Chapter

Sugarcane Response and Its Related Gene Expression under Water Stress Condition

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Abstract

This review paper is to study the different responses expressed by the sugarcane when exposed to water stress conditions, that is, waterlogging and drought. Water stress is one of abiotic stress affecting sugarcane productivity and the development of water-use efficiency and the morphological character get varies with genotypes, duration and intensity of stress and types of tissue damage and expression of variable patterns of a gene that makes a high degree of complexity on sugarcane under water stress condition. Since, there is little stepping towards sugarcane crops coming from genetics, agronomics, and molecular biology. These studies provided the framework for researching the morphological basis of genetic variation and mycorrhizal colonization in water stress tolerance and yield improvement under water-limited conditions.

Keywords: sugarcane, gene, yield, water stress, complexity

1. Introduction

Sugarcane (*Saccharum officinarum* L.) is a unique crop that can accumulate high levels of sugar and is also a commercially viable source of bioelectricity and second-generation bio-ethanol. It provides over 76% of the sugar for human consumption from approximately 27.12 million hectares area with the production of 1900 million metric tons around the globe [1]. Brazil is the leading country in terms of sugarcane area and production followed by India and China [1]. Sugarcane provides juice, which is used for making white sugar, jaggery (gur), and many by-products like bagasse and molasses. Sugar and its fermented products are very important in making and preserving various kinds of medicines like syrups, liquids; capsules, etc. Bagasse is used as a fuel, for the production of fiberboard, papers, plastics, and furfural. Molasses is used in distilleries for the manufacture of ethyl alcohol, butyl alcohol, citric acid, ethanol, etc. Rum is the best potable spirit made from molasses. Molasses is also used as an additive to feed livestock. Green tops of cane are a good source of fodder for cattle. Press-mud, the remains after juice clarification is good manure. Steam produced during the boiling of the juice is used to generate electrical power. Despite its importance in production and productivity sugarcane is affected by various biotic and abiotic stresses. On the other hand, no use of improved and recommended varieties also causes yield decline, among the abiotic stresses Water stress Is one of abiotic stress abiotic stress affecting sugarcane productivity. Therefore, the development of water-use efficient and water-stress-tolerant cultivars is imperative for major sugar-producing countries and industries. The susceptibility of sugarcane to water stress is greatly exposed in tillering and stem elongation phases with both leaf and stem growth [2] but has a positive effect in the maturation phase with sucrose content. The morphological character get varies with genotypes, duration, intensity of stress, and types of tissue damaged [3]. However, reported large potentially exploitable genetic variation for cane yield, responses like leaf rolling, inhibition of stalk and leaf growth, leaf senescence and reduced leaf area, and root development pattern [4]. Knowledge about stress biology in other crops increases coming from genetics, agronomic and molecular biology but still, there is little stepping towards sugarcane crops.

The use of arbuscular mycorrhizal fungi (AMF) has significant importance for more efficient crop development and the use of fertilization, especially in the early stage of development [5], and helps in the absorption of soil nutrients like phosphorus and plant resistance to biotic and abiotic stress. Abdel-Fattah & Asrar [6] reported that its association increases tolerance to water stress, contributing to aggregate stability, increase in soil aeration and subsequent availability [7], the reason behind, plants maintain higher stomatal conductance during water deficit periods and, in return, higher diffusive dependence. However, studies on interaction between AMF and sugarcane are scarce, and need to develop studies of AMF in sugarcane for different water stress conditions in lab conditions to explore the importance to cope with different stress situations.

Plant species go under different alternations in antioxidant enzyme activities. Drought tolerant crops showed the up-regulation of SOD and CAT activity as a general mechanism, where there is a remarkable correlation between antioxidant and proline content that signifies the specific traits for future identification of drought tolerance [8] that contributes protection against oxidative damage [9]. These enzymes were further affected by plant species, cultivars, and stress intensity and duration.

From the perspective of molecular studies, a wide range of gene are explicitly recognized that are expressed in sugarcane under water stress condition [10] that encodes putative chaperones dehydrin (DEH) and late embryogenic (LEA) protein, enzymes involved for metabolism and oxidation of proline [10]. There is an expression of variable patterns of gene that makes a high degree of complexity on sugarcane under water stress conditions. On other hand, signal transduction pathways lead to stress response due to plant hormones like ABA play an important role in signaling and gene regulation. Under water deficit, ABA concentration increases and makes stomata functional to protect against rapid desiccation [11]. Use of beneficial nitrogen fixing bacteria and gibberellic acid (GA_3) involves in gene expression and regulates the metabolic process as a function of sugar signaling and antioxidative enzymes [12].

Study of the morphological and stress response helps us to develop the concept for future breeding programs and selection of the water stress tolerant genotypes. In the present context, there are limited varieties named Jitpur series, that does not resolve the problem of water stress condition, so further study should be conducted for studying the response to water stress and selection of genotypes under different level of stress condition. So, this research should be conducted for studying the morphological and stress-responsive character of different genotypes for better crop improvement and to find a suitable variety for water stress conditions.

The knowledge of response and genetic variation of sugarcane to water stress under field conditions is relatively limited [13] and restricted in a small number of studies. Most studies on water deficits in sugarcane have focused on irrigation management practices [14, 15]. A number of physiological investigations conducted under laboratory or glasshouse conditions together with modeling studies have advanced the knowledge of water relations, stomatal functions and osmoregulation, carbon assimilation, and dry matter partitioning in water-stressed sugarcane [2, 3, 16–20]. Sugarcane breeding program has access to a broad range of parental germplasm and cultivars developed more than 100 years of breeding with selection, which derives from wild cane are relatively less developed and selected.

Despite the other major consequences of climate change, it is predicted that sugarcane yield will increase with increasing average minimum and maximum temperatures in winter, and increasing maximum temperatures in the rainy seasons [21]. Recent Nepalese climatic projections support the above assumptions and suggest that these conditions are likely. However, recent studies on the impacts of climate change on sugarcane [22] indicate that climate change-induced drought is one of the most significant challenges for sugarcane production. Moreover, the crop is more able to withstand natural disasters such as flood than rice and/or other cereal crops. Regardless of the high resilience to natural disasters, less than half of the potential of sugarcane production is currently realized [23]. The morphological basis of genetic variation and mycorrhizal colonization under different water stress situations were studied.

2. Morphological responsiveness of sugarcane under water stress condition

Morphological character is the first character seen when sugarcane gets exposed to water stress conditions. On exposure to drought conditions, there is a reduction in tillering, leaves discoloration, rolling of leaves and leaves folding and shredding, reduced leaf area due to narrow leaves, and decrease in lipid peroxidation. Under the waterlogged conditions, the sugarcane showed pipping in stalks and roots, development of adventitious roots, and the presence of aerenchyma tissue without which sugarcane cannot survive in water-logged condition.

Sugarcane, an important source of sugar and ethanol, demands relatively high water and is highly sensitive to water deficit [24]. The use of one Ml of irrigation produces 8–12 ton cane [25] and water deficit can lead to 60% productivity losses [26]. For this reason, production areas are concentrated in regions with favorable rain regimes for sugarcane growth and development [27], while in other areas crop production requires supplemental or full irrigation [28]. The increasing incidence, duration, and intensity of severe water deficit have promoted large sugarcane crop improvement programs for water use efficiency and water stress tolerant varieties that make impetus to develop biotechnological strategies.

From the perspective of growth stages and water management practices, susceptibility is more in tillering stage ad stem elongation phases [2] with both stem and leaf growth being more affected than other organs. The morphological and physiological responses of sugarcane plants get varies with genotypes and duration and intensity of stress and type of tissue damaged and substantially decreased both cane and cane yield. There, as reported, has a large potentially exploitable genetic variation for cane and cane yield under stress conditions. The most common water stress responses in sugarcane are leaf rolling, stomatal closure, inhibition of stalk and leaf growth, leaf senescence, and reduced leaf area [2]. Besides cell division and elongation get interrupted, so stem and leaf elongation and root development are more seriously affected during growth processes [4, 29].

There is an interesting fact that increased levels of sugar, such as trehalose, can help plants to cope with water deficit, reducing the damage to cell membrane [30].

3. Mycorrhizal colonization variance in water stress condition

Water stress strongly affects mycorrhizal development in roots and soils. The short-term soil drought did not appear to favor or discourage root AMF colonization and longer-term soil drought decreased AMF colonization. So, there are increased levels of root AMF colonization in response to drought stress than in decreased levels, which is related to reductions in plant P levels.

The growth of AMF spores varies with soil moisture available, which alters spore behaviors. Spore germination is favored n soil at or above field capacity but is decreased with decreasing soil water potentials below field capacity [31]. AM-stimulated plant growth enhancement may be more important in the host plant under drought stress than under watered conditions. In addition, helps in increase of P nutrition, water uptake by hyphae, and increase of root length [32].

The survivability of plants under stress conditions is strongly enhanced with AMF inoculation. This may be due to a more effective root architecture for water and nutritional absorption and the developed external hyphae in the soil [33]. Mycorrhizal plants would recover more quickly from wilting than non-mycorrhizal plants after drought recovery [34]. In addition, leaf morphological adaptation, and root morphological adaptation are other strategies for mycorrhization under drought stress. This adaptation provides more exploration of soil volume to absorb water and nutrients from the soil [35], thereby potentially enhancing the drought tolerance of the host plant. Mycorrhizal hyphae help in the uptake and transport of water from bulk soil to the host plant and also considerable quantities of phosphates and nitrogen to the plant from soil zones but yet no significant evidence of transfer of water from hyphae to plants. In sum, AMF-enhanced nutrient absorption is an important physiological mechanism in drought tolerance of the host plant caused by mycorrhization.

4. Genetic variability of sugarcane under water stress condition

Crop growth under water stress has been frequently and usefully conceptualized as the product between the components of total water use and water use efficiency. The high genetic correlation between water levels is likely traits for contributing to increased levels of component under fully irrigated and contributed to high yield under water stress. Genetic gains accomplished from long-term selection in breeding programs conducted under relatively non-stressed situations extend large degrees under water stress.

In relation to water use efficiency, high photosynthetic capacity improves rates of crop growth under non-water-limiting conditions and also improves water use efficiency through reducing leaf internal CO_2 levels, thus, contributing to improved performance also under water-stress conditions. Similar to water use, high radiation use efficiency and fast canopy development under non-limiting conditions contribute to increases above- and below-ground (root) growth. Genotypes with larger canopies and root systems have a greater rate of water use and efficiency and growth which may persist for some time even as stress develops, because larger canopies and above-ground growth support greater root growth, and vice versa. However, when soil water is depleted to a point where extensive root systems cannot provide a benefit (because there is little or no water remaining), the initially vigorous genotypes with relatively high water use would be expected to lose an advantage progressively.

There was very limited genetic variation for traits specifically affecting growth under water stress. If most genotypes had a relatively lower rate of growth under water stress, the relative final yield of genotypes under water stress would be largely determined by differences in growth rates expressed when there was limited water stress. While some genetic variation in traits affecting growth under water stress may exist in sugarcane and related germ-plasm, large deviations in the expression of these traits may have occurred in none or very few genotypes.

5. Physiological mechanism of sugarcane during water stress condition

ABA, regulatory signaling molecules, implicated to stomata closure, reduction of leaf and stem growth, production of deeper root system, higher root and shoot hydraulic conductivity, assimilate remobilization, induction of senescence, maintenance of turgor pressure, expression of antioxidant proteins and seed dormancy [36]. There is concomitant relationship between ABA and stomatal conductance and transpiration rate, by increasing H_2O_2 raises cytosolic Ca^{2+}, concentration in guard cells [37] and by reducing ROS generation and the expression of antioxidant enzymes under stress conditions [38]. Additionally, ABA-mediated responses involve cis-acting elements as dehydration response and trans-acting factors as physiological and stress response regulators that induce ird29A gene, ScbZIP29 and ScbZIP31, ScbZIP21, ScbZIP24, ScbZIP70, and ScbZIP79 for controlling the gene expression [39] that regulates the water status in bundle sheath cells [40].

Reactive oxygen species (ROS), are key regulators for growth, development, response to biotic and abiotic stimuli and programmed cell death [41] as they are the byproducts of metabolic reaction of plants [42]. The exposure of the plant for drought for longer period of drought causes ROS outburst that overrides antioxidant mechanisms and damage cell membranes, DNA, and proteins and results in cell death [43] and low induce activate acclimation and defense pathways [44]. Plant antioxidant defense mechanisms are classified as enzymatic and non-enzymatic ROS scavenging. The enzymatic ROS scavenging consists of superoxide dismutase (SOD) activity and ascorbate peroxide (APX) and catalase (CAT) activities that may be genotype dependent, and display antioxidant response in sugarcane plants during stress conditions. On the other side, non-enzymatic oxidant molecules composed of ascorbic acid (AA), reduced glutathione (GSH), α-tocopherol, carotenoids, phenolics, flavonoids, and proline act as a compatible osmolyte, molecular chaperone and carbon and nitrogen reserve, and balance cytosolic pH. Increased proline is correlated to water tolerance as sugarcane acts as a component of antioxidant defense system rather than an osmoregulator.

The most oxidative stress symptom is peroxidation of lipids, which includes O_2 molecules that are originated from photosystem II and are incorporated into plasmid membranes and catalyzed by lipoxygenase (LOX) into LOOH (lipid hydroperoxide) causing membranes vulnerable to fragmentation and leading to a cascade of damaging events [45]. The product malondialdehyde (MDA) and thiobarbituric acid reactive substances (TBARS) cause changes in cell membrane properties, such as fluidity, ion transport, and enzyme activity [46] and are accepted as a marker of oxidative stress in plants [47]. A low level of lipid peroxidation revealed the water stress tolerance in sugarcane so it can be the best parameter to identify water stress tolerant sugarcane varieties.

6. Gene related to expression of water stress response in sugarcane

Several studies has been conducted to better understand the molecular basis of physiological responses of sugarcane under stress condition. Based on ABA-dependent, high throughput gene expression under water stress focused in extensive signal transduction networks that involve various transcription factors, protein kinases, and phosphates [48]. Expressed sequence tags (ESTs) are important and breakthrough tools for identifying sugarcane genes and assessing their function because of their complex genome size of 7440 Mb. Sugarcane Expressed Sequence Tags Sequencing project (SUCEST) sequenced over 238,000 ESTs from different sugarcane tissues and cultivars, and grouped them into 43,000 Sugarcane Assembled Sequences (SAS) [49] that provides important transcriptome studies [50]. The study conducted by Li et al. used cDNA array for studying the gene expression profile in sugarcane leaves subjected to water stress conditions. However, genes involved in cellular metabolism (cell wall, amino acid, lipid, and protein metabolism), signal transduction (transcription factors, hormone signaling proteins, calmodulins, and kinases), stress response (heat shock proteins and peroxidase) showed substantial similarity in expression under different experimental conditions, the expression pattern of genes varies on the intensity of water deficit condition.

The experiment to correlate the gene expression and drought tolerance of genotypes SP83–5073 (water stress tolerant) and SP90–1638 (sensitive) by use of macro-array 9 [51] from leaf library of SUCEST suggested that 93.3% expressed genes up-regulated in tolerant cultivars, where 36% of expressed gene were repressed un sensitive plants including heat shock proteins and genes involved in photosynthesis which is corroborated to morpho-physiological data. The interesting finding of this experiment opens a new research field to unravel the hitherto unknown genetic mechanism underpinning water stress tolerance in sugarcane. The fact of expression of antisense transcripts in sugarcane under water stress caused more complex in gene regulation under stress conditions [52] so, for instance, expression of miRNAs analysis done to cope with growth phases into different intensities of stress [26]. The gene expressed in field conditions provided a different set of genes and expression profiles as compared to glass-house conditions [53]. Since there are unclear remarks of drought stress gene and sucrose accumulation [10] caused the complexity in both phenomenon. Despite the sugarcane transcriptome response, [10] indicated the strong correlation between the expression of water stress-induced genes and the sequence of dehydrin (late embryogenesis abundant proteins for protection of cellular membranes and organelles during dehydration) so it can be used as molecular marker for water stress response in sugarcane [53]. On other hand, from root samples [54] detected gene coding proteins with protection function (chaperones, heat shock proteins, antioxidants enzymes, and protease inhibitors protein and ABA response (trehalose-phosphate synthase and serine/threonine kinase receptors) that are responsible for water stress protection and adaptation mechanism. Additionally, de Silva et al. verified water channel proteins, aquaporins with isoform PIP1-1, NIP3-1, and SIP1-2 in root were exclusively up-regulated in tolerant varieties for stress avoidance mechanisms in sugarcane [55–58].

Different genes encoding GAPDH, ά-tubulin and histone H1, eEF-1 ά, Eukaryotic initiation factor 4 ά, CUL (cullin), CAC (clathrin adaptor complex), APRT (adenine phosphoribosyltransferase) and TIPS-41 (Tonoplastic Intrinsic Protein 41) are good candidate genes for normalization in various stress caused by hormones, abiotic and drought. Among them, *CAC + APRT, GAPDH + eEF1*, and *CAC + CUL* are the most reliable genes for normalization of sugarcane under stress.

There is a lack of mutant or genetic lines available to confirm the gene expression that is identified by transcriptomic analysis. The translatability of glasshouse pot study to field is very little known so further reverse or forward genetic studies are the impetus for functional assessment and linking them.

7. Conclusion

Despite the numerous studies conducted on behalf of water stress conditions in sugarcane, the result was not significantly remarked. The response and expression shown by the sugarcane at a different intensity, location and genotypes varies and confused the exact reason and corroborate to another consequential sequence, it's all due to its complexity in genome structure. The protein dehydrin encoding gene was finally named as paradigm molecular marker for analysis of water stress in sugarcane. The morphological, physiological study cannot resolve the problem of water stress and developing tolerant varieties so the mutant line generation was necessary for comparing or studying the water stress tolerance. Besides, some *CAC + APRT, GAPDH + eEF1*, and *CAC + CUL* are providing reliable genes for normalization of sugarcane under stress. So further study in greenhouse and field conditions should be translated and the concept of reverse and forward genetics studies should be thoroughly implemented for developing stress-tolerant sugarcane varieties.

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