



# Antioxidant Protection Mechanisms Reveal Significant Response in Drought-Induced Oxidative Stress in Some Traditional Rice of Assam, India

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**Abstract:** Drought tolerance levels and antioxidant protection mechanisms were evaluated for 21 traditional rice varieties of Assam, India, along with Sahbhagi Dhan (drought tolerant) and IR64 (drought sensitive) as controls. Drought was imposed in hydroponic culture with polyethylene glycol 6000 (PEG6000) that was initially standardized with different concentrations. All the rice varieties showed apparent decreases in growth characteristics under drought stress (initially at 15% for 7 d followed by 20% PEG6000 for 7 d in Yoshida medium). On the basis of standard evaluation score (SES), eight rice varieties showed high drought tolerance which were carried forward for further biochemical analyses. Based on different morpho-physiological parameters, SN03 (Bora), SN04 (Prosad Bhog), SN05 (Kola Joha), SN06 (Helash Bora), SN08 (Salihoi Bao), SN12 (Kola Amona), SN20 (Ronga Bora) and SN21 (Sok-Bonglong) were identified as promising drought tolerant varieties. The non-enzymatic antioxidants activities viz., glutathione, ascorbate and enzymatic antioxidant activities such as superoxide dismutase (SOD), catalase (CAT), guaiacol peroxidase (GPX), ascorbate peroxidase (APX), glutathione reductase (GR) in shoots and roots of all the selected varieties revealed significant level of protection mechanisms as compared with controls. Enhancement in activities of the overall antioxidant enzymes including SOD, GPX, CAT, GR and APX under drought stress reflects their role in the adaptation process under water stress.

**Key words:** drought stress; traditional rice; standard evaluation score; antioxidants protection mechanism; antioxidant enzyme

Abiotic and biotic stresses, such as salt, drought, cold, heavy metals, pests and various diseases, are the major factors that grossly affect rice product. As a consequence, plant needs to develop protection mechanisms to cope with environmental stresses (Zhou et al, 2009). Water availability is the most important among the abiotic factors that have shaped and continue shaping plant evolution in general and rice crop in particular (Bray et al, 2000; Nahar et al, 2016). Drought affects growth and development of rice, reduces production and in severe conditions, results in death of rice plants.

Drought stress is a severe threat to sustainable rice cultivation as it causes extensive loss to productivity (Wang et al, 2012).

Rice (*Oryza sativa*) plays a major role as a staple food for more than half of the world's population (Khush, 2005). Rice is found to be most susceptible to damage from water deficit. It is estimated that 50% of world rice production is affected by drought (Lafitte and Bennet, 2003). About 40 million hm<sup>2</sup> of drought-prone area in rice environments globally is available which produces substantially low yield

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(Singh and Mackill, 1991). Soil moisture deficit at early stages of growth intensely impairs the establishment of rice crops. Leaf rolling is one of the acclimation responses of rice and used as a criterion for scoring drought tolerance. Drought-induced spikelet sterility is well documented in rice (O'Toole and Moya, 1981; Guo et al, 2013). Upland rice cropping system, which is entirely dependent on rainfall for growth and yield, often experiences water deficiency. Matsushima (1962) observed that rice is sensitive to water deficit from 20 d before heading (booting) to 10 d after heading. Grain yield reduction due to water stress at the flowering stage is mainly caused by reduction in number of filled spikelets per panicle without a substantial decrease in spikelet number per panicle (O'Toole and Namuco, 1983; Guo et al, 2016). A given level of drought, at the vegetative stage, can cause a moderate reduction in yield but drought can entirely eliminate yield if it coincides with pollen meiosis or fertilization (O'Toole, 1982). Upland rice, which needs to rely on rainfall, is often exposed to drought stress and consequently has developed drought-resistant traits (Yadav et al, 1997).

Drought stress results in stomatal closure, which limits CO<sub>2</sub> fixation and reduces NADP<sup>+</sup> regeneration by the Calvin Cycle (Sato and Murata, 1998). These adverse conditions increase the rate of reactive oxygen species (ROS), such as H<sub>2</sub>O<sub>2</sub> (hydrogen peroxide), O<sub>2</sub><sup>-</sup> (superoxide), O<sub>2</sub> (singlet oxygen) and OH<sup>-</sup> (hydroxyl) radicals, by enhanced leakage of electrons to molecular oxygen. Production of ROS has been found to be stimulated in plants under a variety of environmental stresses (Sgheri et al, 1996). ROS causes peroxidation of lipids, denaturation of proteins, mutation of DNA and various types of cellular oxidative damage (Avramova et al, 2017). Plants possess an efficient antioxidant (enzymatic and non-enzymatic) defense system to cope with ROS-induced oxidative stress (Anjum et al, 2017). To scavenge ROS, plants have evolved specific defence tactics involving both enzymatic and non-enzymatic antioxidant mechanisms. Rapid accumulation of free proline is a typical response to drought stress. When exposed to drought stress in the soil, many plants accumulate high amounts of proline, in some cases several times the sum of all the other amino acids (Lum et al, 2014). Comparison of antioxidant defense systems, lipid peroxidation and proline contents of rice cultivars differing in drought tolerance may be helpful in developing a better understanding of tolerance

mechanisms to drought stress.

Polyethylene glycol (PEG) is a polymer produced in a range of molecular weights. PEG compounds have been used to simulate water stress effect in plants (Murillo-Amador et al, 2002). Lagerwerff et al (1961) indicated that PEG can be used to modify the osmotic potential of nutrient solution culture and thus induce plant water deficit in a relatively controlled manner, appropriate to experimental protocols. It was assumed that PEG of large molecular weight did not penetrate the plant and thus was an ideal osmoticum for use in hydroponics root medium. During the 1970's and 1980's, PEG of higher molecular weight (4 000 to 8 000) was quite commonly used in physiological experiments to induce controlled drought stress in nutrient solution cultures.

Assam is the traditional rice growing area in India, and rice diversity in the region is very high possibly due to different agroclimatic ecosystems (Ahmed et al, 2007). Further, rich ethnic diversity also plays a pivotal role in shaping rice evolution in the state. The state has its climatic and physiographic features favourable for rice cultivation, and the crop is grown in a wide range of agro-ecological situations including hill slopes to very deep-water areas, wet humid months to a drier period of the year. There are three distinct rice growing seasons in Assam viz., ahu (February/March–June/July), sali (June/July–November/December) and boro (November/December–May/June). Rainfed ahu rice suffers from prolonged drought at panicle initiation stage of the crop due to the late arrival of monsoon leading to substantial yield loss in rice. Therefore, identification of drought tolerant rice from these traditional resources would be a boon for future rice improvement program.

## MATERIALS AND METHODS

### Plant materials and growth parameters

Twenty-one traditional rice accessions were collected from six different agro-climatic zones of Assam, India. Sahbhagi Dhan, a drought resistant variety (released by Central Rainfed Upland Rice Research Station, Hazaribagh, Jharkhand), and IR64, a drought sensitive variety, were used throughout the study as positive and negative controls (Table 1). Collected rice samples were kept at 4 °C for 3–4 months for regular use, and fresh new seeds were collected every year until the end of the experiments.

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