



Silicon Dioxide Nanoparticles as Osmoprotectants: A Review of their Role in Mitigating Osmotic Stress in Banana and Citrus Crops

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Abstract: Silicon dioxide nanoparticles (SiO₂-NPs) have emerged as promising abiotic stress alleviators in horticultural crops, yet their specific applications in banana (*Musa* spp.) and citrus (*Citrus* spp.) under osmotic stress remain underexplored. Osmotic stress, primarily induced by salinity and drought, disrupts water uptake, nutrient balance, and metabolic homeostasis, leading to reduced growth, yield, and fruit quality. This review synthesizes current knowledge on the physicochemical properties of SiO₂-NPs, their modes of action as osmoprotectants, and the mechanisms by which they enhance plant tolerance to osmotic challenges. We examine seed priming and foliar application strategies, highlighting how SiO₂-NPs modulate antioxidant defense systems, osmolyte accumulation (e.g., proline, soluble sugars), and membrane stability in banana and citrus. Comparative analyses of nanoparticle size, concentration, and surface modifications reveal optimal treatment parameters that maximize stress mitigation while minimizing phytotoxicity. Furthermore, the review addresses uptake pathways, translocation dynamics, and potential interactions with endogenous silicon transporters. Emerging omics-based insights into gene expression and metabolomic shifts provide a deeper understanding of SiO₂-NP-mediated stress resilience. Finally, we discuss environmental safety considerations, regulatory challenges, and future research directions, including field-scale trials and integration with precision agriculture. By consolidating recent advances and identifying critical knowledge gaps, this review offers a practical framework for the development of SiO₂-NP-based interventions to sustain banana and citrus production under increasingly prevalent osmotic stress conditions.

Keywords: Silicon dioxide nanoparticles, Osmotic stress, Banana (*Musa* spp.), Citrus (*Citrus* spp.), Osmoprotectants, Antioxidant defense, Agricultural nanotechnology

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INTRODUCTION

Osmotic stress, encompassing both drought- and salinity-induced water deficits, represents one of the most pervasive abiotic constraints on global horticultural productivity. In many tropical and subtropical regions, including major banana (*Musa* spp.)– and citrus (*Citrus* spp.)–producing areas, fluctuations in precipitation patterns, soil salinization, and erratic irrigation regimes create environments in which plants continually confront disruptions in water uptake and cellular turgor maintenance. Under such conditions, the osmotic potential of the rhizosphere declines, impeding root water absorption and precipitating a cascade of detrimental physiological and biochemical alterations. These include the overproduction of reactive oxygen species (ROS), membrane lipid peroxidation, impaired photosynthetic performance, nutrient imbalances, and ultimately reductions in vegetative growth, yield, and fruit quality (Munns & Tester, 2008; Farooq et al., 2009).

Traditionally, agronomic and genetic interventions such as the use of tolerant rootstocks, deficit irrigation management, and exogenous application of osmoprotective compounds have been deployed to ameliorate osmotic stress in banana and citrus. However, these approaches often deliver inconsistent field performance and may carry environmental or economic drawbacks. In this context, nanobiotechnology offers a novel frontier: the deployment of silicon dioxide nanoparticles (SiO₂-NPs) as abiotic stress mitigators. Silicon, long recognized for its beneficial ‘quasi-essential’ role in monocot and dicot stress physiology, forms the basis for a burgeoning body of research demonstrating that nanostructured silica can penetrate plant tissues more effectively, interact at the molecular level, and trigger enhanced defense responses relative to bulk Si sources (Ma & Yamaji, 2006; Wang et al., 2014).

Silicon dioxide nanoparticles exhibit unique physicochemical characteristics high specific surface area, tunable porosity, and facile surface functionalization that render them particularly suited to agricultural use. When applied via seed treatment, soil amendment, or foliar spray, SiO₂-NPs have been shown to fortify cellular antioxidant machinery, facilitate osmolyte (e.g., proline and soluble sugar) accumulation, and stabilize membrane integrity under water-deficit conditions (Raja et al., 2017; Rizwan et al., 2019). Yet, while studies on SiO₂-NPs have proliferated in model species and staple cereals, investigations focused on the economically critical banana and citrus crops remain comparatively sparse and fragmented.

Banana, cultivated extensively across humid tropical lowlands, is particularly vulnerable to salinity stress due to its shallow rooting depth and high transpiration rates (Sharma, 2002). Likewise, citrus species, which dominate subtropical fruit markets worldwide, suffer marked declines in photosynthetic efficiency and fruit set under saline irrigation and prolonged drought episodes (Flexas et al., 2006). The integration of SiO₂-NP technology could therefore represent a powerful tool to sustain and enhance production of these fruit crops under escalating osmotic constraints driven by climate change and soil degradation.

This review aims to synthesize and critically evaluate current knowledge on the application of silicon dioxide nanoparticles for osmotic stress mitigation in banana and citrus. We begin by detailing the physicochemical properties and synthesis methods of SiO₂-NPs pertinent to plant applications. Next, we examine the mechanisms by which these nanoparticles are taken up and translocated within banana and citrus tissues, highlighting both apoplastic and symplastic pathways. We then delve into the molecular and physiological bases of stress amelioration specifically, the enhancement of antioxidant defenses, osmolyte biosynthesis, and maintenance of membrane stability. Comparative analyses of seed priming versus foliar application strategies are provided, along with discussions of nanoparticle size, concentration, and surface modifications as determinants of efficacy and phytotoxicity. Subsequently, we explore emerging omics-based insights transcriptomic, proteomic, and metabolomics into SiO₂-NP-mediated stress responses. Finally, we address environmental safety considerations, regulatory frameworks, and the critical knowledge gaps that must be bridged to enable scalable, field-level adoption. Through this comprehensive appraisal, we aim to furnish researchers and practitioners with a cohesive framework for leveraging SiO₂-NPs to bolster banana and citrus resilience in an era of mounting osmotic stress (Avestan et al. 2019).

PHYSICOCHEMICAL PROPERTIES AND SYNTHESIS OF SIO₂-NPS

Nanoparticle Characteristics

Silica nanoparticles (SiO₂-NPs) are among the most widely studied nanomaterials due to their exceptional physicochemical stability, biocompatibility, and ease of functionalization. These nanoparticles exhibit a high surface-to-volume ratio, which enhances their reactivity and surface interactions. Typically, silica NPs display uniform spherical morphology, although their shape can be tuned to form rods, tubes, or irregular particles depending on the synthesis method.

Key characteristics include

- **Particle size:** Generally ranges between 10 to 100 nm. Smaller sizes offer higher reactivity but can affect biocompatibility.
- **Surface area:** Specific surface areas can reach up to 1000 m²/g, particularly in mesoporous silica structures, enabling high loading capacities for drug delivery and catalysis.
- **Pore diameter and structure:** Controlled between 2 to 50 nm. The pore volume and geometry (hexagonal, cubic, etc.) can be fine-tuned using templating agents.
- **Surface chemistry:** Rich in silanol groups (–Si–OH), which can be modified with organic or inorganic functional groups to tailor properties such as hydrophobicity, charge, and reactivity.
- **Zeta potential:** Typically negative in aqueous solution due to the deprotonation of surface silanols, influencing colloidal stability and interaction with biological membranes.

These intrinsic characteristics make silica NPs highly suitable for applications in drug delivery, catalysis, biosensing, and environmental remediation (Al Murad et al. 2020).

Synthesis Methods

The synthesis of SiO₂ nanoparticles can be tailored to achieve specific size, porosity, and surface functionality. Below are the primary methods:

Stöber Method:

The Stöber method, introduced in 1968, is a widely used sol–gel-based technique for synthesizing monodisperse, non-porous silica spheres ranging from the nanometer to micrometer scale. This method operates on the principle of controlled hydrolysis and condensation of alkoxysilanes most commonly tetraethyl orthosilicate (TEOS) in an alcohol medium such as ethanol or methanol, with water and ammonia acting as the hydrolyzing agent and base catalyst, respectively. During the process, TEOS undergoes hydrolysis to form silanol groups (Si(OH)₄), which subsequently condense to form siloxane bonds (Si–O–Si), resulting in the formation of a solid silica network. The reactions can be represented as follows: hydrolysis – $\text{Si}(\text{OC}_2\text{H}_5)_4 + 4\text{H}_2\text{O} \rightarrow \text{Si}(\text{OH})_4 + 4\text{C}_2\text{H}_5\text{OH}$; and condensation – $\text{Si}(\text{OH})_4 \rightarrow \text{Si–O–Si} + \text{H}_2\text{O}$. One of the major advantages of the Stöber method is its ability to yield highly uniform silica particles with excellent monodispersity. Additionally, the size of the resulting particles can be finely tuned by adjusting the molar ratios of TEOS, ethanol, ammonia, and water. The method is also noted for its simplicity and reproducibility, making it suitable for laboratory-scale and industrial applications. However, a notable limitation is that the particles produced are typically non-porous, and achieving specific porosity or surface

functionalities requires further post-synthetic modifications (Hasan et al. 2019).

Sol–Gel Route with Templating

The sol–gel route with templating is an advanced synthesis method employed to produce mesoporous silica nanoparticles (MSNs) featuring highly ordered and tunable pore structures. This process involves the use of surfactants such as cetyltrimethylammonium bromide (CTAB) or triblock copolymers like P123 as templating agents that self-assemble into micelles or liquid-crystalline phases, which serve as soft templates. Silica precursors like tetraethyl orthosilicate (TEOS) or tetramethyl orthosilicate (TMOS) co-condense around these templates through hydrolysis and condensation reactions. Upon formation of the silica framework, the organic surfactant templates are removed via calcination or solvent extraction, resulting in mesoporous structures with well-defined pore sizes typically ranging from 2 to 10 nm. These MSNs often display ordered morphologies such as hexagonal (e.g., MCM-41), cubic (e.g., SBA-1), or worm-like arrangements. The resulting materials exhibit high surface area, large pore volume, and uniform particle distribution, making them highly suitable for applications such as drug delivery, catalysis, and adsorption. The major advantages of this method include precise control over porosity and morphology, uniform particle size, and the ability to incorporate functional molecules within the mesopores, enhancing the utility of MSNs in both industrial and biomedical fields (Kaur et al. 2018).

Green Synthesis

Green synthesis of silica nanoparticles (SiO₂-NPs) has gained considerable attention as an eco-friendly and sustainable alternative to conventional chemical methods, primarily focusing on minimizing environmental toxicity and enhancing biocompatibility. This approach utilizes natural raw materials such as plant extracts derived from *Aloe vera*, *Azadirachta indica* (neem), and *Ocimum sanctum* (tulsi), which are rich in polyphenols, flavonoids, and natural reducing agents. Additionally, biopolymers like chitosan, starch, or alginate may serve as stabilizing or capping agents during synthesis. The mechanism typically involves the reaction of silica precursors like sodium silicate or tetraethyl orthosilicate (TEOS) in aqueous media, wherein the biomolecules not only reduce the precursor but also stabilize the forming nanoparticles, thereby controlling particle size and preventing aggregation. One of the key advantages of green synthesis is its eco-friendly nature, as it avoids the use of harmful solvents and high energy input, often being conducted at ambient temperatures. Moreover, the resulting nanoparticles are highly suitable for biomedical and therapeutic applications due to their improved biocompatibility. However, despite its sustainability, green synthesis may face challenges related to achieving uniform size distribution and monodispersity, and its scalability and reproducibility remain areas requiring further optimization for large-scale applications (Khan et al. 2019).

UPTAKE, TRANSLOCATION, AND DISTRIBUTION IN BANANA AND CITRUS

Root Absorption Pathways

The uptake of silica nanoparticles (SiO₂-NPs) through plant roots is a highly regulated and complex physiological process, particularly in species like banana (*Musa spp.*) and citrus (*Citrus spp.*), which are

known to exhibit variable silica accumulation behavior. Upon application in the rhizosphere, SiO₂-NPs enter the root system primarily through two distinct pathways: the apoplastic and symplastic routes. In the apoplastic pathway, the nanoparticles diffuse passively through the extracellular spaces and cell walls without crossing the plasma membrane. This mode of movement allows for relatively unregulated transport but may be restricted at the Casparian strip in the endodermis, necessitating a switch to symplastic transport.

The symplastic route, by contrast, involves the regulated entry of SiO₂-NPs or their ionic derivatives particularly monosilicic acid (H₄SiO₄) across the plasma membrane into the cytoplasm, followed by movement from cell to cell via plasmodesmata. This pathway is facilitated by specific membrane transport proteins, most notably Lsi1 (Low Silicon 1) and Lsi2, which are responsible for the influx and efflux of silicic acid in many plant species. Lsi1 is generally expressed in root epidermal and cortical cells and functions as a channel-type transporter, while Lsi2 is an active transporter located in the endodermis and pericycle. In banana and citrus, recent studies suggest the presence of homologous genes encoding similar transporters, although their expression and activity may vary with species, developmental stage, and stress conditions. These coordinated mechanisms ensure controlled uptake and radial transport of silica from the root surface to the vascular tissues for systemic distribution.

Foliar Entry Mechanisms

In addition to root-mediated absorption, foliar application of SiO₂-NPs represents a promising alternative route for silica supplementation, especially under conditions where root uptake is compromised. The entry of nanoparticles through the leaf surface occurs via multiple mechanisms, depending on their size, surface charge, formulation, and environmental factors. The two major pathways include cuticular penetration and stomatal uptake. The leaf cuticle, although generally considered a barrier, contains nano- and micro-pores that can permit the diffusion of ultrafine particles, especially when the surface tension of the applied solution is reduced. Surfactants are commonly added to foliar formulations to enhance wettability and promote the spreading and adhesion of SiO₂-NPs on the leaf surface, thereby improving their chances of penetration.

Stomatal uptake is another important route, where nanoparticles can directly enter the substomatal cavity when stomata are open, typically during daylight hours under favorable humidity conditions. Once inside, the particles or their ionic forms may be translocated through the apoplast or symplast into deeper mesophyll tissues. In citrus leaves, which possess a thick waxy cuticle and deeply embedded stomata, the efficiency of foliar uptake may vary depending on nanoparticle formulation and leaf age. Similarly, in banana plants with large, moisture-retaining leaves, foliar application is effective, especially when combined with surfactant agents that aid in particle dispersion and adherence. Overall, foliar entry mechanisms offer a practical route for targeted delivery of SiO₂-NPs in both crops, enhancing bioavailability while bypassing soil interactions.

Tissue Localization

Following uptake via roots or leaves, the distribution and accumulation of SiO₂-NPs within plant tissues determine their physiological effects and potential benefits. Advanced imaging techniques such as

Transmission Electron Microscopy (TEM), Scanning Electron Microscopy (SEM), and Energy-Dispersive X-ray Spectroscopy (EDX) have been instrumental in revealing the intracellular and extracellular localization of silica nanoparticles in banana and citrus plants. In roots, SiO₂-NPs are often found deposited in the cortical apoplast, especially in intercellular spaces and cell walls, suggesting that much of the initial accumulation occurs before xylem loading. In species where symplastic movement is dominant, particles may be observed within vacuoles or associated with vesicle-mediated endocytosis pathways, indicating active internalization by root cells.

Once translocated into the xylem, silica nanoparticles can move upward with the transpiration stream, reaching aerial organs such as stems, leaves, and fruits. In the vascular tissues, they are often localized in or near xylem vessels, either as discrete particles or as precipitated silica. In leaf tissues, TEM imaging has identified their presence in mesophyll cells, chloroplasts, and near cell walls, potentially contributing to structural reinforcement and stress resistance. Furthermore, in fruit tissues, silica deposits have been observed in parenchyma cells, often correlating with improved mechanical properties and reduced pathogen susceptibility.

Elemental mapping techniques such as X-ray fluorescence (XRF) and EDX spectral analysis provide corroborative evidence for silicon accumulation in specific tissues and cellular compartments. These studies show that SiO₂-NPs do not distribute uniformly but rather accumulate preferentially in sites of active metabolism, transport, or mechanical stress. The spatial distribution also depends on the mode of application, plant species, growth stage, and environmental conditions. The precise localization of SiO₂-NPs is therefore critical to understanding their role in alleviating osmotic and other abiotic stresses in banana and citrus cultivation.

MECHANISMS OF OSMOTIC STRESS ALLEVIATION

Osmotic stress, primarily induced by drought, salinity, or high evaporative demand, severely impacts plant physiological and metabolic functions by disrupting water balance and inducing oxidative damage. The application of silica nanoparticles (SiO₂-NPs) has shown promising results in alleviating such stress conditions in banana (*Musa* spp.) and citrus (*Citrus* spp.), owing to their ability to modulate key biochemical and structural pathways. These mechanisms are primarily attributed to improvements in antioxidant defenses, osmotic regulation, and membrane stabilization.

Enhancement of Antioxidant Defense

One of the primary consequences of osmotic stress is the excessive generation of reactive oxygen species (ROS), including superoxide radicals (O₂^{•-}), hydrogen peroxide (H₂O₂), and hydroxyl radicals (•OH), which cause oxidative damage to cellular structures. SiO₂-NPs play a critical role in enhancing the plant's innate antioxidant defense system by upregulating the activities of enzymatic antioxidants such as superoxide dismutase (SOD), catalase (CAT), and peroxidases (POD). SOD catalyzes the dismutation of superoxide radicals into hydrogen peroxide, which is subsequently broken down into water and oxygen by CAT and POD, thus detoxifying ROS and reducing oxidative stress.

Experimental evidence from both banana and citrus plants indicates a significant increase in these antioxidant enzyme activities following the application of SiO₂-NPs under stress conditions. This

biochemical upregulation leads to a marked reduction in malondialdehyde (MDA) levels, a key indicator of lipid peroxidation and membrane damage. Lower MDA content suggests that lipid membranes are better protected from oxidative degradation, preserving cell functionality. The nanoparticles may also act as indirect signaling molecules, activating stress-responsive genes and triggering transcriptional cascades related to ROS detoxification. Thus, SiO₂-NPs serve not only as structural additives but also as functional mediators of stress tolerance through oxidative stress regulation.

Osmolyte Accumulation

Under osmotic stress, plants accumulate small organic solutes known as osmolytes, which help maintain cell turgor and enzyme activity despite low water availability. The application of SiO₂-NPs has been shown to significantly enhance the synthesis and accumulation of key osmolytes, including proline, glycine betaine, and soluble sugars in banana and citrus crops. These osmoprotectants function by stabilizing proteins and membranes, scavenging free radicals, and adjusting the cellular osmotic potential, thereby facilitating water retention and physiological activity.

Proline, in particular, is a multifunctional osmolyte that not only contributes to osmotic adjustment but also acts as a ROS scavenger and metal chelator. SiO₂-NP treatment enhances proline biosynthesis by upregulating the expression of pyrroline-5-carboxylate synthetase (P5CS), a key enzyme in the proline biosynthetic pathway. Similarly, increased glycine betaine accumulation is associated with the protection of photosynthetic machinery and membrane stabilization. Soluble sugars such as sucrose and glucose serve dual roles as osmotic balancers and metabolic energy sources, crucial for stress recovery and growth resumption.

These osmolytes act in synergy to buffer osmotic pressure, reduce cellular dehydration, and maintain biochemical homeostasis under water-deficit conditions. The enhanced osmolyte accumulation mediated by SiO₂-NPs thus represents a crucial adaptive strategy for stress mitigation and survival.

Membrane Stability

Membrane systems, particularly the plasma membrane and chloroplast envelope, are highly vulnerable to osmotic stress, leading to increased electrolyte leakage, loss of compartmentalization, and impaired ion regulation. SiO₂-NPs contribute to maintaining membrane integrity by reinforcing structural stability and minimizing permeability under stress conditions. The nanoparticles may interact with membrane-associated proteins or form a protective barrier on the cell surface, reducing the rate of water loss and ion efflux.

Empirical studies have shown a significant reduction in electrolyte leakage in banana and citrus leaves treated with SiO₂-NPs under salinity or drought stress. This suggests that SiO₂-NPs enhance the selective permeability of membranes, maintaining ionic homeostasis and cell viability. Furthermore, microscopic analyses such as transmission electron microscopy (TEM) reveal better-preserved chloroplast structures with intact thylakoid membranes and fewer signs of swelling or distortion in nanoparticle-treated plants. This preservation is critical for maintaining photosynthetic efficiency and reducing photoinhibition under stress.

By enhancing membrane rigidity and resilience, SiO₂-NPs also facilitate better nutrient uptake and ion

transport, which are essential for metabolic activity during osmotic challenges. The stabilization of membrane systems ensures sustained physiological processes and confers a level of stress endurance critical for crop productivity in arid and semi-arid environments.

APPLICATION STRATEGIES AND OPTIMIZATION

The effectiveness of silicon nanoparticles (SiO_2 -NPs) in alleviating osmotic stress and promoting crop resilience depends greatly on how they are applied. In banana and citrus crops, where environmental stressors like salinity, drought, and high temperature can critically impair growth and yield, strategic delivery of SiO_2 -NPs is essential to ensure optimal uptake and bioavailability. Various application methods ranging from seed priming to soil incorporation and foliar spraying have been developed and optimized to align with specific physiological needs and stress stages of the plant.

Seed Priming

Seed priming is an effective pre-sowing treatment involving the controlled exposure of seeds to SiO_2 -NP suspensions for a limited duration. This technique enhances the physiological and metabolic preparedness of seeds to withstand adverse environmental conditions upon germination. When banana or citrus seeds or plantlets are primed with SiO_2 -NPs, especially under saline substrates, there is a noticeable increase in germination percentage, seedling emergence rate, and early root-shoot elongation. The nanoparticles may facilitate water uptake, modulate hormonal activity (particularly gibberellins and abscisic acid), and enhance early antioxidant enzyme activities, giving the seedling a competitive edge in stress-prone soils.

Studies have demonstrated that the pre-treatment of seeds with SiO_2 -NPs (typically in concentrations of $10\text{--}50\text{ mg}\cdot\text{L}^{-1}$) results in stronger root systems, greater cell viability, and improved chlorophyll content in early developmental stages. This is especially important for citrus rootstocks and tissue-cultured banana plantlets, which are sensitive to initial transplant shock. By "preconditioning" the seeds, priming acts as a cost-effective and low-tech strategy for improving field establishment and ensuring uniform stand development under osmotic stress.

Soil Amendment

Soil application of SiO_2 -NPs represents another vital route for silicon delivery, particularly in field settings. In this approach, SiO_2 -NPs are incorporated directly into the rhizosphere, where they can serve as a slow-release source of bioavailable silicon, either through direct dissolution or microbial-assisted mineralization. This method allows for a sustained supply of silicon to the plant roots over an extended period, improving long-term stress resilience and nutrient-use efficiency.

Beyond their role in plant nutrition, SiO_2 -NPs also influence the soil microbiome. Recent research indicates that the inclusion of silica nanoparticles in soil can modulate microbial community structure, often favoring beneficial groups such as plant-growth-promoting rhizobacteria (PGPR), nitrogen-fixers, and phosphate-solubilizing bacteria. These microbes, in turn, contribute to enhanced nutrient uptake, reduced pathogen incidence, and improved soil aggregation.

In banana and citrus orchards, where soil health directly influences productivity, nanoparticle-amended soil

may lead to improved water retention, reduced sodium uptake under saline conditions, and better root colonization. However, optimizing particle size, concentration (commonly 100–300 mg·kg⁻¹ soil), and frequency of application is crucial to avoid toxicity or nanoparticle accumulation. Incorporation can be done manually, via fertigation systems, or through pelleted formulations.

Foliar Sprays

Foliar application remains one of the most practical and responsive strategies for delivering SiO₂-NPs during active growth phases or under imminent environmental stress. When sprayed on the leaves, these nanoparticles bypass the soil-root barrier, enabling rapid absorption and targeted physiological effects. The efficiency of this method depends significantly on the particle concentration, droplet size, pH of the spray solution, and presence of surfactants that enhance wettability and penetration.

Studies in banana and citrus indicate that foliar sprays with SiO₂-NP concentrations ranging from 50 to 200 mg·L⁻¹, when combined with non-ionic surfactants (such as Tween-20 or Silwet L-77), result in increased nanoparticle adhesion to the leaf surface and improved stomatal uptake. This, in turn, translates to higher tissue silicon levels, reduced leaf temperature, enhanced photosynthetic efficiency, and better tolerance to drought and salinity. Application timing is critical: pre-stress foliar sprays are most effective when administered a few days before expected stress onset, allowing plants to "prime" their antioxidant and osmotic responses.

In banana, foliar applications during early vegetative stages and just before flowering have shown the greatest impact on leaf turgor and chlorophyll retention. In citrus, repeated applications at intervals of 10–15 days during dry spells help sustain fruit size and peel integrity, especially in sandy soils with poor water retention. Thus, foliar spraying of SiO₂-NPs offers a flexible, non-invasive method to deliver micronutrients and stress-mitigating agents exactly when and where they are needed.

OMICS-BASED INSIGHTS INTO SiO₂-NP-MEDIATED RESPONSES

The advent of omics technologies including transcriptomics, proteomics, and metabolomics has revolutionized our understanding of how plants perceive and respond to external stimuli at the molecular level. These high-throughput platforms provide comprehensive insights into the mechanistic underpinnings of SiO₂ nanoparticle (SiO₂-NP)-mediated stress alleviation in plants like banana (*Musa spp.*) and citrus (*Citrus spp.*). By dissecting changes in gene expression, protein abundance, and metabolite profiles, omics approaches help identify key regulatory networks and biochemical pathways activated in response to nanoparticle treatment under osmotic stress.

Transcriptomics

Transcriptomic studies focus on the global expression patterns of genes, offering valuable insights into the molecular signaling pathways that are triggered upon SiO₂-NP exposure. Under osmotic stress conditions, plants treated with SiO₂-NPs often exhibit upregulation of stress-responsive genes involved in detoxification, water transport, and signal transduction. Notably, genes encoding Dehydration-Responsive Element-Binding proteins (DREBs) are significantly upregulated. These transcription factors are central to the regulation of abiotic stress tolerance, particularly in orchestrating downstream responses related to

osmolyte biosynthesis, ROS scavenging, and stomatal control.

In addition to DREBs, increased expression of Heat Shock Proteins (HSPs) has been observed. These molecular chaperones play a protective role by stabilizing proteins and membranes during stress-induced denaturation. The induction of HSP70 and small HSPs suggests that SiO₂-NPs may enhance the thermal and osmotic stress tolerance of banana and citrus by preserving proteome integrity. Furthermore, aquaporin genes, especially Plasma membrane Intrinsic Proteins (PIPs) and Tonoplast Intrinsic Proteins (TIPs), are differentially expressed in treated plants. These genes regulate water transport across membranes and are essential for maintaining cellular hydration under osmotic pressure. Such transcriptomic shifts highlight the priming effect of SiO₂-NPs on genetic pathways that confer resilience against abiotic stress.

Proteomics

While transcriptomics provides a snapshot of gene activity, proteomic analysis reveals the actual functional proteins present within the cell and how their abundance is modulated in response to nanoparticle exposure. Proteomic studies in SiO₂-NP-treated plants have identified increased accumulation of antioxidant enzymes, such as superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), and glutathione reductase (GR). These proteins are central to maintaining redox homeostasis and directly neutralize reactive oxygen species generated under osmotic stress.

In addition, enzymes involved in osmolyte biosynthesis, including Δ^1 -pyrroline-5-carboxylate synthetase (P5CS) for proline synthesis and choline monooxygenase for glycine betaine production, show enhanced expression and activity in SiO₂-NP-treated samples. These changes corroborate the physiological observations of increased osmolyte accumulation in treated plants. Proteomic mapping has also revealed modulation of stress-associated kinases, signal transduction proteins, and structural proteins like actin and tubulin, suggesting a broader reprogramming of cellular machinery to accommodate stress adaptation.

Moreover, differences in chloroplast-associated proteins such as Rubisco activase and chlorophyll-binding proteins indicate that SiO₂-NPs help sustain photosynthetic efficiency under stress. These findings underscore the importance of nanoparticle-mediated proteomic modulation as a key mechanism supporting improved growth and stress tolerance.

Metabolomics

Metabolomics, the study of small-molecule metabolites within biological systems, provides the most direct measure of physiological responses to environmental and treatment-induced changes. In the context of SiO₂-NP-mediated stress alleviation, metabolomic profiling reveals significant alterations in the levels of compatible solutes, phenolic compounds, and organic acids. These metabolites play critical roles in osmoprotection, antioxidant defense, and pH regulation.

Among compatible solutes, proline, glycine betaine, and soluble sugars such as sucrose and raffinose accumulate to higher levels in nanoparticle-treated banana and citrus tissues under drought or salinity stress. These molecules contribute to osmotic adjustment, membrane stabilization, and free radical scavenging. The elevated presence of phenolic acids (e.g., ferulic acid, caffeic acid) and flavonoids (e.g., quercetin, kaempferol) further highlights the role of SiO₂-NPs in inducing secondary metabolite pathways

associated with stress protection.

Additionally, organic acids such as malate, citrate, and fumarate, involved in the tricarboxylic acid (TCA) cycle, show altered levels, indicating metabolic adjustments aimed at sustaining energy production and maintaining cellular redox balance. Some studies also report increased levels of GABA (γ -aminobutyric acid), a non-protein amino acid that functions in stress signaling and ion flux regulation.

Altogether, the metabolomic data demonstrate that SiO₂-NPs orchestrate a complex and dynamic biochemical response that enhances the plant's metabolic plasticity, facilitating rapid adaptation to osmotic stress. When integrated with transcriptomic and proteomic insights, this provides a systems-level understanding of how SiO₂-NPs prime banana and citrus plants for enhanced stress tolerance and physiological performance.

ENVIRONMENTAL SAFETY AND REGULATORY CONSIDERATIONS

As the application of silica nanoparticles (SiO₂-NPs) gains traction in sustainable agriculture for improving stress resilience and crop productivity, it becomes essential to evaluate their environmental safety and regulatory compliance. Despite the numerous agronomic benefits, questions remain regarding potential phytotoxicity, ecological effects, and regulatory oversight. Responsible deployment of SiO₂-NPs in banana and citrus cultivation must be guided by a thorough understanding of their behavior in biological and environmental systems, alongside evolving national and international policy frameworks.

Phytotoxicity Thresholds

While SiO₂-NPs are generally regarded as safe and biocompatible, dose-dependent phytotoxic effects have been observed at elevated concentrations, underscoring the importance of identifying optimal dosage ranges. At low to moderate concentrations (typically 25–100 mg·L⁻¹), SiO₂-NPs enhance seed germination, root elongation, and chlorophyll biosynthesis in banana and citrus plants. However, at higher concentrations (above 200–300 mg·L⁻¹), growth inhibition, oxidative damage, and chlorosis have been reported, likely due to nanoparticle aggregation, physical blockage of stomata or vascular tissues, or excessive oxidative stress.

Germination assays and root toxicity tests in model plants have shown that excessive SiO₂-NP exposure can impede radicle emergence, reduce cell division in meristematic tissues, and disrupt root hair development. Similarly, prolonged exposure has been associated with declines in total chlorophyll and photosynthetic efficiency, indicating potential disruptions in light-harvesting complexes or oxidative imbalance. Therefore, the establishment of phytotoxicity thresholds and safe application windows is critical to avoid unintended adverse effects. Field-specific factors such as soil pH, organic matter content, plant developmental stage, and method of application should be considered to tailor nanoparticle dosing for maximum efficacy with minimal risk.

Soil and Water Dynamics

Understanding the fate and transport of SiO₂-NPs in soil and water systems is essential for evaluating their long-term environmental impact. Once introduced into the rhizosphere through soil amendments or runoff

from foliar sprays, SiO₂-NPs interact with various abiotic and biotic components. In soil, nanoparticles may persist in the solid phase, become immobilized on clay and organic matter, or undergo gradual dissolution into orthosilicic acid (H₄SiO₄) the plant-available form of silicon. This transformation is pH-dependent and typically occurs over days to weeks, depending on the surface properties and crystallinity of the nanoparticles.

While the conversion to orthosilicic acid facilitates plant uptake and reduces environmental persistence, concerns arise regarding the impact on non-target soil organisms, including earthworms, mycorrhizal fungi, and beneficial rhizobacteria. Some studies suggest that low concentrations of SiO₂-NPs may stimulate microbial activity and enhance nutrient cycling, but high concentrations or repeated applications can alter microbial community structure, reduce enzymatic activities, and disrupt trophic interactions in the soil food web.

In aquatic ecosystems, SiO₂-NPs can enter water bodies through leaching or surface runoff, where they may form colloidal suspensions, aggregate with other particles, or adsorb onto sediment surfaces. Though silica is a naturally occurring component of aquatic environments, the nano-sized form poses different physicochemical behaviors, including higher reactivity and mobility. As such, comprehensive risk assessments are required to determine safe application practices that minimize off-site transport and accumulation.

Policy Landscape

The regulatory landscape for nano-enabled agricultural inputs, including nanopesticides and nano-fertilizers, is still evolving and presents several challenges. Currently, SiO₂-NPs fall under the category of engineered nanomaterials (ENMs), and their agricultural use is subject to scrutiny under both chemical and fertilizer regulatory frameworks. However, most regulatory agencies, including the United States Environmental Protection Agency (EPA), the European Chemicals Agency (ECHA), and India's Central Insecticides Board and Registration Committee (CIBRC), do not yet have specific, harmonized guidelines for the evaluation and approval of SiO₂-NP-based products.

One major regulatory hurdle is the lack of standardized testing protocols for nanoparticle toxicity, environmental persistence, and bioaccumulation. Traditional risk assessment models developed for bulk materials may not accurately predict the behavior of nanoscale formulations. Furthermore, product registration requires extensive data on particle characterization, synthesis methods, safety profiles, and field-level efficacy—data which are often costly and time-consuming to generate.

In many jurisdictions, nano-enabled products must be registered as either conventional agrochemicals or novel materials, depending on their intended use. The absence of clear regulatory pathways has created uncertainty for manufacturers, researchers, and farmers alike. Therefore, there is an urgent need for international harmonization of nano-agriculture regulations, along with the development of guidelines for labeling, safety testing, and risk communication. Only through a science-based and transparent policy framework can the benefits of SiO₂-NPs be realized while ensuring environmental sustainability and public trust.

KNOWLEDGE GAPS AND FUTURE DIRECTIONS

Despite the promising potential of silica nanoparticles (SiO₂-NPs) in mitigating osmotic stress and enhancing productivity in banana and citrus crops, several critical knowledge gaps remain that must be addressed to enable large-scale, sustainable implementation. Future research should prioritize multidisciplinary approaches, combining nanotechnology, plant physiology, agronomy, soil science, and regulatory sciences to unlock the full potential of SiO₂-NPs in precision agriculture. Four major areas of focus are discussed below:

Field-Scale Validation

Most of the current evidence supporting the efficacy of SiO₂-NPs has emerged from controlled-environment studies, such as growth chambers, hydroponics, or small-scale greenhouse trials. While these conditions are useful for mechanistic insights, they often fail to replicate the complex and heterogeneous field conditions experienced in commercial orchards and plantations. Factors such as variable soil texture, pH, organic matter content, weather patterns, pest pressure, and irrigation regimes can significantly influence nanoparticle behavior and plant response.

To facilitate mainstream adoption, it is essential to conduct multi-location, multi-season field trials under real-world agronomic conditions. These trials should assess not only physiological and yield-related outcomes but also economic viability, labor requirements, and compatibility with existing cultivation practices. Moreover, studies in mature banana and citrus orchards where plants interact with long-standing soil microbiomes and have deep root systems are particularly important to understand nanoparticle distribution, uptake kinetics, and residual effects in perennial systems.

Long-Term Impact Studies

A key limitation of current research is the lack of long-term and lifecycle studies assessing the cumulative impact of SiO₂-NP applications on soil health, ecological balance, and crop rotation systems. Most toxicity assessments are short-term and focused on immediate physiological responses. However, chronic exposure to nanoparticles, even those deemed biocompatible, could lead to unintended consequences such as nanoparticle accumulation in soil, disruption of microbial communities, changes in nutrient cycling, or impacts on subsequent crops grown in the same fields.

To mitigate these risks, longitudinal studies should track the persistence, mobility, and bioavailability of SiO₂-NPs over multiple seasons. Investigations should also explore whether residual nanoparticles interfere with root-microbe interactions, symbiotic nitrogen fixation, or the efficacy of organic soil amendments. In banana-citrus intercropping systems or rotational agriculture, understanding how previous SiO₂-NP applications influence soil legacy effects and subsequent crop performance will be critical for sustainable land management.

Integrated Precision Agriculture

Advancing the application of SiO₂-NPs beyond traditional spraying and broadcasting methods requires integration with precision agriculture tools. Sensor-based and data-driven systems such as GPS-enabled sprayers, soil moisture sensors, and canopy reflectance tools can be employed to design site-specific

nanoparticle delivery protocols that optimize dosage, timing, and spatial coverage. Such systems would minimize wastage, reduce environmental runoff, and enhance efficiency by synchronizing nanoparticle application with plant demand and environmental conditions.

Developing real-time decision-support platforms that use remote sensing, machine learning, and big data analytics can help farmers apply SiO₂-NPs only when and where needed. For instance, early detection of osmotic stress through drone-based multispectral imaging could trigger targeted foliar sprays in affected zones. Additionally, smart irrigation systems could deliver SiO₂-NPs through fertigation channels, adjusting delivery rates based on evapotranspiration forecasts. Thus, incorporating SiO₂-NPs into the framework of integrated precision agriculture represents a promising frontier for scalable and environmentally responsible deployment.

Multifunctional Nano-Formulations

Another exciting avenue for future research lies in the development of multifunctional nano-formulations that combine SiO₂-NPs with other agro-inputs. Rather than applying silica alone, co-formulation with essential micronutrients (e.g., zinc, iron, magnesium), biostimulants, or plant defense activators could produce synergistic effects, reducing the number of inputs and improving plant resilience against multiple stresses. These smart formulations can be engineered with controlled-release properties, pH sensitivity, or stress-responsive release kinetics to enhance uptake efficiency.

Moreover, encapsulating bioactive molecules such as phytohormones (e.g., salicylic acid, gibberellins), antifungal peptides, or beneficial microbes along with SiO₂-NPs could turn them into nano-biocarriers with dual or triple functionality. For instance, a silica nanoparticle functionalized with chitosan and loaded with micronutrients could act as a growth promoter, stress mitigator, and antimicrobial agent all in one formulation. Such innovations would align well with the goals of sustainable intensification, reducing chemical load while maximizing crop productivity and resilience.

CONCLUSION

Silicon dioxide nanoparticles (SiO₂-NPs) have emerged as a promising nano-enabled solution for combating osmotic stress in high-value fruit crops such as banana and citrus. Their nanoscale dimensions, high surface area, and tunable surface properties enable them to interact effectively with plant systems, triggering a cascade of physiological, biochemical, and molecular responses that collectively enhance stress tolerance. From reinforcing antioxidant defenses and promoting osmolyte accumulation to stabilizing membranes and improving water-use efficiency, SiO₂-NPs offer a multifaceted approach to stress mitigation. Moreover, their adaptability across various application modes whether as seed primers, soil amendments, or foliar sprays allows for flexible integration into existing agronomic practices. However, to fully harness their potential, several challenges must be addressed. These include the optimization of nanoparticle synthesis for consistency and biocompatibility, in-depth elucidation of uptake and translocation pathways, long-term studies on environmental safety and soil health, and, crucially, large-scale validation under diverse field conditions. Bridging these gaps will not only enhance the scientific credibility of SiO₂-NPs but also ensure their safe and effective use in commercial agriculture. In doing so, SiO₂-NP technology can become a vital component of sustainable fruit production systems, offering

resilience against increasing water scarcity and contributing to food security in a changing climate.

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