Combined Drought and Heat Stress in Rice: Responses, Phenotyping and Strategies to Improve Tolerance

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Abstract: Simultaneous occurrence of drought and heat stress will have significant negative impact on rice yield, especially under upland conditions. The projected increase in global temperatures and reduced precipitation will increase the frequency of occurrence and intensity of these stresses, threatening rice production. Despite recognizing the importance of combined stress in rice, the knowledge generated in this area is very limited. Though complex, understanding combined stress tolerance of rice under water saving cultivation is more critical towards development of climate resilient rice cultivars. Here, we summarized the effects of combined stress on rice physiology with more emphasis on reproductive stage. Omics responses, phenotyping and physiology challenges and potential strategies for improving combined stress tolerance in rice are also discussed.

Key words: rice; drought stress; heat stress; combined stress; flowering; grain filling; omics response; phenotyping; genetic enhancement

Agriculture worldwide is predicted to face the drastic effects of climate change in the near future with increased frequency and magnitude of hot and dry spells (Gornall et al, 2010). This will cause yield losses as high as 60% depending on the crop, developmental stage, as well as type, severity and duration of the stress (Jedmowski et al, 2015). Under tropical field conditions, drought and heat stress frequently co-occur. Drought stress and low atmospheric humidity will increase the vapour pressure deficit (VPD) causing rapid water loss from the soil as well as the plants. Plants generally close stomata in response to increasing VPD as a conservative strategy, but often fail to meet the transpirational demand to cool off the surface, resulting in increased tissue and canopy temperatures. Similarly, high temperature can also directly create drought stress due to rapid evapotranspiration, resulting in a sudden decrease in soil and plant tissue water status. This direct effect of heat is exacerbated under water limited conditions (Prasad et al, 2008). Therefore, under field conditions, drought and heat interact to create a more formidable combined stress (Fig. 1). Plants exposed to a combination of drought and heat stress exhibit unique physiological, metabolic and molecular responses that are different from individual stress (Silva et al, 2010; Jin et al, 2016). Therefore, specific strategies for combined stress are being developed towards improving crop tolerance (Pandey et al, 2015).

Rice is a major crop feeding more than half of the world’s population. Incidentally, it is a high-water demanding crop, requiring nearly 3 000–5 000 L of fresh water to produce 1 kg of grains, which is the highest among major cereals (Bouman, 2009). It is predicted that by 2025, nearly 15–20 million hectares of rice growing area, especially in the tropics and low
latitudes, will suffer from water scarcity (Ben et al, 2017). The reduced water availability and ever-increasing demand for fresh water are forcing farmers to adopt water saving practices such as alternate wetting and drying, dry-direct seeding and semi-irrigated aerobic cultivation (Korres et al, 2017). These practices will often expose rice to both drought and heat stresses, threatening rice production (Wassmann et al, 2009). Drought and heat stress responses in rice have mostly been studied individually with considerable success (Jagadish et al, 2011; Kumar et al, 2015; Kumar A et al, 2017; Oladosu et al, 2019; Wang et al, 2019). Here, we reviewed the effects of combined drought and heat stress on physiology, omics responses, phenotyping approaches and strategies to improve combined stress tolerance in rice.

**Effects of combined drought and heat stress on rice physiology**

Combined stress might cause pandemonium on photosynthetic carbon metabolism and mitochondrial respiration at the seedling and vegetative stages in rice. So far, there is no evidence to show that short-term combined stress at the vegetative stage will contribute to significant changes in rice growth and yield. However, long-term stress has been shown to significantly affect growth, leaf gas-exchange and water use efficiency (WUE), with more reduction in total biomass under combined stress compared to individual stress (Perdomo et al, 2015, 2016). The effects of combined stress on rice physiology are illustrated in Fig. 2. Combined stress can limit photosynthesis, mostly due to reduced stomatal conductance, and hence reduce biomass. The mitochondrial respiration at pre-dawn, maximum velocity of RuBisCO carboxylation and maximum rate of electron transport are highly influenced by temperature than drought (Perdomo et al, 2016). This is also evident from the higher resilience of photosynthetic biochemistry under drought than under high temperatures. Therefore, photosynthetic carbon metabolism seems to be strongly temperature dependent during long-term (acclimated) combined stress. Under drought, increased WUE is often associated with reduced transpiration and a corresponding decrease in biomass (Galmes et al, 2011). However, under combined stress, WUE is positively correlated with growth, though this needs further investigation at the whole plant level (Perdomo et al, 2015). Cooler canopies even under stress are always associated with higher yields (Zhang et al, 2007). This is achieved through higher transpiration which is counterproductive under limited root and water. Besides, selection for reduced transpiration and transpiration efficiency will result in smaller plants and hence reduced biomass (Blum, 2009). This will aggravate when drought and heat stress co-occur. Therefore, to keep a balance between cooling and carbon assimilation, genotypes with tight stomatal regulation should be identified (Vadez et al, 2014). This is possible through developmental stage, diurnal and VPD driven coordinated regulation of stomata. Higher density and smaller stomata might help in achieving this as it provides plants more options to regulate stomatal movement under stressful conditions (Cuine et al, 2019). Diurnal regulation of photosynthesis might further help in alleviating combined stress effects with active photosynthesis during cooler
periods. Nocturnal transpiration becomes critical from this perspective as it helps in sustaining sugar transport to the reproductive structures and filling grain, enhancing next day carbon assimilation (de Dios et al, 2016; Sadok, 2016) that is crucial under combined stress. However, the amount of water lost through nocturnal transpiration must be optimized so as to maximize WUE.

Rice is more sensitive to drought, heat and their combination at reproductive stage, more specifically at flowering. High temperatures in most rice-growing areas of Asia coincide with drought stress at flowering to early grain filling stages (Wassmann et al, 2009; Kumar N et al, 2017). In these regions, the temperature during flowering ranges from 36 °C to 40 °C, which is above the optimum. With changing climate, several predictive models suggest an increase in frequency of high temperatures towards the end of crop growing season when the irrigation water availability and/or precipitation is very low, leading to severe yield loss and hence posing serious concerns on future rice production in these regions (Timsina and Humphreys, 2006; Yoshida et al, 2015). Combined stress at flowering has been shown to significantly affect peduncle length, anther dehiscence, pollen number, pollen germination and spikelet fertility in rice (Li et al, 2015). Nagina 22 (N22), a drought and heat-tolerant cultivar, grows better both under heat and combined stress compared to Moroberekan, a drought tolerant but heat-sensitive cultivar which has no pollen, resulting in absolute sterility under heat and combined stress. Better performance of N22 might be due to good pollen viability and germination percentage under all conditions (Jagadish et al, 2010). Similar observations were made in another study involving five rice cultivars (two N22 accessions, IR64, Apo and Moroberekan) (Rang et al, 2011). In terms of spikelet fertility, heat stress dominantly reduces fertility when drought and heat stress co-occurred. Interestingly, even with higher anther dehiscence, the pollen count on the stigma reduces in Moroberekan. This could be due to asynchrony in pollen shedding and the stigma receptivity in this cultivar at high temperatures (Jagadish et al, 2010). There is also carryover effect of heat and combined stress on spikelet sterility even after the stress is relieved, suggesting that stress even before anthesis might still affect spikelet fertility. With rice being a monoecious crop, grain set depends on the fertilization success. Therefore, synchrony in anther dehiscence and stigma receptivity, which is not observed in Moroberekan, is critical not only under stress but also under normal conditions. Increased panicle temperatures along with reduced panicle conductance under combined stress was observed in three rice cultivars (N22, Dular and Anjali) under field conditions (Lawas et al, 2018). Canopy-air temperature increase was also observed under drought treatment altering the canopy microclimate. Significant reductions in grain yield, number of spikelets per unit area, seed-setting rate and harvest index have been reported in all cultivars under combined stress. Combined stress at grain filling affects yield in all the three cultivars, but is less significant compared to stress at flowering. However, stress at grain filling has greater effect on chalkiness compared to that at flowering, suggesting the effect of combined stress even at the grain filling stage (Lawas et al, 2018). Better yield and physiological performance of N22 under combined stress at panicle initiation and anthesis have also been shown by Radhakrishna et al (2018). Apart from daytime temperatures, nighttime temperatures negatively affect rice yields especially at grain filling. In fact, rapid increase in nighttime temperatures than daytime temperatures has been observed in the last few decades. With every 1 °C increase in nighttime temperatures above the optimal (24 °C), 10% reduction in yield and biomass was observed (Peng et al, 2004; Welch et al, 2010). Starch accumulation during grain filling stage will happen during the nighttime, and high nighttime temperatures will severely perturb the activities of enzymes involved in starch metabolism (Bahuguna et al, 2018). When high nighttime temperatures are associated with drought, it will affect carbon metabolism during grain filling causing high chalkiness (Lanning et al, 2011).

It appears that not all drought-tolerant cultivars are tolerant to heat stress by default at sensitive stages. Heat stress seems to have a dominant effect especially at the reproductive stage. Therefore, empirical screening for heat tolerance at the reproductive stage and testing the heat tolerant genotypes under combined stress would hasten the process of improving combined stress tolerance in rice. N22 is the only known combined stress tolerant cultivar so far identified. Tolerance of N22 is due to better anther dehiscence, more pollen and higher pollen viability, resulting in better seed set and yield. Even at the vegetative stage, N22 maintains better photosynthesis with minimal damage to photochemistry translating into better combined stress tolerance and hence superior yield.
Similar to individual stress, source-sink interactions determine plant tolerance to combined stress (Rosa et al, 2009). Remobilization of assimilates to reproductive structures during stress depends on sink strength (Bihmidine et al, 2013). During anthesis, sink strength is higher for pollen than pistils with ovary having more sink strength after fertilization and during grain filling (Zinselmeyer et al, 1995; de Storme and Geelen, 2014). Hence, sucrose requirement for pollen is higher than pistils at anthesis which is highly affected by combined stress (Li et al, 2015; Perdomo et al, 2015, 2016). Besides, the physical location might protect ovaries from severe damage of combined stress when the tissue and canopy temperatures increase. Though the sensitivity of pollen to combined stress has been proven in rice (Rang et al, 2011; Jagadish et al, 2014), the role of stigmatic surface during pollination and fertilization process cannot be ignored. Therefore, strategies to enhance combined stress tolerance should include traits that improve resource accumulation, mobilization and partitioning from vegetative to reproductive structures during stress conditions. Combined stress will also severely reduce biomass and delays flowering leading to significantly reduced yields compared to individual stresses, suggesting the importance of growing degree days (Parent and Tardieu, 2012).

**Omics responses of rice to combined drought and heat stress**

Like any other stress, combined drought and heat stress has been shown to evoke differential transcriptome, metabolome and proteome responses in plants. Though several responses under combined stress are shared with drought, heat or both, some of them are specific to combined stress (Rizhsky et al, 2002, 2004; Lawas et al, 2018). Nevertheless, the numbers of omics-based studies under combined stress are very limited in rice.

Pollinated pistils from N22 and Moroberekan showed more differentially expressed genes (DEGs) under combined stress than one-day heat stress compared to control (Li et al, 2015). In N22, the number of DEGs are much lesser in combined stress treated anthers compared to those in Moroberekan, suggesting better anther physiology in N22 due to constitutive regulation of several genes required for stress tolerance. On the contrary, a greater number of DEGs in combined stress treated pollinated pistils of N22 compared to Moroberekan suggests the possible active fertilization process in N22 even under stress. In one-day combined stress treated pollen, 716 and 82 DEGs specific to combined stress were found in Moroberekan and N22, respectively. Similarly, 767 and 494 DEGs specific to three-day combined stress treated pollen were found in Moroberekan and N22, respectively. These findings confirm that some transcriptome responses are specific to combined stress as well, in addition to shared responses. In general, genes related to heat shock transcription factors and heat shock proteins which play important roles in cellular tolerance through their chaperone activities are upregulated by multi-fold in N22 under combined stress compared to Moroberekan, suggesting the intrinsic tolerance ability of N22 rather than escape mechanism. Besides, differential regulation of a greater number of genes in pollinated pistils of N22 can be attributed to a greater number of germinated pollens per stigma initiating the fertilization process than Moroberekan, wherein, none or a very few viable pollens were found on stigma leading to no fertilization process. In addition, several genes related to sugar metabolism are differentially regulated under combined stress, including MST8 (sugar transporter) and INV2 (cell wall invertase). Moroberekan shows increased expression of carbon starved anthers (CSA) gene, suggesting the greater sink strength and sucrose supply to reproductive structures in N22 compared to Moroberekan. Further, there are 4 and 31 metabolites up- and down-regulated in N22 compared to Moroberekan under combined stress. Nine potential marker metabolites responsive to combined stress are identified, which are mainly involved in regulation of sugar metabolism and tricarboxylic acid cycle, and three of these are constitutively expressed in N22 under the control condition. In a recent study, metabolite profiling of three rice cultivars (N22, Dular and Anjali) at flowering and early grain filling stages in flag leaves, flowering spikelets, developing seeds treated to mild/severe-drought and re-watering under natural summer temperatures shows differential expression of metabolites between organs, cultivars and tissues (Lawas et al, 2019). Glucose-6-phosphate, arabitol, succinic acid and pyruvic acid in spikelets, glutamic acid, vanillic acid and arbutin in flag leaves and pyruvic acid in developing seeds are suggested to be potential metabolic markers for combined stress tolerance in rice. Arabitol, arbutin and vanillic acid may help in osmotic adjustment and oxidative damage reduction under stress (Hincha et al, 1999; Parvin et al,
2020; Schaarschmidt et al, 2020). Glucose-6-phosphate, succinic acid, pyruvic acid and glutamic acid are feeding into or intermediates of tricarboxylic acid cycle which might help in sustaining the energy requirement during the stress.

The proteomic responses of rice to combined stress have also been studied during anthesis with 29 differentially expressed protein spots identified under drought, heat, and/or combined stress (Jagadish et al, 2011). Of these, 11 spots which show 2-fold or more expression are identified as cell wall loosening proteins, extension and biosynthesis proteins such as pollen allergens, low-molecular-weight heat shock proteins (HSPs), beta expansin, soluble inorganic pyrophosphatase, putative fructokinase and two unknown proteins. All these proteins are responsive to all stresses with no protein specific to combined stress. However, six of these proteins reverted back to control levels, especially two small HSPs (16.9 and 17.4 kDa), are stronger and corresponded with their transcript abundance under combined stress.

Collectively, with these limited studies covering only a few cultivars, it is difficult to draw specific conclusions at present on the potential omics candidates for improving combined stress tolerance of rice. Besides, these studies concentrated only on identifying genes, metabolites and proteins specific to combined stress. However, there are genes, metabolites and proteins which are shared between individual stresses and, between individual and combined stresses. Though these represent generic responses of plants to a stress, they might play critical role in combined stress tolerance. Therefore, strategies to mitigate combined stress should include the responses common to all stresses as well as specific to combined stress (Fig. 3) (Pandey et al, 2015).

**Phenotyping for combined drought and heat stress in rice**

It is critical to consider prevailing stress intensities, genotypes grown and developmental stages affected by combined stress in the target environment while phenotyping for combined stress. Though field-based drought stress imposition using managed drought environments such as rainout shelters has been in practice for a while, limited effort towards understanding the combined stress responses could be attributed to limited or non-availability of heating systems to impose heat and combined stress on a large scale (Rehmani et al, 2011; Chiba and Terao, 2014). In most of the rice growing regions, the flowering and grain filling stages coincide with high temperatures where temperatures are more than the optimum required for maximum rice yields (Jagadish et al, 2010). The critical temperature at reproductive stage for the drought and heat tolerant cultivar N22 itself is 36.5 ºC (Jagadish et al, 2011). Temperatures similar to this can be easily attained during summer, especially when it is associated with drought stress (Bahuguna et al, 2015; Prasanth et al, 2016). Increase in soil, canopy and air temperatures have been reported in drought treated plots compared to control conditions (Lawas et al, 2018). By using rainout shelters and staggered sowing, Lawas et al (2018) and Radhakrishna et al (2018) effectively increased canopy air temperatures during critical growth stage of rice, which are similar to temperature increase obtained by heating systems (Rehmani et al, 2011; Chiba and Terao, 2014). Therefore, natural summer temperatures can be effectively used to impose and evaluate effects of combined stress in rice while taking care of the photosensitivity. After identifying the superior cultivars, laboratory experiments can be conducted to understand the genetic basis of stress tolerance with precise imposition of drought, heat and combined stress using environment-controlled chambers/greenhouses.

Stress imposition at the critical reproductive stage is important for accurate quantification of combined stress effects. Coinciding drought at critical growth stages might be difficult as there is a large genetic

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*Fig. 3. Omics approaches for understanding and improving combined drought and heat stress tolerance.*
variation for water use, which is highly influenced by VPD and phenology among cultivars. The later can be taken care to an extent with prior knowledge on flowering time of cultivars and tagging the tissues which reach specific growth stage (Lawas et al, 2018). Desired water status at specific growth stages can be achieved, irrespective of the differences in water use and VPD, using open drought phenomics platforms (Vijayaraghavareddy et al, 2020). This type of transpiration-interphased phenotyping platforms will maintain the same soil moisture status across the pots irrespective of the differences in transpiration among the genotypes. Besides, interaction of temperature and relative humidity (RH) will significantly affect rice yields. Higher RH in association with moderate temperatures cause greater negative impact than lower RH on rice yields (Weerakoon et al, 2008). Therefore, testing combined stress responses of rice cultivars in hot and dry regions or seasons will have less interference from RH. However, it is also important to develop varieties which can withstand dry and hot conditions under relatively higher RH for vulnerable regions (Weerakoon et al, 2008; Wassmann et al, 2009).

Anthesis is more sensitive to combined stress as high temperatures after pollination cause no effect on spikelet fertility (Jagadish et al, 2011). In most rice cultivars, anthesis occurs early in the morning (Julia and Dingkuhn, 2012; Bheemanahalli et al, 2017). Under field conditions, hot days are mostly associated with warmer early morning temperatures. Therefore, controlled environment experiments should consider combined stress responses with elevated early morning temperatures as well. Stamens, specifically pollens, seem to be more affected by combined stress than pistils (Jagadish et al, 2010; Li et al, 2015). The stress during booting which coincides with microsporogenesis causes lesser damage but leads to carryover effect after the stress is relieved (Lawas et al, 2018). Further, stress before heading will result in impaired panicle exertion, trapping considerable number of spikelets within the flag leaf sheath causing severe spikelet sterility (Jagadish et al, 2011; Lawas et al, 2018). Hence, genetic variation in panicle exertion under combined stress is another important trait to be studied. In addition, duration of the stress also has greater influence on cultivar responses to combined stress. Even though a few hours of combined stress is enough to cause major yield loss, more yield reductions have been shown when stress is given for a longer period (Rang et al, 2011). Therefore, it is also important to study the acclimation responses of plants to combined stress similar to natural conditions. With the advance in phenotyping technologies, real-time measurements of canopy temperatures and fluorescence are possible in the field, and it will definitely help hasten phenotyping for combined stress in rice.

**Approaches to improve combined drought and heat stress tolerance in rice**

**Breeding**

In major cereals including rice, conventional breeding efforts in the last few decades have mostly focused on increasing yield under optimal conditions with little attention to unfavourable environment. However, the yields have started plateauing even under optimal conditions, threatening food security (Grassini et al, 2013). On top of that, rice growing regions are facing increased frequency and intensity of dry and hot spells (Barnabás et al, 2008). Therefore, the present need is to develop high-yielding cultivars under stressful as well as optimal environments. Plant breeding plays a major role in understanding and improving tolerance of rice to individual drought or heat stress. Our understanding of the genotype and environmental interaction from the individual stress may not be of much help while studying the combined stress. Besides, while selecting for only yield, the gene pool required for combined stress tolerance might have been lost in the presently grown cultivars. This is supported by the fact that combined stress studies have been done with only a few cultivars and N22 emerged as the only tolerant cultivar. Therefore, systematic screening of rice germplasms and mapping populations to identify quantitative trait loci (QTLs) and introgressing the QTLs into elite cultivars are necessary. This has been shown in rice under other stress combinations. For example, marker-assisted introgression of individual drought and submergence tolerance QTLs in Swarna results in combined drought and submergence tolerance (Sandhu et al, 2019). The availability of a large number of drought QTLs and continued efforts towards identification of heat QTLs would pave the way for stacking large effect drought and heat QTLs for improved combined stress tolerance (Kobayashi et al, 2013; Shanmugavadivel et al, 2017; Prakash et al, 2019). With genome sequencing becoming cheaper and availability of whole genome sequence information for 3K rice accessions, genome-wide association study plays a critical role in dissecting the genetic basis of combined stress tolerance through identification of QTLsgenes. Further, such
QTLSgenes can be quickly integrated into the elite background through targeted breeding in association with genome editing tools without much linkage drag.

### Genetic engineering

Understanding the mechanisms and pathways regulating plant responses to combined drought and heat stress through omics approaches is critical in an endeavour to develop combined stress tolerant rice. Genetic engineering tools play vital roles in introducing these mechanisms and pathways. Large number of drought and heat stress responsive genes have been identified and transgenic approaches have been used to engineer these genes towards improving individual drought and heat stress tolerance of rice (Yu et al., 2016; Latha et al., 2017). Besides, several of these genes are responsive to both drought and heat stress. Engineering such genes has been shown to improve both drought and heat stress tolerance, when given independently. For instance, overexpression of SNAC3 transcription factor confers tolerance to individual drought and heat stress in rice through reactive oxygen species (ROS) homeostasis (Fang et al., 2015). Similar observations were made with overexpression of OsRab7 through modulation of osmolytes, ROS and stress-responsive genes (El-Esawi and Alayafi, 2019). The only gene which has been tested for combined stress tolerance in rice is OsWRKY11, expressed under OsHSP101 promoter, with transgenic plants showing enhanced tolerance at the vegetative stage (Wu et al., 2009). In addition, there is substantial transcriptome data available from rice subjected to drought and, to some extent, heat stress (Jin et al., 2013; Huang et al., 2014; Zhang et al., 2016). Shared molecular responses from these individual stresses can be explored through meta-analysis to identify genes and test them using the transgenic approach for improved tolerance to combined stress (Fig. 3). Besides, transgenic approach can be used to evaluate the role of genes such as HSPs and sugar transporters which are specifically induced under combined stress in improving stress tolerance (Li et al., 2015). Recently, omics data analysis and functional characterization of genes identified from plants exposed to individual stresses have revealed a convergence of signalling pathways for stress adaptation (Kissoudis et al., 2014; Shaik and Ramakrishna, 2014). Understanding such signalling components shared between drought and heat stress is critical in improving rice tolerance to combined stress. Knowledge from other plant species suggests ROS as converging signals involved in crosstalk between drought and heat stress (Koussevitzky et al., 2008; Fang et al., 2015; Choudhury et al., 2017; El-Esawi and Alayafi, 2019). Role of genes involved in ROS homeostasis can be tested in imparting combined stress tolerance. Systems and gene network approaches to identify genes and related physiological responses will also be helpful towards improving combined stress tolerance of rice.

### Conclusions and future perspectives

Though the effects of combined drought and heat stress on rice under field conditions are known for a long time, the effort towards understanding and improving tolerance is limited. Global concerted efforts are needed in this direction to sustain rice production to feed the growing population under changing climate. Traits of interest which can potentially contribute to combined stress tolerance in rice are shown in Fig. 4. From the physiological perspective, WUE, carbon assimilation and partitioning, and respiration are critical traits to be explored. Identifying genotypes with better mesophyll conductance and carboxylation efficiency would help in improving WUE and yield under stress. The basic instinct of plants under individual drought or heat stress is to partition more photosynthates towards root growth in anticipation of finding more water to meet the transpirational demands. Partitioning more assimilated carbon towards reproductive structures and filling grain is critical in maintaining grain yield and quality during terminal stress. Genetic variability and interaction between nighttime photo-assimilates transport and daytime photosynthesis is another area to be explored. There is a high association between availability of carbon assimilates, dark respiration and grain filling. Reduction of nearly 10% rice grain yield for every 1 °C

![Fig. 4. Potential traits for improving combined stress tolerance in rice.](https://example.com/figure4)

HSPS, Heat shock proteins; ROS, Reactive oxygen species; VPD, Vapour pressure deficit.
increase in nighttime temperatures has been suggested as against 3.2% for daytime temperatures. Therefore, it is critical to understand the effects of combined stress on carbon assimilate partitioning and respiration using realistic temperature and soil water changes under field conditions. Besides, combined stress screening should be carried out in arid and semi-arid regions through staggered sowing during early winter to coincide flowering to natural high day/night temperatures with the help of rainout shelters.

Another area, which has not been explored so far, is the phytohormonal responses of rice to combined stress. Apart from stress responses, hormones tightly regulate developmental processes especially reproduction which are highly affected by combined stress. Like other stress, enriching the basic knowledge on physiological, biochemical and molecular responses of rice to combined stress through omics approaches will help in better understanding and improving tolerance to combined stress.

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