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MOLECULAR MECHANISM OF SALT TOLERANCE IN MILLETS - A REVIEW

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SUMMARY

Millets are nutrient-dense staple crop grown in a wide range of agro-ecological conditions, including plains, coasts, and hilly regions, throughout the semiarid tropics of Asia and Africa. Millets are a potential agricultural crop with several uses, including food, feed, fodder, and raw materials for the brewing and biofuel industries. Climate resilience and the rising demand for nutrient-dense food, feed and hay have given them a remarkable importance in agriculture. Salinity stress severely hinders the growth and development of crop plants. Salinity affects over 20% of the world's irrigated land, which lowers agricultural output by 20%. Therefore, it is necessary to develop salinity tolerance in the major forage and nutri-cereal crops for sustainable global food, nutritional, biofuel and fodder security.

Key words : Forage crops, millets, salt-overly-sensitive (SOS3)

Millets are regarded as sustainable crops with enormous potential to impart global food security and nutrition in the face of escalating agricultural costs and climatic changes. They are climate resilient crops with human health benefits, supplementary wellbeing profit and needs only fewer effort overheads for growing. Millets are a group of small-seeded annual panicoid grasses with C_4 photosynthetic pathway. They have shorter growing periods and are considered as favourable crops for stressed environments (Blatensperger, 2002). Salinity, drought, flooding, high temperature, cold, *etc* are among the abiotic factors that have a huge impact on world agriculture and account for more than 50% reduction in average yields for most food and fodder crops (Wang *et al*., 2003). Over 35% of the world's land is arid or semiarid. In these areas, precipitation is often inadequate for most of the agricultural practices. Only 9% of the world area is conducive for crop production, while 91% is afflicted by various stresses. Yield losses due to high temperature (40%), salinity (20%), drought (17%), low temperature (15%) and other forms of stresses have been reported (Ashraf *et al*., 2008). A major challenge towards global sustainable agriculture is the requirement for production of 70% more food crops for an estimated additional 2.3 billion population by 2050 (FAO 2009). Salinity stress is a major limiting factor in meeting the increase in the demand for food crops. More than 20% of the cultivated land worldwide is affected by salt stress and the area is increasing day by day. In India, about 6.72 M ha has been affected by salinity. The extent of salt-affected soils (SASs) is projected to increase to 16 million ha by 2050, as a consequence of climate change and/or human activities (CSSRI, 2015). The estimates suggest India incurs soil salinity related loses to an extent equivalent to 17 million tons of food grains annually (Sharma *et al*., 2015). Climate resilient crops such as grasses: guinea grass, bajra -napier hybrids and tri-specific hybrids, dinanath grass, (Dheeravathu *et al*., 2018, Singh *et al.,*2020, Dheeravathu *et al*., 2021a, Dheeravathu *et al*., 2021b, Dheeravathu *et al*., 2022a), Antony *et al*., 2021, Dheeravathu *et al*., 2022b), pulses: cowpea, berseem, *clitoria, centrosema, siratro* (Dheeravathu *et al*., 2017a, Dheeravathu *et al*., 2017 b, Dheeravathu *et al*., 2021a, Dheeravathu *et al*., 2021c, Dheeravathu *et al*., 2022b, Dheeravathu *et al*., 2022c), forage cereals including millets: pearl millet and sorghum (Singh *et al*., 2010, Prasad *et al*., 2015, Tara Satyavathi *et al.,* 2021, Dheeravathu *et al*., 2022a, Malathi *et al*., 2022), have been proven to be climate smart. Considering the adverse effect of accumulated salts in soil and irrigation water, elevated CO_2 and temperature on soil health as well as forage yield and productivity, it is high time for in depth understanding of physiological and biochemical changes in forage crop varieties/ genotypes/ lines in response to climate change. Adoption of good bioremediation practices could play a major role in improving soil health management, sustaining livestock production and thus will be helpful in future soil health management and breeding programs. Average yield losses of 15-90% have been well documented in major crops due to salinity stress. For instance, maize, wheat, and cotton exhibited 55%, 28%, and 15% grain yield loss respectively under moderate soil salinity, while up to 55% and 93% yield losses were observed in cotton and tef (*Eragrostis tef)* respectively under high saline conditions (Tadele, 2018; Zörb *et al.,* 2019). Very little information is available on millets regarding salt stress responses and yield losses, as compared to other crops. In finger millet, a significant reduction in grain yield by 23–27% was estimated (Krishnamurthy *et al*., 2014). In a recent study, 13-22% reduction in pearl millet grain yield was observed under salinity levels of 8-12 dSm^{-1} (Yadav *et al.,* 2020). Yuan *et al.,* (2021) reported that proso millet is among the resistant and adaptable crops in arid and semi-arid regions and it has strong sodium ion (Na^+) toxicity resistance strategies.

Salt tolerance mechanism in millets

Plants are classified broadly into two major types depending on their response to salinity: the halophytes that can withstand salinity and the glycophytes that cannot withstand salinity and eventually die. Glycophytes are defined as a group of species which has evolved by adaptation under natural selective pressures in ecosystems with low soil sodium levels and which maintains low sodium levels in its aboveground tissues, especially in its leaves (Cheeseman, 2015). Millets are categorized as glycophytes and can tolerate average salt threshold of about 6 EC_e (dS/m) with some variation from species to species (Table 1). Plants develop various physiological and biochemical mechanisms in order to survive in soils with high salt concentration. The different types of salt tolerance mechanism include (a) ion homeostasis and compartmentalization, (b) ion transport and uptake, (c) biosynthesis of osmoprotectants and compatible solutes (d) activation of antioxidant enzymes and synthesis of antioxidant compounds, (e) synthesis of polyamines, (f) generation of nitric oxide (NO), and (g) hormone modulation. Ion homeostasis and compartmentalization (K^+/Na^+) mechanisms are among the major salt tolerance mechanism in plants and discussed in detail below.

Many studies have suggested that salinity increases sodium in the shoots and roots and reduces potassium in various plant organs. Potassium (K^+) mostly contributes to a plant's survival when exposed to various environmental stresses such as drought, salinity, and cold (Wang *et al.,* 2013). The positive role of K^+ in response to salinity is due to: (1) it is competitive with sodium (Na^+) for binding and maintaining relative water content (RWC) in plants (Capula-Rodríguez *et al*., 2016); (2) K^+ can regulate the balance between ROS and antioxidants to adjust protein synthesis and stomatal function, thereby improving the plant's photosynthetic status (Wang *et al*., 2013). Salinity-tolerant cultivars have different K^+/

TABLE 1
Threshold salt tolerance in millets

S. No.	Common name of millets	Botanical name	Tolerance based on	Salt threshold and rating [ECe (dS/m)/Or mM]		Reference
1.	Sorghum	*Sorghum bicolor* (L.) Moench	Grain yield	6.8	MT	Francois *et al.,* (1984)
2.	Pearl millet	*Pennisetum glaucum* (L.) R. Br.	Seedling stage	100 mM NaCl		Rahim *et al.,* (2000)
3.	Finger millet	*Eleusine coracana* Gaertn.	Germination/ seedling stage	6 dS/m		Shailaja, and Saminadane (2007)
4.	Barnyard millet	*Echinochloa frumentacea* L	Germination	200 mM		Arthi *et al.,* (2019)
5.	Foxtail millet	*Setaria italica* (L.) P. Beauvois	Grain yield/ dry matter	6.0 -	MS	Kubsad *et al.,* (1995)
6.	Little millet	*Panicum sumatrense* Roth. ex Roem. & Schultz)	Grain yield	Moderate levels		Siddique *et al.,* (2022)

MT-Moderately tolerant, MS-Moderately susceptible.

Na^+ ratio in different organs and at different growth stages compared to salinity-sensitive cultivars (Dheeravathu *et al.,* 2020, and 2021). The K^+/Na^+ ratio increases salinity tolerance in millets. Krishnamurthy *et al.,* (2007) reported that high K^+/Na^+ and Ca^{2+}/Na^+ ratios were positively related to salt tolerance in sorghum. Dheeravathu *et al.,* (2021) reported that photosynthetic pigments showed positive correlation with K^+ and K^+/Na^+ ratio while there is a negative correlation between Na^+ and chlorophyll content in bajra napier hybrids. Numerous studies have shown that salt tolerance is ultimately manifested in plants through several physiological processes including Na^+ uptake and exclusion, in homeostasis, especially between K^+/Na^+ and partitioning. Various studies have shown that plants increase Na^+ uptake and reduce K^+ uptake under salt stress. The K^+ ions are beneficial to plants and by increasing K^+ content, plants can reduce the absorption of Na^+ ions to a certain extent, thus increasing the K^+/Na^+. The entry of Na^+ into cells causes severe ion imbalance and excess uptake can cause significant physiological disorder(s). High Na^+ concentration inhibits the uptake of K^+ ions, ultimately resulting in lower production eventually leading to death (James *et al*., 2011).

Salt Overly Sensitive (SOS): Hypothetical salt tolerance mechanism in millets

Salt stress increases the intracellular Na^+ (increased Na^+ /K^+ ratio), induces Ca^{2+} signalling and activates SOS3 (salt-overly-sensitive), which is a calcium B-type like calcium-binding protein. SOS3 binds to plasma membrane and interacts with SOS2 which is a serine/ threonine protein kinase (Fig-1). SOS3/ SOS2 complex then activates SOS1 which is an ATP-dependent Na^+ /H^+ exchanger which can be phosphorylated by SOS3/ SOS2 complex. Active SOS1 ensures Na^+ exclusion from plant cells *via* ion exchange (Na^+ is exchanged for H^+) at the cost of ATP. SOS1 controls Na^+ loading into xylem thus regulating Na^+ level in transpiration stream and its root to shoot transport. Thus, the up regulation of SOS pathway proteins provides tolerance under salt stress. Proteomic studies supporting elevation in the expression of SOS proteins have been carried out under salinity. The OsSOS1 plasma membrane gene plays a crucial role in sodium extrusion from root epidermal cells under salinity. Transfer of OsSOS1 gene from *Oryza sativa* to finger millet and its overexpression conferred high salt tolerance in finger millet, promoted

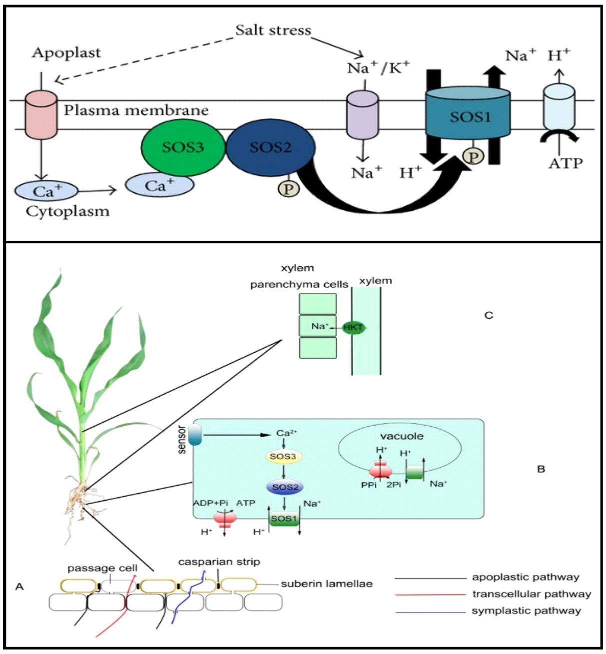

Fig. 1. Model of SOS Pathway for salinity stress responses (adapted from Gupta and Huang 2014).

seed germination and increased root length, shoot length, chlorophyll, membrane stability index and reduction in reactive oxygen species (ROS) relative to wild type plants (Pushpa *et al.,* 2020).

CONCLUSIONS

India's sustained food, nutritional, biofuel and fodder security are under risk due to increased soil salinization. Higher daytime temperatures during summer have led to more evapotranspiration, which in turn cause salt build up on soil surface. It has negative influence on crop plants' physiological, biochemical, and molecular salt tolerance processes as well as the biophysical characteristics of soil. Millets have the potential to grow under challenging and resource-poor marginal conditions, and they hold enormous potential for global food security and nourishment in the face of changing climatic conditions. This is especially true in the scenario, where, cultivable lands are decreasing due to soil salinization, increased urbanization, and industrialization. As compared to other crops, very little is known about the advantages of millets: The impact of saline stress, salt tolerance, and mitigation of salt stress. Therefore, a thorough investigation of the physiological, biochemical, and molecular mechanisms of salt stress in the millet species, more specifically, the mechanisms underlying stress tolerance at the varietal level will be fruitful. The development of salt-tolerant millets and several potential tactics for enhancing salt tolerance are encouraged.

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