Physiological basis of submergence tolerance in rice and implications for crop improvement

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The environments of rainfed lowland rice are highly variable both over time and location. Flash-flooding and submergence adversely affect at least 16% of the rice lands of the world (~22 m ha). In eastern India, ~13 m ha of rice lands are unfavourably affected by excess water and periodically suffer from flash-floods and complete submergence. Improvement of germplasm is likely the best option to withstand submergence and stabilize productivity in these environments. However, progress in germplasm improvement has been slow but can substantially be enhanced if the physiological and genetic bases of submergence tolerance are well understood and extent of damages over time and location is known. This review focuses on current physiological understanding of tolerance to submergence in rice with greater emphasis on floodwater environments, new genetic resources and potential of DNA marker technology for incorporating multiple traits associated with tolerance, to enhance and speed progress through breeding. Research on the aspect has been further facilitated by the recent application of chlorophyll fluorescence spectrophotometry as a rapid and non-destructive technique to screen submergence-tolerant cultivars.

Keywords: Crop improvement, physiology, rice, submergence.

IN India, the area under rice cultivation is 44.5 m ha (data of 2000–01)[1] with an annual production of 85.5 million tons and an average productivity of 1.9 t ha^{-1}. Rice is grown in a wide range of ecologies ranging from irrigated to uplands, rainfed lowland, deep water and tidal wetlands. About 29% of India's total rice area, i.e. ~13 m ha is rainfed lowland, which contributes only 19% to national rice production. In a normal year, about 4 m ha is drought-prone, while 3 m ha is favourable, another 3 m ha is medium deep waterlogged with water standing for up to 50 cm. The remaining 3 m ha is submergence or flood-prone, where plants are completely submerged for 1–2 weeks or so, resulting in partial or even complete crop failure. Rainfed lowlands constitute highly fragile ecosystems, always prone to flash-floods (submergence) with an average productivity of only 1.2 t ha^{-1} in normal years and hardly 0.5 t ha^{-1} in case of submergence. Among the 42

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biotic and abiotic stresses affecting rice production, submergence has been identified as the third most important constraint for higher rice productivity in eastern India[2], because it sometimes resulted in near total yield loss. Besides India, flooding is widespread in other South and Southeast Asian countries such as Bangladesh, Thailand, Vietnam, Myanmar and Indonesia. Suitable germplasm as well as management technologies are therefore needed to enhance and stabilize rice productivity in these areas.

The effects of submergence on rice as well as physiological base of tolerance were recently reviewed[3–7]. The present article focuses on the multifaceted problems to which rice crop is exposed during submergence. It also emphasizes various tolerant traits or mechanisms that are necessary for high and stable productivity in submergence-prone areas, such as tolerance during germination and seedling emergence, tolerance to repeated floods and fast regeneration after submergence, all of which could contribute to better and stable productivity. Due consideration is also given to the use of innovative research approaches to build on the progress achieved so far for germplasm improvement.

Inference from field studies

There are many environmental and biophysical stresses to which plants are exposed during complete submergence in the field, which include reduced movement of gases to and away from plant surfaces. Gases are known to diffuse 10,000 times slower in water than air[8]. Hence, poor plant growth and survival during submergence or waterlogging is often considered a consequence of the decreased diffusion of gases which affects plant growth and metabolism. Reduced O_2 supply limits respiration, reduced CO_2 supply limits photosynthesis and reduced ethylene diffusion away from the plant triggers chlorosis and excessive elongation of leaves of intolerant cultivars[9,10].

Measurement of the concentration of gases in floodwater during submergence gave important clues as to the causes of reduced growth and survival. Studies at NDUAT, Faizabad, eight locations in Bihar and Uttar Pradesh and by Central Rice Research Institute (CRRI), Cuttack also helped to understand the variations observed in plant survival at different locations of eastern India, when rice genotypes are exposed to submergence for similar durations. Oxy-

gen concentration was found to range between 0.0 and 0.6 mol m^{-3} (air equilibrium 0.24 mol m^{-3} at 30°C), carbon dioxide concentration varied between 0.004 and 0.020 mol m^{-3} (air equilibrium 0.01 mol m^{-3}) and pH varied[11,12] from 6.6 to 9.7. The pH was high in alkali soils of Bihar. In eastern India, submerged plants received lower irradiance due to high turbidity of floodwater and cloudiness. Light intensity even at a depth of 30 cm was almost nil due to the presence of silt in floodwater. During submergence, plant survival is greatly affected by depth of water and by its physico-chemical characteristics (oxygen and carbon dioxide concentration, pH, turbidity, temperature, etc.). Studies on survival percentage of the same set of cultivars in different locations in India as well as in Bangladesh, Thailand and Indonesia support this contention[3,12]. Plant survival under complete submergence can considerably be affected by altering the quality of the floodwater. Low floodwater pH or enhanced concentration of CO_2 is known to enhance photosynthesis under submerged condition and increase the concentration of O_2 for respiration, resulting in plant survival for longer periods[13,14]. Although the idea of manipulation of floodwater for higher photosynthesis through lowering the pH and/or providing higher concentration of CO_2 is not practical, these studies indicated that germplasm or breeding materials tolerant to submergence need to be location-specific to meet local variations of light availability, floodwater conditions, particularly turbidity and turbulence, and prevailing water temperature at the time of flooding among others.

Physiology of submergence tolerance

Submergence tolerance is a metabolic adaptation in response to anaerobiosis that enables cells to maintain their integrity so that the plant survives hypoxia without major damages. A critical evaluation of submergence-tolerant and intolerant rice cultivars revealed that seedlings of tolerant species have 30–50% more non-structural carbohydrates compared to the susceptible cultivars[15–17]. These non-structural carbohydrates are utilized during submergence to supply the required energy for growth and maintenance metabolism[18,19].

Quick regeneration following submergence is a desirable trait under frequent or prolonged flooding, as it can ensure early recovery and production of sufficient biomass for optimum productivity. The old leaves die after flooding, particularly when floodwater is turbid or when flooding is prolonged. Initiation of new leaves and their subsequent growth requires availability of non-structural carbohydrates. Cultivars that maintained more than 6% of their initial non-structural carbohydrate at the time of re-aeration were found to be capable of developing new leaves rather quickly[20,21]. Therefore, it is sufficiently reasonable to conclude that high carbohydrate status after submergence, which is the consequence of its level before submergence

and extent of turnover and consumption during submergence, is the key factor that determines the ability of plants to withstand submergence stress.

Development of submergence-tolerant cultivars has required the use of stress-specific screens, because direct evaluation of tolerance is not as simple as it might seem. Complete submergence hastens degradation of chlorophyll content in susceptible rice cultivars compared to tolerant ones[10,22], which can also be used as an indicator of submergence tolerance. A comparison between chlorophyll content and chlorophyll fluorescence parameters suggests that the latter is more sensitive to submergence[23,24]. By measuring chlorophyll fluorescence, several valuable parameters could be quantified which can clearly differentiate between sensitive and tolerant cultivars as early as 4–6 d of submergence, when even sensitive cultivars showed no signs of mortality.

In India, submergence tolerance alone is not sufficient to release cultivars for rainfed lowland ecosystems. This also applies to other countries in South and Southeast Asia, and is probably the reason that the varieties released so far for rainfed lowland lack submergence tolerance. In normal years when flooding is not too severe, farmers can get reasonable returns from these types of cultivars. However, adoption of the so-called high-yielding cultivars is meagre because of the frequent floods commonly experienced in these flood-prone areas, coupled with the sensitivity of released cultivars. Hence, farmers still use their local low-yielding landraces, which are relatively more tolerant to submergence. Adoption of alternative screening techniques such as chlorophyll fluorescence could help speed up the progress in breeding to incorporate tolerance into modern varieties, where prolonged submergence will not be required for phenotyping. This technique can also help differentiate between tolerant and sensitive breeding lines without losing the valuable high-yielding breeding materials.

Rice plants that exhibit only limited elongation during submergence often show tolerance to complete flooding. Some researchers have suggested that the ideal response to submergence is submergence tolerance (survival under water) together with some elongating ability[4,25]. This ideotype is suitable only if the water level increases and then (i) stays at that level, (ii) recedes only partly, or (iii) recedes but then rises again and stays for longer duration. However, reduced elongation under flash-flood conditions (complete submergence for 1–2 weeks) is vital for survival because elongating plants would tend to lodge as soon as the water level recedes. Further, the rationale behind classifying a genotype that exhibits limited elongation during submergence as tolerant, is that such a genotype is likely to use only a small quantity of available carbohydrate for elongation growth, leaving enough for survival during submergence[16,18,26] as well as for growth resumption after the water recedes[21]. Studies involving manipulation of elongation growth upon submergence have also

demonstrated the beneficial effects of its suppression during submergence. For example, GA application to plants 48 h before submergence resulted in more elongation during submergence, with consequent reduction in plant survival. In contrast, when GA biosynthetic inhibitor, paclobutrazol was applied, shoot elongation under submergence was reduced resulting in higher survival percentage[21,26]. Furthermore, dwarf mutants exhibiting little or no capacity for GA biosynthesis showed submergence tolerance on par with the highly tolerant landrace FR 13A, when plants of the same mass or carbohydrate content were used in the experiment[26]. Introduction of a genomic clone (OS-ACS5) encoding 1-aminocyclopropane-1-carboxylic acid (ACC) synthase in the modern cultivar 'IR36', renders it more sensitive to submergence damage[27]. Such genetically engineered plants showed more elongation, which might be due to enhanced production of ethylene resulting in depletion of reserve carbohydrates at a faster rate and consequently poor survival percentage[17,28]. Application of an ethylene biosynthesis inhibitor, 1-methyl cyclopropene led to the reduction in elongation growth and better survival[10]. Plants also maintained higher carbohydrate levels and as a result survival percentage increased. The accumulated ethylene during submergence adversely affected antioxidant mechanism in intolerant rice cultivars, especially after de-submergence[29]. Ethylene also directly affects chlorophyll degradation as we have shown previously[10]. Hence, biotechnological manipulation aimed at reducing of GA/ethylene biosynthesis can be an approach to develop high-yielding, submergence-tolerant rice cultivars.

Protein synthesis is known to be adversely affected during submergence. However, certain new anaerobic proteins, many of them being enzymes of the carbohydrate metabolism, are induced during submergence. Some other existing enzymes, particularly those associated with energy metabolism, showed enhancement in activities as efficient utilization of reserve carbohydrate is essential to survive against submergence stress[5,30]. Plants endowed with scheme 1 (below) for starch utilization seem to survive the submergence stress better than those dependent on scheme 2.

Scheme 1: α-amylase $\rightarrow$ debranching enzyme $\rightarrow$ starch phosphorylase
Scheme 2: β-amylase $\rightarrow$ debranching enzyme $\rightarrow$ α-glucosidase.

The glycolytic pathways in which α-amylase acts together with the debranching enzyme result in the formation of oligosaccharides which undergo starch phosphorylase-catalysed phosphorolysis using inorganic phosphate to yield D-glucose-1-phosphate. This conserves the energy of the glycosidic bond in a phosphodiester bond. The glycolytic product of scheme 2 that begins with β-amylase is maltose, which is converted to glucose, the phosphorylation of which requires expenditure of an additional ATP. From the viewpoint of energetics, scheme 1 is therefore more favourable[5]. α-Amylase and starch phosphorylase activities have been found to increase under submergence in tolerant rice cultivars compared to susceptible ones[31,32], suggesting that the tolerant cultivars are able to use carbohydrates in more efficient ways.

Pyruvate decarboxylase (PDC) and alcohol dehydrogenase (ADH) are the two enzymes whose activities are known to increase during submergence leading to the production of ethanol and regeneration of NAD$^+$. Ethanol concentrations, however, do not reach toxic levels in submerged rice seedlings[33–35], though acetaldehyde may be toxic if its levels exceed a certain limit[36]. Acetaldehyde concentration has been found to be low in plants having higher activities of ALDH, a lesser known enzyme, with concomitant increase in submergence tolerance[37]. During re-aeration, acetaldehyde-oxidizing aldehyde dehydrogenase (ALDH) activity increases, thereby causing acetaldehyde content to decrease in rice[38,39]. As alcoholic fermentation is the primary means by which plants produce energy under anaerobiosis[40,41], much attention has been paid to the enzymes related to alcoholic fermentation and the endogenous level of substrate, i.e. carbohydrates[18,16,42–44]. Reports are available on the effect of genetic manipulation of PDC and ADH levels on submergence tolerance in rice[45,46]. These studies indicated that PDC transgenic rice is much more susceptible to submergence than the wild type as the former accumulates higher level of acetaldehyde. Further, higher activity of ADH in engineered plants did not contribute to submergence tolerance. Rather it made them more susceptible due to faster depletion of reserve carbohydrates. An optimum level of induction of this pathway is probably more associated with tolerance to provide minimum energy required for maintenance metabolism, but without excessive use of carbohydrates as the alcohol fermentation pathway is much less efficient than normal aerobic respiration.

Low light intensity and hypoxia are the two key factors that limit the ability of rice plants to survive long periods of submergence, due to severe damage to old leaves and the failure of sensitive plants to develop new leaves. Moreover, the submerged plants are promptly exposed to an environment with high light and oxygen tension, leading to the formation of reactive oxygen species (ROS) such as superoxide anion, hydroxyl radical and hydrogen peroxide that if not mitigated, can cause severe damage to cellular organization leading to plant death. Cultivars that are more efficient in detoxifying the ROS upon exposure to air are more capable of retaining their chlorophyll, maintain growth of older leaves, regenerate new leaves relatively fast and hence sustain plant growth. There appear to be two mechanisms by which tolerant rice plants protect themselves against oxidative damage:

(i) Presence of natural antioxidants: Naturally occurring, low-molecular weight compounds such as ascorbate, α-tocopherol, glutathione, carotenoids and phenols have been found to reduce oxidative damage in plants because of their antioxidant properties. Among these, only the role of ascorbate has been studied to some extent. Reports are available which showed that higher levels of particularly reduced forms of ascorbate did play an important role in protecting the plants against damage by ROS[29,47,48].

(ii) Presence of antioxidant enzyme systems: The pathway involving superoxide dismutase $\rightarrow$ catalase $\rightarrow$ peroxidase scheme is not so efficient in removing the ROS in rice, perhaps because the activity of superoxide dismutase (SOD) decreases under submergence. Moreover, after exposure to air, the activity of this enzyme remains below the level of control plants[22]. Ella et al.[49], however, found that the activity of SOD increases substantially in both 'FR 13A' and 'IR 42' upon de-submergence. Increase in SOD activity depends on the severity of stress and more studies are required to ascertain its role in scavenging ROS upon de-submergence. Another pathway, the ascorbate–glutathione cycle (Asada–Halliwel pathway), may play an important role in scavenging ROS upon de-submergence. This pathway regenerates the reduced forms of ascorbate and glutathione as primary scavengers. Higher concentrations of reduced ascorbate and lower levels of malondialdehyde (MDA) were observed in tolerant cultivars after desubmergence. MDA is the product of lipid peroxidation and its concentration is indicative of the extent of damage caused by ROS[49,50]. Kawano et al.[29] found strong negative correlations between MDA formation with ascorbate concentration ($r = -0.93$) and percentage survival ($r = -0.98$), and thus concluded that ascorbic acid is an important antioxidant in vivo for the recovery of submerged rice seedlings. Huang et al.[19] reported that sensitivity to salt stress increased in an ascorbate-deficient Arabidopsis mutant. These studies suggest that maintenance of higher levels of reduced forms of ascorbate is essential to mitigate the oxidative damage both under submergence as well as salt-stress situations. Ascorbate works in cooperation with glutathione and also helps maintain the regeneration of α-tocopherol, providing a synergetic role in the protection of cellular membranes[51,52]. Importance of plant phenolics and ascorbate in detoxifying H_2O_2 through peroxidases has been proposed by Yamasaki and Grace[53]. It was presumed that the H_2O_2 generated in the mitochondria diffuses to vacuoles where it is degraded by peroxidases using phenolics as primary electron donors. Phenoxyl radicals generated during the reaction are then reduced by both ascorbic acid (AA) and the monodehydroascorbic acid radical. If regeneration of AA occurs in the cytosol, it is transported to the vacuole[53]: a perxidase/phenolics/AA system in the vacuoles could then function to scavenge H_2O_2. This could well be the case in rice to remove these oxidative species generated during stress[47] and needs confirmation.

Germplasm improvement

Water-control measures in submergence-prone areas can help reduce the damage caused by flooding, but this normally entails huge investment beyond the reach of resource-poor farmers normally living in these areas. Floods also cannot be predicted and the damage could occur at any stage of plant development including germination. Development of suitable germplasm with higher levels of tolerance provides a better option, given the fact that sources of tolerance at different stages were identified and tolerance is genetically simple. The effect of flash-flooding in rice interacts with growth stage with germination being highly sensitive, and with tolerance increased progressively with seedling age.

Direct seeding is a common practice in rainfed lowlands and is becoming increasingly important because of labour scarcity and the high cost incurred during transplanting. In eastern India, sometimes early rains cause water stagnation in the field just after sowing which results in poor crop establishment. We have identified certain landraces like 'Panikekoa' and 'T 1471', which can establish relatively well under water. More cultivars with higher levels of tolerance need to be identified and the mechanism of tolerance is to be established. This trait also has immense value for irrigated conditions if seedlings could be established under water, as this will substantially suppress weed growth and reduce the cost of weeding and herbicide use.

In most cases, rice crop is submerged more than once during a single season. This repeated stress normally is more devastating even to the previously identified tolerant lines, though the extent of damage is dependent on the duration of each cycle. This necessitates searching for cultivars that can tolerate such conditions, particularly those that have the capacity for faster regeneration after submergence. This holds true for most of the submergence, avoiding types which are capable of extending their leaf tips above the water surface and thus avoid complete submergence. 'Sabita', a well-known submergence-avoiding variety, showed only 35% survival when submerged twice with a gap of ten days between, whereas 'Matia', a local landrace had more than 80% survival under similar conditions[54], suggesting that genetic variability in tolerance to repeated flooding may also exist in rice.

Most of the existing rice cultivars are seriously damaged if they are completely submerged for more than three days. However, a few tolerant landraces were identified that can withstand complete submergence for 10–14 days. Examples of these are 'FR 13A', 'FR 43B', 'Goda Heenati', 'Kurkaruppan' and 'Thavalu'. However, these tolerant landraces have poor combining ability and agronomic characters and possess the same locus that controls most of the submergence-tolerance phenotype ($Sub1$)[55,56]. Tolerant breeding lines with improved agronomic characteristics have now been developed, and some breeding lines such as IR49830-7 had yields equivalent to the irrigated

checks. Other new breeding lines with adequate tolerance to submergence have been recommended for release in India[4]. Despite this progress in breeding, adoption of these new varieties has been somewhat limited probably because none of them combines desirable levels of flooding tolerance with grain yield and quality preferences. Hence, identification of new donors through exploring new genetic resources is highly desirable. Recently, we have identified some new rice landraces with reasonably higher levels of tolerance to submergence but with better agronomic traits than the landraces identified before. These lines are 'Atiranga', 'Khoda', 'Khadara', 'Kusuma' and 'Kalaputia'. However, these lines are yet to be tested for their combining ability as well as the dominant mechanisms associated with submergence tolerance[57]. Among these new submergence-tolerant genotypes, 'Atiranga' was found to have higher levels of tolerance compared to other tolerant types, including the traditionally most tolerant check, 'FR 13A'. This line has higher regeneration capacity and survival rate even when submerged for long period ($\geq$ 18 days, our unpublished data). Identification of such lines suggested the possibility of identifying new genes or alleles for higher levels of tolerance.

Concerted efforts made by CRRI, Cuttack and other Rice Research Stations in eastern India on varietal improvement for these adverse situations in flood-prone areas under the ICAR–IRRI collaborative Rainfed Lowland Shuttle Breeding Programme, led to the development of a number of improved lines for submergence tolerance. Some lines like 'CR 2003-13', 'CR 2006-7', etc. are found to have submergence tolerance at par with 'FR 13A'. A number of improved lines developed/identified under this programme namely 'Kishori', 'Satyam', 'OR 1206-25-1 (Jagabandhu)', 'OR 1234-12-1 (Upahar)', 'CN 1035-61 (Bhudev)', 'CRLC 899 (Varshadhan)', 'TTB 238-3-38-3 (Prafulla)', 'NDR 8002', 'CR 2003-2', 'CR 2003-3', 'CR 978-8-2', etc., have been released in different states of eastern India or are in the advanced stages of testing in national coordinated trials. Besides, a set of advanced lines, namely 'IR40931-26-3-3-5', 'IR49830-7-1-2-1', etc. with strong submergence tolerance and desirable agronomic features developed at IRRI, Philippines are being used in breeding programmes to incorporate submergence tolerance into some of the popular high-yielding lowland varieties using conventional as well as molecular breeding approaches.

Genetic studies suggested both simple and quantitative inheritance for submergence tolerance[56,58]. Using a population developed from a cross between an *indica* submergence-tolerant line ('IR40931-26') and a susceptible *japonica* line ('PI543851'), a major QTL was mapped[55] to chromosome 9, designated as *Sub1*, and this QTL accounted for about 70% of the phenotypic variation in submergence tolerance in the population studied. In a subsequent study[59] the importance of *Sub1* in submergence tolerance was confirmed and four additional QTLs were identified

on four different chromosomes. Moreover, the *Sub1* locus has been fine mapped, paving the way for its positional cloning[60,61]. Siangliw *et al.*[62] and Toojinda *et al.*[63] identified several major QTLs for plant survival, plant height, stimulation of shoot elongation, visual tolerance score and leaf senescence, each mapped to the same locus on chromosome 9. Minor QTLs influencing tolerance were also identified on chromosomes 1, 2, 5, 7, 10 and 11. Some of these QTLs are specific to particular traits, environments, or genetic backgrounds. All identified QTLs were found to contribute to submergence tolerance by reducing underwater shoot elongation, maintaining chlorophyll levels, or by triggering one or more of the events discussed earlier to be associated with submergence tolerance[22,49]. Introduction of *Sub1* locus in a highly adapted and popular Thai rice cultivar 'KDML 105' enhanced its submergence tolerance[62,64], while maintaining all the desirable characters of the parent material. Through marker-assisted breeding, *Sub1* was also introgressed in 'Swarna', a popular rice cultivar in South and Southeast Asia and is currently being tested in farmers' fields in India and Bangladesh. Introgression of *Sub1* into 'Swarna' greatly enhanced its survival under submergence (Figure 1), and with no other obvious effects on original characteristics of 'Swarna'. Our preliminary field experiments under normal conditions showed that both cultivars have similar grain yield (Figure 2). The dramatic effect of *Sub1* on what is essentially a quantitative trait, suggests a regulatory locus rather than a specific enzyme. Efforts are currently in progress to introduce *Sub1* into popular high-yielding rice varieties of rainfed lowland. This approach will substantially enhance submergence tolerance of these varieties without sacrificing their quality aspects or yield potential, and will dramatically shorten the breeding cycle. Another level in the hierarchy of genetic controls which have a bearing in regulating stress responses is exercised at the signal transduction pathway. Small GTP-binding proteins
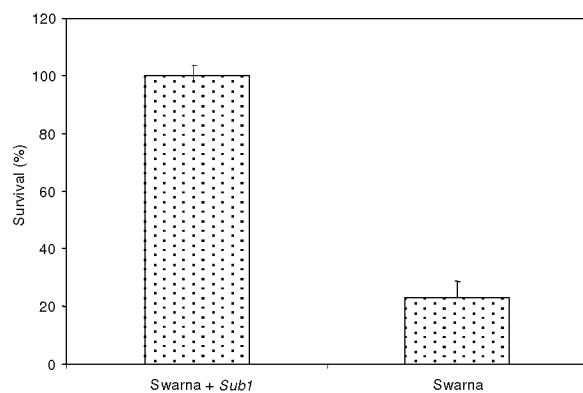

Figure 1. Percentage survival of 'Swarna + *Sub1*' and 'Swarna'. Twenty-one-day-old seedlings were completely submerged for 10 days and then allowed to recover for another 10 days. Data are means of four replications and vertical bars represent standard deviation.
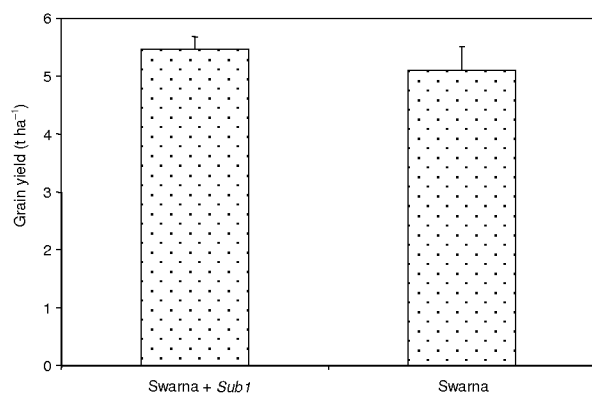
Figure 2. Grain yield of 'Swarna + *Sub1*' and 'Swarna' under favourable rainfed lowland conditions. Data are means of four replications and vertical bars represent standard deviation.

play critical roles in signal transduction in mammalian and plant systems[65,66]. The prospects for altering signalling pathway in flooding stress tolerance are yet to be tested.

Future prospects

Rice is the only cereal crop that is well adapted to the conditions of waterlogging or partial flooding. Performance of high-yielding varieties can further be augmented if their submergence tolerance is enhanced, which is now becoming more feasible. Current understanding of the physiological and biochemical bases of submergence tolerance has progressed well in recent years, making it possible to design efficient phenotyping protocols and has laid the infrastructure for further genetic and molecular studies, to discover genes underlying component traits associated with tolerance. This will subsequently speed up the breeding process if individual genes can be combined in favourable phenotypes through marker-assisted selection, as the example shown with *Sub1* locus.

So far, only one submergence tolerant landrace ('FR 13A') has been extensively exploited in breeding as well as in mechanistic studies, because of its higher level of tolerance compared with all other lines that were tested before. We recently identified few more tolerant genotypes distinct from 'FR 13A', in being agronomically more desirable. This could probably provide better donors and sources of new genes, as some of these lines showed better performance than 'FR13A'. Substantial efforts are needed to characterize these new materials, to study their genetic and physiological bases of tolerance and to discover useful genes and alleles associated with tolerance. These can then be developed into molecular markers to pyramid them into popular varieties.

Another important finding is the identification of rice cultivars with an inherent capacity of seedling establish-

ment under anaerobic conditions. These cultivars shall be a boon for rainfed lowlands as well as for irrigated direct seeding systems. Greater efforts are now being devoted to identify more sources and understand the bases of such tolerance. Our ultimate goal is to combine tolerance to flooding both during germination as well as during vegetative growth to develop robust varieties for better and stable productivity in flood-prone areas. Cultivars are also needed that can withstand repeated flooding and have better regeneration capacity as well as faster growth after flooding to produce sufficient biomass in a shorter period and to minimize delay in flowering when submerged.

Maintenance of high levels of stored carbohydrates in the seedlings prior to submergence coupled with minimum shoot elongation and retention of chlorophyll are all desirable traits for submergence tolerance. Biotechnological and genetic approaches to enhance the efficiency of pathways involved in carbohydrate metabolism and regulation of plant hormones such as GA, an ethylene to restrict elongation and reduce chlorophyll loss under submergence could help in developing germplasm with enhanced tolerance. An important area of research on submergence tolerance and probably for tolerance to other abiotic stresses is to develop well-defined and efficient antioxidant systems to effectively remove toxic radicals generated during stress and preserve cellular membranes and organelle structure.

Beside genetic enhancement, some of the traits associated with submergence tolerance could also be manipulated through certain management practices, such as proper nursery management to enhance the build-up of carbohydrates in seedlings before transplanting through proper spacing and balanced nutrition. Excessive use of some nutrients, such as nitrogen, was found to cause excessive shoot growth, deplete carbohydrate storage and alter carbohydrate partitioning between roots and shoots even without submergence, all of which aggravated seedling mortality upon submergence. Proper nutrient management after water recedes may also enhance recovery and grain yield. These simple management approaches could substantially enhance survival and productivity in flood prone areas, particularly when combined with genetic tolerance. Further investigations are needed to test the feasibility of these management packages in farmers' fields.

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