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Enhancing Abiotic Stress Tolerance in Banana (Musa Spp.) Using In Vitro Methods and Conserving Germplasm

Divya Prakash Ojha[1,*]

Abstract

Both biotic and abiotic stressors are the main factors limiting banana output. Research on biotic stressors in bananas has received enough attention globally, whereas abiotic stressors receive less attention. Water availability, salinity, and a wide variety of temperatures are the main problems that must be resolved to maintain banana farming in the twenty-first century. Samples were introduced with water, salt and heat stresses in invitro condition. Under different stress and concentration different results were obtained as follows, for heat stress 45°C showed the optimal growth, for salinity stress 100mM NaCl concentration have shown the most growth and for water concentration at 30 g/L PEG best growth was observed. The study will be helpful in providing knowledge and information on studies related to abiotic stresses on horticultural and different crops. This study will be helpful in production of GMOs of banana and different species challenging abiotic stresses as well as in germplasm conservation. Looking at the current scenario of global warming introducing different types of abiotic stresses this study will be helpful in production of such crops.

Keyword: Banana, abiotic stresses, germplasm conservation, survival rate, *invitro* Propagation, PEG, NaCl

INTRODUCTION

Banana is a long, edible fruit — botanically a berry – produced by numerous species of big monocotyledonous herbaceous flowering plants of the genus Musa. Sreedharan et al. (2013) state that bananas are members of the Musaceae family and genus Musa. In some locations, cooking bananas are called "plantains" to differentiate them from desert bananas. Although the fruit's size, color, and firmness vary, it is typically elongated and curved, with soft, starchy flesh that is coated in a rind. Bananas (Musa spp.) and plantains (Musa spp.) are among the first crop plants to be domesticated by humans. Although these tropical fruiting plants are usually referred to as trees, they are actually large herbaceous plants with no wooden stems. Their massive, oblong, bright green leaves are produced by strong, tall stalks. The large, elongated green or yellow fruit appears after the beautiful blossoms in the spring.

***Author for Correspondence**
Divya Prakash Ojha
E-mail: 02dprakashojha08@gmail.com

[1]**Student**, Department of Biotech, Meerut Institute of Engineering and Technology

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Musa acuminata, Musa balbisiana and Musa paridisiacsa are some of the wild species that produce almost all modern edible seedless (also known as parthenocarp) bananas, depending on their genetic composition. Bananas (which include bananas, plantains, and cooking bananas) are a high-value commercial as well as a low-value subsistence food crop. It was one among the first crop plants to be domesticated. Originally, they were tailored to climatic conditions in the humid tropics and the broad subtropics. Bananas were first cultivated in Indochina and Southeast Asia, where wild Musa species abound (Musa acuminata AA and Musa balbisiana BB). Banana provide Vitamin B6, Vitamin C, Fiber, Potassium, Manganese.

Certain physical, chemical, and biological elements are necessary for the growth, development, and commercial output of horticultural plants. Any departure from these guidelines could result in aberrant metabolic alterations in the plant, which would reduce crop output and stress the plant. Abiotic stressors (wind, touch) are caused by nonliving factors such light (high light, UV, darkness), water (deficient, desiccation, flooding), salt, temperature (frost, cold, heat), nutrients (nutrient imbalance), oxidative stress, hypoxia, and physical effort. These environmental factors lead to non-infectious diseases. Extremes in water, temperature, salts, nutrients, and light can all cause stress to plants. The soil-plant atmosphere continuum (SPAC) is changed by abiotic stress, which may lead to decreased plant performance and yields. Plants detect these pressures and respond by improving their tolerance or avoiding them through morphological changes and/or physiological, biochemical, and molecular mechanisms. These tactics cause physiological and developmental changes in the tree, which affect its production and growth. Unfavorable conditions of these abiotic and biotic variables have a negative impact on horticultural crops' plant growth, development, and full expression of genotype potential yield 91% of the world's land is under stress, and only 9% of it is suitable for growing grains. Abiotic stressors have the ability to reduce yield by 65–87 percent. The plant's tolerance to various stimuli is determined by its species, genotype, and developmental stage. Avoidance (preventing the plant from being exposed to the stress), tolerance (enabling the plant to survive the stress), and acclimatization are the three stress resistance mechanisms (altering the physiology in response to the stress). A variety of abiotic pressures has a significant impact on banana production in key growing regions, endangering the livelihoods of smallholder farmers in developing countries. The crop is very subject to abiotic stressors such as drought, temperature, nutrition, and salinity because of the little genetic variety in this commercial plantation. However, there is a great amount of variety available all across the world, and we believe that optimal research and use of this diversity can help to alleviate concerns with stress, pests, and diseases. The Laboratory for Tropical Crop Improvement hosts the world's largest collection of banana biodiversity (>1500 accessions) in the International Transit Centre of Biodiversity (KU Leuven).

Plants ability to acclimate to various forms of stresses is influenced by the duration and rate at which stress occurs. Abiotic stress restrictions have been overlooked by present technology for increasing banana and plantain productivity are the most common abiotic stresses.

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Plants ability to acclimate to various forms of stresses is influenced by the duration and rate at which stress occurs. Abiotic stress restrictions have been overlooked by present technology for increasing banana and plantain productivity (Wairegi *et al*. 2010). Salinity (Ravi & Uma, 2011), heat (Wairegi, van Asten, Tenywa, & Bekunda, *et al.,* 2010), and drought (van Asten, Fermont, & Taulya, *et al.,* 2011; Taulya, van Asten, Okech, & Gold, *et al.,* 2006) are the most common abiotic stresses.

REVIEW OF LITERATURE

Banana

Rich in minerals, phosphorus, calcium, potassium, and vitamin C, bananas are a popular food in India because they are available year-round, are produced in large quantities, and are well-liked by customers.

Additionally, it is necessary for the synthesis of tannin, latex, and fiber. The banana is Asia's and the Pacific's most popular fruit. In Bangladesh, Vietnam, Indonesia, Thailand, the Philippines, and the South Pacific Island nations, it is the most significant fruit. In the agricultural economies of Australia, Malaysia, Taiwan, Sri Lanka, and South China, bananas also play a significant role. The export of bananas generates significant revenue for Taiwan and the Philippines. Domestic marketplaces are where the vast majority of the bananas grown in our nation are traded and consumed. Numerous biotic and abiotic elements are to blame. for India's low banana output and yield.

Concept of Micropropagation of Banana

The lengthy cultivation cycle and the requirement for broad areas due to the size of the plants make field experiments to choose genotypes of drought-tolerant bananas challenging. In vitro selection is therefore beneficial since it is quicker and requires less plant material, enabling the quick multiplication of chosen genotypes ,condition control, and the identification of variations in decreased.

Abiotic Stress

The basic elements of the environment that affect plant productivity and distribution are known as abiotic variables. In nature, plants are constantly challenged by adverse abiotic environmental conditions such as drought, heat, cold, nutrient deficiencies and excess salt or toxic metal levels in the soil. These abiotic stressors have a detrimental impact on agricultural productivity and restrict the global use of arable lands. Therefore, it is essential for global food security to comprehend how plants interpret stress signals and adjust to unfavorable environmental situations. Abiotic stressors alter cellular functions broadly and have an impact on many facets of plant physiology [1]. The deleterious alterations in protein structure and membrane fluidity brought on by heat or cold stress, as well as the disturbances in enzyme kinetics and molecular interactions brought on by toxic ions, are examples of non-adaptive reactions that merely reflect damage produced by a stressor. Crop improvement may be possible because many of the changes are adaptive reactions that result in greater stress tolerance. Repairing damage caused by stress, restoring cellular homeostasis, and adjusting growth to levels appropriate for the specific stress situation are all steps in the adaptive response In our project, we will focus on three key abiotic stresses that, when combined, affect horticulture crops during their production. The following are the principal abiotic stresses: Water stress, heat stress and salinity stress.

Water Stress

The plant is said to be under water stress when water is not available to the roots or when transpiration becomes excessive. Water stress can be caused by a lack of water or a high level of salinity in the soil. Drought is defined as a lack of precipitation for an extended period of time in which any area receives annual rainfall that is less than average, whereas water logging is defined as a condition in which water is present in excess of its optimum requirement, causing anaerobic conditions around roots and plants that are unable to absorb water. There is still water in the soil solution in the case of excessive salinity, periods of inundation, and low soil temperature, but the plants are unable to absorb it, resulting in a situation known as "physiological drought". When there is water in the soil solution but plants cannot absorb it because of high salt concentrations, water logging, or low soil temperature, this is known as physiological drought. Particularly in arid and semi-arid regions, water stress disrupts the growth and development of horticulture crops, which lowers yield [2]. Plants react to water stress in a number of ways, including altered cellular metabolism and gene expression, as well as modifications to root development, production, leaf movement and shape, and plant growth. Relative water content, photosynthetic rate, stomatal conductance, and leaf water potential are all impacted by drought stress.

Finally, it causes cell death by disrupting membrane structure and permeability, as well as protein structure and function. Drought-tolerant plants have a variety of processes, including reduced water loss through stomatal conductance reduction or morphological alteration, improved water uptake through effective root systems, and osmolyte buildup. The weight of the bunch is impacted because of the influence of water deficit during the flowering stage, which restricts the number of fingers produced. Water stress occurs in plants when transpiration surpasses the rate of absorption or when the water supply to the roots is restricted. Plants under water stress have lower water potential and wall pressure, which makes it harder for them to perform normal physiological processes. Lack of water, such as drought or high salt levels, is the main cause of water stress. In dry and semi-arid regions with fluctuating precipitation, drought is frequent.

The agricultural sector is forecast to demand an additional 45 percent water by 2050, while its share is expected to fall by 10%. Rainfed agriculture continues to account for about 58 percent (80 million ha) of India's net sown area, contributing 40 percent of food grain output and supporting two-thirds of the cattle population [3,4] (Anonymous). Under stress fruit TSS, kiwi, strawberries, apricots, pear, grapes, and tomatoes all showed increases in titratable acidity, firmness, color, ascorbic acid, anthocyanins, phenols, flavonoids, and post-harvest acceptable quality.

When a plant experiences water stress, it slows down growth, photosynthesis, and other processes to preserve water As water loss rises, the color of some species leaves may shift to a blue-green or pale hue. The plant dies when the foliage wilts and the leaves fall off. Drought reduces a plant's root's water potential, causing abscisic acid accumulation and, eventually, stomatal closure. This lowers the relative water content of a plant's leaves. The amount of time it takes for drought stress to develop is determined by the soil's water holding capacity, weather conditions, plant growth stage, and plant species [5]. They discovered that compared to plants growing in sandy soils with little water-holding ability, plants growing in clay soil are less susceptible to drought stress.

If the root system is restricted, drought stress will manifest itself more rapidly. Competing roots may hinder a root system due to container size, high water tables, and compacted soils. Because leaves lose water more quickly than roots can restore it, plants with a large leaf mass relative to their root system are more vulnerable to water stress. Early excessive foliage growth and inadequate root system development make newly established orchards vulnerable to drought stress.

Plants use a variety of physiological mechanisms to adjust to water stress, including changes in the amount of chlorophyll in leaf tissue, the stability of chlorophyll and membranes, the relative water content of tissues, osmotic potential (OA), stomatal conductance, transpiration, and antioxidant defense. The translocation of assimilates to bunches is inhibited by a lack of water during finger growth.

Heat Stress

Heat stress is one of the most dangerous abiotic variables that reduces productivity and quality, resulting in significant financial losses. The morphological, anatomical, physiological, and biochemical alterations in the plant system are all affected by high temperatures. Due to a lack of understanding of heat stress at vital periods of fruit crops, success in overcoming it is limited. Bananas are thermophiles, perennial herbaceous plants that grow and produce fruit all year. As a result, the plant is usually subjected to low temperature stress in the winter, resulting in a significant reduction in yield. As a result, temperature influences plant functions such as growth and metabolism.

Reduction in growth, chlorosis, and wilting of the leaves as well as alterations in membrane integrity, loss of cell compartmentation, and impairment of photosynthesis performance impairment of enzymatic activity, miss-formation of protein synthesis, and over- accumulation of reactive oxygen species (ROS) are some of the most common cold stress symptoms. Temperature is the primary determinant of banana growth and yield under adequate water supply). Furthermore, a moderate temperature (20–30°C) is required for proper vegetative growth, fruit development, and maximum yield Temperatures below 20°C, on the other hand, have a negative impact on the phenological and ripening phases, depending on the severity of the cold and the length of exposure (Stover and Simmonds, 1987). Below 10°C, leaf emergence ceases, resulting in bunch malformations and discoloration of peel tissues, as well as abnormal fruit ripening. Furthermore, irreversible chilling damage to the entire growth was reported in banana after four hours of exposure to 4°C [6,7]. The optimum temperature is 31°C or 32°C, which is only reached in the banana growing districts during the summer months. Around 9 or 10°C, leaf emergence ceases. Each plant may yield four or five leaves every month in the summer, but only roughly half a leaf per month in the winter. The amount of growth that the plant has produced can be expressed by its dry weight. The ideal temperature for leaf appearance rate is around 10°C higher than the ideal temperature for development. Higher temperatures cause the leaves, pseudo stem, corm, and roots to use a larger percentage of the carbohydrates they produce for respiration [8,9]. As a result, there is less space for growth. Since temperature greatly affects how quickly fruit grows, bunch coverings are utilized because they are believed to warm the fruit and speed up growth.Fruit from covered bunches is more consistent than fruit from unprotected bunches, and bunch covers help lessen the temperature gradient throughout the bunch.

Heat-burn: Bright sunlight and high air temperatures (often above 38°C) are associated with sunburn damage to exposed fruit, especially the upper hands of the bunch. Care must be used when handling bunch covers if they are employed. Using coverings with a reflective coating on one side or sandwiching a protective covering, such paper, between the fruit and the cover will help prevent sunburn.

Choking: The plant appears "choked" or "rosette" when there is less space between the petioles (leaf stalks) of alternating leaves. From late winter to early spring, this is most prevalent. In August, bunches might not properly develop. The Williams variety does not experience this as frequently as the Dwarf Cavendish variety. Choked bunches yield fruit that is challenging to pack and are more prone to sunburn. Choking can also result from dehydration and high heat (over 30°C).

Salinity Stress

One of the most important abiotic stresses in the world, salinity limits plant growth in general and agricultural crop productivity in particular. Over the past ten years, salinity has damaged almost 900 million hectares worldwide [10]. One of the most prominent cultures studied in irrigated regions across the globe is the banana. Nevertheless, among other things, rising soil salinization limits its yield. Around the world, bananas are a staple crop and one of the most significant table fruits. Many environmental factors have been causing it to lose yield. One of the main abiotic factors restricting growth is salinity, which lowers the pace at which plants propagate both naturally and artificially

In contrast, the rate of plant propagation under in vitro settings has been found to be significantly higher than that under ex vitro conditions. In dry and semi-arid regions of the world today, agricultural land is becoming less fertile as soil salinity increases over time. It happens as a result of saline crop irrigation or natural processes

When cultured under appropriate and balanced nutritional conditions, normal plant development is anticipated. When one or more nutritional components (inorganic or organic) [11]. Adequately supplied (deficient or excess), aberrant expression occurs. Excess NaCl is to blame for the majority of salinity issues. It causes three types of issues, such as exceeding external osmotic pressure while maintaining internal osmotic pressure. Disturbances in the equilibrium of nutritional ions in cells may have direct harmful effects on membranes and enzymes. Furthermore, correct metabolic activities are required for plant growth. Metabolic dysfunction is caused by nutritional deprivation, osmotic stress, and ion poisoning. Under stress, plants need to produce and store a range of appropriate solutes, including sugars, polyols, and free amino acids (proline, betaine, etc.).

These serve a critical function in compensating for osmotic stress caused by salinity in a variety of species, including plants. Through micro-propagation, variations in metabolites can be seen in growing plantlets Salinity is a crucial physical condition that limits plant proliferation effectiveness in ex-vitro and in vitro. As a result, the plant tissue culture method has proven to be an effective instrument for evaluating the effects of different salts on plant growth-related characteristics.

This study provides valuable insights into the impact of abiotic stresses—heat, salinity, and water—on the growth and development of banana plants (Musa spp.). The findings reveal that optimal growth conditions under stress include a temperature of 45°C for heat stress, 100mM NaCl concentration for salinity stress, and 30 g/L PEG for water deficit stress. These results highlight the potential for identifying and developing stress-tolerant genotypes through in vitro techniques.

The research underscores the significance of leveraging biotechnological approaches, such as genetic modification and micropropagation, to address the challenges posed by abiotic stressors. This is especially important in light of the changing global climate and the loss of arable land.

Furthermore, the study contributes to germplasm conservation efforts, which are vital for sustaining banana cultivation and ensuring food security.

By advancing our understanding of plant stress tolerance mechanisms, this research paves the way for developing resilient horticultural crops capable of thriving under adverse environmental conditions. The outcomes of this study hold promise for enhancing banana productivity and can be extended to other crop species, ultimately benefiting both smallholder farmers and global agriculture.

CONCLUSION

Water availability, soil type, and plant-specific characteristics significantly influence a plant's ability to cope with drought stress. Plants growing in clay soils exhibit better resilience compared to those in sandy soils, primarily due to the higher water-holding capacity of clay. However, restricted root systems, whether due to limited container size, compacted soil, or high-water tables, exacerbate drought stress. Additionally, plants with a disproportionate leaf-to-root ratio are particularly susceptible to water stress, as their water demand exceeds the roots' replenishment capacity. Newly planted orchards are especially vulnerable, emphasizing the need for balanced growth between foliage and root systems during early stages. To counteract these challenges, plants deploy diverse physiological mechanisms, underscoring the intricate interplay between plant structure, environment, and adaptive strategies in mitigating water stress.

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