of its fresh weight owing to water presence and about 60 - 90% is located inside the cell for its proper functioning and other in cell wall. Water availability is going to be the most critical and burning issue for the agriculture system as it requires 70% of global withdrawals for its working. About 20% of world land is cultivated through irrigation which yielded almost 40% of global food (Mahindawansha et al., 2018). Water scarcity is caused by various factors like heat driven high evaporative demand, low water retention by soil particles, decline of underground water level and inadequate precipitation. More than 24% yield losses were reported due to impaired growth under drought conditions in rice. Rice is highly sensitive plant among the cereals and showed repressed growth and reduced yield performance at initial salinity level (3 dSm^{-1}) (Z. Y. Feng et al., 2021). When Rice crop is grown on saline soil then yield reduction percentage ranges from 12 to 50% for salinity level from 3 to 6 dSm^{-1} respectively. Earlier studies reported that about 4.03 billion people are under the effect of soil salinity for agricultural commodities and this number is expected to climb at 5.02 billion of the world population by the end of 2050 (Zeng & Shannon, 2000). Estimated saline or sodic area is more than 1000 million hectares from which 25 to 30 % is totally barren land. The rise in sea level, sea water intrusion and accumulation of salts in underground water are the driving factors for increase in sodic or saline soils (Wong et al., 2010).

STRESS EFFECTS ON DIFFERENT PRODUCTIVE TRAITS OF RICE

Seed germination loss, impaired seedling growth, yellowing of leaves, leave curling and wilting are reported for stress established (> 40 °C) at early and pre flowering stages in rice plant. This leads to reduction in tillering capacity, leaf area, photosynthetic potential and plant height which ultimately cause a loss of total biomass during vegetative stages (Li et al., 2023). Yield reduction up to 86% is reported for rice plants when exposed to high temperature (> 37 °C) for 15 days at tillering stage. *Japonica* cultivars are more sensitive under heat stress than *indica* type rice during vegetative stages. Similarly, 21-50% yield losses in rice crop were determined for moderate to severe draught stress arrival during vegetative stages (Shi et al., 2016). Flower initiation, formation and fertilization got disturbed when rice plant faced a spell of heat stress pre or at flowering stages. The losses of stress onset at these stages are more than vegetative phase due to deformed floral parts and more susceptibility of spikelet formation, pollination, pollen viability and stigma receptivity (Y. Wang et al., 2019). At high temperature, decrease of 66% in spikelet number, 48% in seed establishment, 78% in pollen viability and complete spikelet infertility was observed in rice plants. The failure to pollination process resulted from poor anther dehiscence, pollen number, viability, germination on stigma and pollen tube formation under hot climate casus such severe losses. About 51 to 90% yield reduction is reported in different studies when rice plant was exposed to droughts spells during flowering stages (Mukamuhirwa et al., 2019). Seed formation, development, filling and assimilation movement is reduced under heat stress onset at post flowering period. Relative reduction in grain filling period up to 37%, sink size and grain weight (24-39%) with poor quality grain formation having high chalky combined percentage (92%) and lower amylose contents (16%) were reported through heat onset during grain filling period (Sandhu et al., 2021).

PHYSIOLOGICAL BASIS FOR STRESS EFFECTS IN RICE PLANT

PHOTOSYNTHESIS

Photosynthesis is thought to be the most sensitive physiological process and any alteration in its attribute serves as a stress indicator due to its close association with growth. It decreases with reversible effects under moderate stress while irreversible damage under acute stress conditions could occur. Any change in photosynthesis apparatus or process can restrict plant growth under abiotic stresses. Thylakoid membrane is thought to be more sensitive than carbon metabolisms of the stroma. Several alterations occur in chloroplast as a result of stress onset including changes in structure of thylakoid, swelling and loss in grana stacking (Nouri et al., 2015). This may be due to changes induced in thylakoids membranes of chloroplast. On the other side, RUBISCO activities are also reported to decrease under heat conditions and complete inactivation at higher temperatures, resultantly inhibiting photosynthesis. However, PSI are stable in thermos due to activation of cyclic electron pathway that contribute proton gradient. Furthermore, it is also observed that performance of Photosystem I get reduced while oxidation of PSII increases due to stress onset (Lal et al., 2021). Abiotic stress drastically effects leaf water contents (LWCs), intercellular CO_2 concentration and stomatal conductance (C). One of the major reasons for impaired photosynthesis is the closure of stomata due to change in its aperture. The main reasons of reduced photosynthesis or loss of photosynthetic activity is due to alteration in membrane integrity increased costs of respiration maintenance and reduced radiation use efficiency (Cechin et al., 2006). Gradual increase in leaf temperature over 38 ° C is reported to be linked with inhibition of net photosynthetic rate (Pn) in rice and maize. Studies revealed that high temperature (45 ° C) led to permanent damage in the photosynthetic system with no recovery when exposed to 96 h. Higher photosynthetic rates were reported for the cereal crops grown under optimal condition than the stress environment. Furthermore, it could also be affected by the inhibition of root growth which reduces leaf water potential and water-absorbing of leaf and stomatal conductance. Stress effects were also observed to have great influence on the synthesis of starch or sucrose by impairing the activities of ADP glucose pyro phosphorylase, invertase and sucrose phosphate synthase. Additionally, poor endosperm development due to reduced endospermic cell division could be the possible reason for lower reduced grain sugar contents (Al-Khatib & Paulsen, 1999). Growth rate and seed weight are also affected by limited endosperm capacity and reduced starch deposition under acute levels of abiotic stresses which leads to lower grain yield in rice. Inhibition of photosynthesis decreases food supply to grain, thus leading to decreasing kernel weight and grain yield in rice. Net photosynthetic rate is not enough under stress establishment to meet the demand of filling grains for assimilates because some assimilate started to be utilized in vegetative plant parts (Thitisaksakul et al., 2012).

ASSIMILATE PARTITIONING

Impaired photosynthetic rate causes reduced assimilation capacity which is primarily associated with reduction in grain yield. A low to intermediate stress level could reduce the source and sink activities resulting in inhibited or severely reduced plant growth, harvest index and economic yield. Studies revealed that relative growth rate (RGR) decreases under acute conditions and resulted in reduction of net assimilation rate, altered stability of membranes, enhanced respiration maintenance costs and reduced radiation use efficiency (Yamori et al., 2016). Assimilate mobilization towards rice panicle is reported to decrease under elevated stress levels which ultimately affect the harvest index. This shows the importance of source-sink relationship under stress onset. Furthermore, high temperature (33-40 °C) also decreased the light capture capability of plant, reduce biomass and harvest index which is direct directly involved in reduction in panicle yield of rice plant (Wu et al., 2016). Grain development rate during grain filling period is the main determinant of grain weight. The reduction in growth rate, shoot dry weight, root density and net assimilation causes 79 to 95% reduction in grain dry weight (W. Yang et al., 2008). High temperature reduces final grain weight due to less carbohydrate reserves of the stem and high rate of loss of water from grains. Declined in endospermic cell division, starch synthesis and dry matter accumulation leads to panicle abortion. Cell division and amyloplast replication were reported to get damaged due to the exposure of higher temperature which reduced the sink capacity of grain and ultimately grain yield (Kobata & Uemuki, 2004). Furthermore, pollination failure and less availability of photo assimilates are also among the main reasons for panicle abortion in rice. Stress onset shortened the grain filling period and further reduced assimilation mobility and availability in rice panicle. Grain growth rate has a linear relationship with crop growth rate during seed formation. However, differential response is observed for grain weight under moderate to high levels of stress (Ali et al., 2019). Therefore, it is expected that decline in grain yield under abiotic stresses is due to reduction of crop growth rate critically during important grain developmental stages.

MEMBRANE THERMO-STABILITY

Biological membranes are the most sensitive parts of plant cell. Under stress, the rupturing of structure of these membranes occurs and their associated functions get inhibited. Such change increase membranes permeability and allows the solute leakage. When the integrity of these membranes gets lost due to high temperature it results in the increased loss of electrolytes to the outer environment. Increase in the solute leakage is the indicator of decreased cell membrane thermo-stability (CMT) and it is an indirect indicator of stress tolerance in field crops including rice. The plasma membranes of mature leaves showed greater sensitive to heat injuries than young developing leaves (Mohammed & Tarpley, 2009). The loss of production is mainly due to the inhibition of assimilation capacity and it serves as a cause of reduction in net photosynthetic rate due to extreme negative effects of higher temperature on membrane stability and higher respiration maintenance costs. However, increased levels of certain membrane components i.e. phosphatidic acid in tolerant plants suggested vitality of the components in conferring stress tolerance (Kim & Portis, 2005). Some Osmo protectants like glycine betaine also provide a source of protecting cellular membranes form the adverse effects. The stability of these cell membranes plays a very important role in coping with drastic effects of stress and hence provides a mechanism of creating stress tolerance in rice. The very first impact of heat is on membrane fluid and it serves as the point of perception (Farooq et al., 2008).

HORMONAL BALANCE

Plant's ability to assess and cope to hostile environmental conditions through different stress tolerance mechanisms made their way to stress management. Abiotic stress is reported to have adverse effects on biosynthesis, compartmentalization and hormonal homeostasis, Imbalance in the hormonal profile of grains could leads towards disrupted development. Abscisic acid (ABA), Ethylene (C_2H_4), Cytokinin and gibberellin A_3 are the most important hormones that are involved in the physiological regulation under stress by acting as signaling molecules. Among these, ABA is the most important hormone that is reported to be linked with thermos tolerance via altering the gene expression and regulation and inducing several HSPs (Hsp 70 and Hsp101) (Zinta et al., 2016). Similarly, higher levels of cytokinin were also detected in stress tolerant germplasm during grain development. Whereas activity of certain enzymes like zeatin or zeatin riboside get reduced in stress affected grains. Ethylene acts as multifunctional Phyto-hormone that governs and regulates the development and senescence of leaves, flowers, and fruits depending upon the ethylene concentration, time of application and plant species. In cereals, the highest production is at the top and lowest in central parts, suggesting its contribution in assimilating partitioning and grain filling. Salicylic acid is linked with stress responses in plants. It helps in binding of heat shock elements to the promoter of genes. Salicylic acid based heat tolerance can be induced in plants and it involves antioxidant mechanisms and Ca^{2+} homeostasis (Peleg et al., 2011).

STRATEGIES FOR ABIOTIC STRESSES IN RICE

Global rice productivity is threatened by heat, cold, drought, and abiotic factors including salinity and salt (Jagadish et al., 2012) . Development of abiotic stress resistance in rice plays a significant impact on food security, thus give rice crop a crucial importance as a staple grain (Uyeh et al., 2021). Despite the constitution of high yielding rice verities with improved stress tolerance, traditional breeding has not reached its full potential because of complex genetic manipulations of yield and its attributing characters. Biotechnology advancements such as molecular breeding and genetic engineering has improved the process of producing crops that can withstand abiotic stress (Villalobos-López et al., 2022). Furthermore, new genes identification and quantitative trait loci (QTLs) connected to the abiotic stress response have improved breeding and transgenic techniques targeted at building stress tolerance. The goal of introducing several stress-related genes into rice cereals is to increase the crops' ability to withstand abiotic stressors. A variety of transgenic rice strains with increased resilience to abiotic stressors have been developed and assessed (Raina et al., 2020). However, the use of contemporary reproductive technologies and procedures cannot be entirely disregarded. More effectively producing genotypes tolerant to abiotic stress may be achievable through the use of an integrated, comprehensive breeding strategy (Razzaq et al., 2021).

CONVENTIONAL BREEDING TECHNIQUES

Conventional plant breeding deals with increasing the number of beneficial alleles that a plant possesses in its genome that aids stress tolerance (Pérez-Méndez et al., 2021). Development of stress-resilient rice cultivars is well documented by utilizing germplasm collections of crops that flourish under diverse climatic conditions (Roy et al., 2023). By using the pedigree method, segregating generations having high heritability of gene-driven traits are used to initiate the selection process for desirable plants. But only when a line becomes homozygous can selection be made for the agronomic attributes that are likely to be inherited by offspring, like abiotic stress tolerance controlled by several alleles (Withanawasam et al., 2022). One crucial selection criterion is screening under naturally harsh or experimentally produced stress conditions in order to find stress-tolerant recombinants. As a result, breeding for abiotic stress resistance to select elite stress-tolerant lines takes five to ten years which is quite unjustifiable to meet the present-day food security. Therefore, keeping in view the importance of conventional breeding, plant breeders are opting for a more balanced and robust integration and conventional and modern breeding techniques and tools to develop a-biotic stress resilience in rice (Rahman et al., 2021).

MOLECULAR BREEDING STRATEGIES

Abiotic stressors like temperature; salinity, submersion / flooding, and drought are becoming more prevalent (Chaudhry and Sidhu, 2022). Consequently, in all crop breeding initiatives, crop quality and productivity maintenance has taken equal precedence over the development of high-yielding cultivars (Mohidem et al., 2022). Despite its vulnerability to unfavorable environmental conditions such flooding, salinity, drought, and submersion, rice continues to be an essential agricultural product in ensuring the world's food security. Rice breeders are currently more focused on creating abiotic stress resilient varieties (Dar et al., 2021). Commonly used technique in variety generation is reconstitution of elite varieties via the introgression of more than one gene and quantitative trait loci that confer resistance to abiotic stress via trait-linked or functional markers (Nair and Shylaraj, 2021). Likewise, with the identification and utilization of Sub1A and SKC1, rice was the first crop to widely use precision breeding methodology for stress tolerance towards abiotic stresses (Haque et al., 2021). New advances in the genomic analysis of rice are encouraging for the production of tolerant cultivars. Deep advancements in the fields of molecular genetics, structural and functional genomics, plant physiology, biochemistry, and allied fields have demonstrated the ability to identify complex mechanisms behind desirable traits. These insights have been deployed to create breeding products for rice that combine different abiotic stress tolerances. These developments speed up the process of developing and implementing breeding strategies for improving germplasm. The discovery and cloning of the SUB1A gene made it easier to introduce into several high-yielding cultivars currently available in South and Southeast Asian countries. Vital progress was made in the mapping of significant quantitative trait loci (QTLs), including SalTol. The introduction of SalTol QTL into established cultivars is facilitated in Philippines, Bangladesh, Vietnam and India. QTLs for drought tolerance are found by combining selection and genomics techniques (Krishnamurthy et al., 2020). The use of accurately mapped quantitative trait loci (QTLs) controlling abiotic stresses resistance as the primary characteristics for marker-assisted selection has increased the potential of rice production in rainfed lowland areas. Rice breeders are compelled by climate change to combine multiple abiotic traits (salinity and submergence, salinity and drought, salinity and drought, and salinity-submergence-drought) into a single genotype as these variables coexist throughout a single cropping season (Dwiningsih et al., 2020). Rice scientists are now using modern biotechnological methods, understandings of genetic mechanisms, and sources of tolerance cultivars that are resistant to climate change and beneficial to farmers.

MARKER-ASSISTED SELECTION (MAS)

In modern agriculture, traditional methods of rice breeding are insufficient to feed the world's growing population in a sustainable manner. Modern approaches to breeding and genetics in rice have emerged, such as gene pyramiding, as a result of developments in molecular techniques, such as marker-assisted selection (MAS) and the development of DNA markers that show strong associations with target genes or quantitative trait loci (QTLs) on rice chromosomes (Ye et al., 2022). The overexpression of numerous genes that give resistance to a wide range of biotic and abiotic stimuli is allowed by "gene pyramiding," which is the process of merging two or more genes from distinct parents into a single genotype (Muthu et al., 2020). A deliberate recombination of back-crossing and marker-assisted pedigree selection has been proved to be a paradigm shift in reducing the No. of breeding generations involved in gene pyramiding process (Vanitha et al., 2023). Factors influencing pyramiding include the amount of transferred genes, the separation between genes and surrounding markers, the number of selected populations in each breeding generation, the properties of genes and germplasms, and the ability of breeders to identify target genes. In the next few decades, stress-tolerant rice varieties will be developed as a result of modern breeding techniques such as marker-assisted backcrossing, which have improved the accuracy and reliability of gene pyramiding. Every year, rain fed lowlands areas of 42 million hectares cultivating rice are affected by drought, which results in yield reduction by 13-35%. Most rice cultivars are vulnerable to drought, which makes the difference between expected and actual yields. Because the mechanisms affecting drought are complex, traditional efforts to increase rice genetic variability against drought stress have shown limited results.

GENOME WIDE ASSOCIATION STUDIES (GWAS)

Genome wide association studies (GWAS) is one of the most widely used tools to identify stress resilient QTLs in rice (Lv et al., 2022). Several case studies were reported by various groups on usage of GWAS to identify salt-tolerant genes. Kumar et al. (2015) genotyped 220 rice types using the high-throughput test using specialized gene-based SNP arrays. Genes showing a stress response were included among the 6000 SNPs on the gene-based SNP array. Twelve unique traits are grouped together in phenotypic datasets and are linked to stress. A study was conducted to investigate the relationship between 12 characteristics and 6000 SNPs. Regarding the Na+/K+ ratio, twenty out of the forty-four SNPs linked to different stressors were significantly localized. In addition, genomic areas on chromosomes 1, 4, 6, and 7 were linked to quantitative trait loci (QTLs) encoding genes or alleles for salt tolerance. Several studies are well documented in identifying the heat-tolerant QTLs in rice. Lafarge et al. (2017) investigated the effects of temperature sensitivity during anthesis on 167 rice varieties' secondary features, such as the fertilization process as well as spikelet sterility (SPKST). The datasets were ranked by allocating one marker at every 29 kilobases. Three different methods were used to conduct GWAS: single-marker regression, haplotype regression and fitting of all markers. Results showed a significant correlation between fourteen loci and SPKST. The loci were shown to control heat shock proteins, wall-associated kinases, cell division, gametophyte development, sense of abiotic stress, and plant response. Moreover, correlations with secondary attributes were found. 3,000 rice genomes include SPKST favorable alleles, according to locus analysis. Furthermore, certain Taiwanese and Indian cultivars, such as N22, are believed to have heat resilience. In a nutshell, strategies to develop abiotic-stress-resilient rice genotypes are well documented. However, an integrated utilization of these approaches could create new vistas in stress-resilient rice breeding.

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