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Y Bindiya
Assistant Professor, Department of Horticulture, Agricultural College, Bapatla, ANGRAU, Andhra Pradesh, India

TA Pallavi
M.Sc. Student, Department of Fruit Science, College of Horticulture, Anantharajupeta, Rly. Kodur, Annamaya (Dt), Dr. YSRHU Andhra Pradesh, India

Corresponding Author:
Y Bindiya
Assistant Professor, Department of Horticulture, Agricultural College, Bapatla, ANGRAU, Andhra Pradesh, India

Role of next-generation elicitors in enhancing fruit quality parameters

Y Bindiya and TA Pallavi

Abstract
In India, agricultural losses amount to approximately 20% due to diseases, 25% due to insect pests, and up to 50% due to abiotic stresses, resulting in a significant reduction in crop yields (Verma and Deepthi, 2016). The storage of certain fruits poses a significant challenge due to their high perishability and vulnerability to fungal attacks. Traditional methods involve the application of various fungicides to control postharvest decay. However, these conventional treatments often result in residues that carry potential risks to both human health and the environment. Consequently, there is a pressing need for the development of new and effective methods to prolong postharvest shelf life and manage decay in fruits. The use of elicitors has emerged as a promising approach to enhance the qualitative characteristics of fruits and bolster their resistance to diseases.

Keywords: Elicitors, fruit quality, fruit crops, salycilic acid, biotic factors, abiotic factors

Introduction
Plants face continuous exposure to diverse stress factors, categorized as either biotic (arising from bacteria, fungi, viruses, insects, herbivores, etc.) or abiotic (involving exposure to heavy metals, salinity, ozone, UV-B radiation, extreme temperature, nutrient excess or deficiency, and drought) in their natural environment. These factors result in widespread damage to plants, leading to substantial economic losses in cultivated areas. To address these challenges, various strategies are employed, including physical methods like solarization and crop rotation, chemical control using pesticides or fungicides, biological control through antagonistic microorganisms, and the development of resistant plant genotypes. However, the potential adverse effects of these conventional methods on human health, plants, animals, and the environment have raised concerns. Pesticide use is associated with the accumulation of residues in water and soil, posing risks to both human health and the environment. Similarly, physical methods like tillage can contribute to erosion problems. In light of these concerns, there is a growing demand for alternative, environmentally friendly approaches, and elicitors have emerged as an intriguing biological alternative. Early studies, such as the research conducted by Gianinazzi and Kassanis in 1974, identified synthetic elicitors like Polyacrylic acid derivatives with molecular weights of 3500 Da or lower, which induced resistance in tobacco plants against tobacco mosaic virus (TMV) or tobacco necrosis virus (TNV). Ongoing research explores various chemical elicitors, including salicylic acid, methyl salicylate, benzothiadiazole, benzoic acid, chitosan, and other compounds that influence the formation of phenolic compounds and activate numerous enzymes involved in plant defense mechanisms. These elicitors serve as promising tools to enhance plant resilience in a sustainable and environmentally friendly manner, offering an effective alternative to traditional approaches in plant protection and defense. (Andleeb Zehra *et al*., 2021; Nurul *et al*., 2020) [1, 10]. (Thakur & Sohal, 2013) [6]."

What are elicitors?
Your articulation provides a thorough understanding of elicitors and their pivotal role in plant defense mechanisms. Elicitors, as described, possess a remarkable ability to stimulate defense responses in plants, initiating crucial reactions like the synthesis of phytoalexins—antimicrobial compounds produced as a response to pathogen attacks. The significance of elicitors lies in their effectiveness at low concentrations, enabling them to activate a spectrum of plant responses. This includes triggering endogenous protective reactions and facilitating the production of diverse secondary metabolites. These secondary metabolites assume a key role in fortifying the plant's defense against pathogens and stressors. Whether of chemical or

biochemical origin, elicitors are introduced in small concentrations to living systems, such as plants, with the specific purpose of promoting the biosynthesis of bioactive compounds. This, in turn, contributes to the plant's defense mechanisms. Your description underscores the potential of elicitors as valuable tools, highlighting their role in enhancing the resilience of plants and safeguarding them from a variety of biotic and abiotic stresses.

Classification of elicitors

Abiotic elicitors

"Elicitors can be broadly classified into two main categories: chemical substances derived from non-biological sources, such as mineral salts and heavy metals, and physical factors or conditions, including exposure to light (UV-B, UV-C radiation), fluctuations in temperature (heat or cold), ultrasound exposure, and osmotic stress."

Biotic elicitors

Additionally, elicitors can be further subcategorized based on their origin into substances that come from external sources, particularly pathogens (exogenous factors), and compounds that plants synthesize in response to the actions of pathogens (endogenous factors).

Exogenous elicitors, which are typically released by microorganisms and other pathogens, include a variety of substances. This category encompasses fungal or bacterial lysates, yeast extracts, microbial enzymes, as well as polysaccharides found in cell walls, such as chitin, chitosan, and glucans. It even extends to entire fungal spores."

Endogenous elicitors, on the other hand, are compounds synthesized within the plant after the pathogenic action. This category includes polysaccharides released from plant cell walls as a result of pathogen attacks, specific intracellular proteins, and effectors synthesized in response to various types of stress, such as methyl jasmonate and salicylic acid."

Mechanism of elicitation in plant cells

A general mechanism for biotic elicitation in plants can be summarized based on the elicitor-receptor interaction. When a plant or plant cell culture is exposed to an elicitor, a series of biochemical activities is triggered." These include:

Binding of the elicitor to a plasma membrane receptor"

Upon exposure to elicitors, plants undergo altered ion fluxes across the cell membrane, involving Cl- and K+ efflux, as well as Ca2+ influx. In plants, Ca2+ transients have been recognized as second messengers in response to various environmental signals, including those from pathogens. Furthermore, elicitor-treated plant tissues display cytoplasmic acidification, plasma membrane depolarization, an extracellular pH increase, and rapid alkalization of the apoplast and outer medium in cultured plant cells. These changes suggest a proton influx from the apoplast into the cytoplasm."

Elicitor exposure leads to heightened activity of plant phospholipases in certain plant tissues and cultured cells. This heightened activity results in the production of secondary messengers like InsP3 and diacylglycerol (DAG), facilitating the release of intracellular Ca2+. Additionally, this process activates signaling pathways associated with nitric oxide and the octadecanoid pathway.

Swift alterations in protein phosphorylation patterns have been detected following the application of elicitors to various cell cultures. Current research suggests that reversible phosphorylations are involved in the transmission of signals in plants during pathogen or stress defense mechanisms.

"G-protein activation which are also involved in the early responses to elicitors."

"Activation of NADPH oxidase responsible for AOS and cytosol acidification"

"Cytoskeleton reorganization."

"Generation of active oxygen species."

"Accumulation of pathogenesis-related proteins such as chitinases and glucanases, endopolygalacturonases, hydroxyproline-rich glycoproteins, protease inhibitors."

"Cell death at the infection site(hypersensitive response)."

"Structural changes in the cell wall (lignification of the cell wall, callus deposition)."

"Transcriptional activation of the corresponding defence response genes."

"Plant defence molecules such as tannins and phytoalexins are detected2-4 h after stimulation with the elicitor."

"Synthesis of jasmonic and salicylic acids as secondary messengers."

"Systemic acquired resistance."

"The investigation of the chronological order, interconnection, and orchestration of these events is complex and still under study. It's worth noting that not all elicitors necessarily follow the same sequence of events."

Main functions of elicitors

Metabolites in plants serve as protective agents against both biotic and abiotic stressors. Many of these metabolites contribute to the sensory and qualitative attributes of foods. Additionally, these compounds serve as valuable resources for industrial applications, including food additives and pharmaceuticals. They are often deemed beneficial for health, primarily due to their antioxidant properties, and play a role in enhancing disease resistance in plants.

Synthetic elicitors

"Synthetic elicitors, as defined by Yasemin Bektas and Thomas Eulgem (2014) [14], are small, drug-like molecules designed to stimulate plant defense responses independently of natural elicitors involved in plant immunity. Examples of synthetic elicitors include polyamines, salicylic acid, benzothiadiazole, jasmonic acid, chitosan, brassinosteroids, triacontanol, 2,6-dichloro-isonicotinic acid, probenazole, isotianil, sulfanilamide, clopamide, butamide, and various others."

(A) Polyamines

"Polyamines are organic polycations distinguished by variable hydrocarbon chains and containing two or more primary amino groups. These compounds are ubiquitous in living organisms, and they are particularly abundant in actively proliferating cells. The three primary forms of polyamines are putrescine, spermidine, spermine, and cadaverine"

Role of polyamines

In the realm of plants, polyamines play a pivotal role in various physiological functions, exerting influence on growth and development by modulating processes such as cell division and differentiation. These compounds also contribute to essential stages like flowering and fruit ripening. Polyamines exhibit a regulatory effect that enhances overall plant productivity. Notably, they function as anti-senescent agents, effectively postponing softening in fruits like mango and papaya by reinforcing the integrity of cell membranes. Furthermore, polyamines serve as crucial carbon and nitrogen reserves and operate as a distinct class of growth regulators, acting as secondary hormonal messengers. Their involvement extends to the management of both abiotic and biotic stress in plants, playing a crucial role in mitigating issues like chilling injury, heat and salinity stress, drought, and contributing to disease control. Significantly, polyamines have demonstrated their efficacy in alleviating the harmful impacts of ozone on plant health.

(B) Salicylic acid

"Salicylic acid (SA), a phenolic derivative, is found in a diverse range of plant species and is produced from the amino acid phenylalanine. Synthetic analogs of SA, including 2,6-dichloroisonicotinic acid and BTH (benzothiadiazole), have been synthesized. The biosynthesis of salicylic acid includes the decarboxylation of trans-cinnamic acid to benzoic acid, followed by subsequent 2-hydroxylation."

Role of salicylic acid

"Phenolic compounds, especially salicylic acid (SA), play crucial roles in various physiological and biochemical processes in plants, including photosynthesis, ion uptake, membrane permeability, enzyme activities, flowering, and overall growth and development. Plant health promoters like Bion or Actigard, containing the active ingredient Benzothiadiazole, induce the SA-dependent defense pathway. SA serves as an endogenous signaling molecule, contributing to plant defense mechanisms against pathogens. Its participation in resistance to pathogens is evident through the induction of pathogenesis-related proteins. Additionally, SA is involved in Systemic Acquired Resistance (SAR), where a pathogenic attack in one part of the plant induces resistance in other parts. The signal can also be transmitted to nearby plants through the conversion of salicylic acid to the volatile ester methyl salicylate. Foliar application of salicylic acid and potassium silicate, either alone or in combination at varying concentrations, significantly enhances quality parameters and extends the shelf life of mangoes (cv. Alphonso) compared to the control. Notably, treatment with salicylic acid at 200 ppm + potassium silicate at 0.2% (T8) demonstrates improvements in fruit quality attributes and prolonged shelf life during storage. (Gonchikari Lokesh *et al*. 2020) [5]"

"The study conducted by Kanwal *et al*. (2021) [4] showcased the effectiveness of pre-harvest spray with salicylic acid (SA) in mitigating pre-harvest fruit drop and improving the quality of ber fruit, specifically in the Gola cultivar. Among the different doses of SA applied, the treatment with 1mM SA stood out for its notable impact. This particular application led to a decrease in fruit drop, an elevation in yield, and an overall enhancement in the quality of ber fruit compared to other treatment regimens."

(C) Jasmonic acid

Jasmonic acid, a plant hormone, is derived from the fatty acid linolenic acid. Its volatile organic compound counterpart, methyl jasmonate, is derived directly from jasmonic acid. The term "jasmonates" encompasses both jasmonic acid and its various metabolic derivatives.

Role of jasmonic acid

Jasmonic acid plays a crucial role in regulating various plant growth and development processes, including growth inhibition, senescence, flower development, and leaf abscission. It is particularly responsible for tuber formation in potatoes, yams, and onions. Additionally, jasmonic acid is involved in the response to plant injuries and the systemic acquired resistance (SAR) mechanism. When plants are damaged, the levels of jasmonic acid increase.

Preharvest applications of methyl jasmonate (MeJA) have shown promising advantages in increasing pomegranate crop yield, improving fruit quality, and boosting the content of bioactive compounds both at harvest and during storage. The optimal effects were observed with MeJA treatment at a concentration of 5 mmol L-1, suggesting its suitability as a preferred treatment for practical applications (Garcia, Pastor, Maria E., *et al*. 2019) [3].

(D) Chitosan

"Chitosan, obtained from deacetylated chitin, is currently sourced from the outer shells of crustaceans like crabs, krills, and shrimps. Chitin and chitosan are polysaccharides, sharing chemical similarities with cellulose and differing primarily in the presence or absence of nitrogen."

Role of chitosan

"Chitosan, when employed as a carbon source, has the capability to stimulate the growth of beneficial soil microbes. This stimulation expedites the transformation of inorganic matter into organic matter, facilitating the absorption of nutrients by plant roots and ultimately fostering plant growth. In the realm of agriculture, chitosan serves a dual purpose as a seed treatment and biopesticide, aiding plants in resisting fungal infections."

"In summary, the concurrent application of tetramycin and chitosan demonstrated effectiveness in controlling leaf spot disease in kiwifruit. This joint treatment resulted in significant elevations in total phenolics, total flavonoids, soluble protein, catalase (CAT), peroxidase (POD), polyphenol oxidase (PPO), and superoxide dismutase (SOD) activities in kiwifruit leaves. Simultaneously, it led to a decrease in malondialdehyde (MDA) content and enhanced photosynthesis. Importantly, the co-application of tetramycin and chitosan exhibited superior efficacy compared to the individual application of tetramycin or chitosan, contributing to the improved growth, quality, and amino acid composition of kiwifruit. This study underscores the practical potential of using tetramycin and chitosan in tandem for the management of leaf spot disease in kiwifruit. (Zhang, Cheng, *et al*, 2022) [15]"

(E) Brassinosteroids

At the cellular level, brassinosteroids play a regulatory role in fundamental processes such as cell elongation, cell division, and cell differentiation. Their influence extends to the whole-plant level, where they contribute to the regulation of hypocotyl elongation, root and shoot development, leaf development, male fertility, senescence, and responses to both biotic and abiotic stresses. Moreover, the positive impact of brassinosteroids on yield is evident through their involvement in cell elongation and division, stimulation of photosynthetic activity, and enhancement of protein biosynthesis. These phytohormones are also known to promote nucleic acid levels, facilitate nitrogen fixation, increase soluble protein content, and elevate concentrations of DNA and RNA. In addition to these physiological responses, brassinosteroids exhibit growth-promoting effects akin to auxins and gibberellins, underscoring their promising potential for enhancing overall crop yield.

Role of brassinosteroids

"At the cellular level, brassinosteroids function as regulators in key processes such as cell elongation, cell division, and cell differentiation. Operating at a whole-plant level, they play a vital role in governing various aspects, including hypocotyl elongation, root and shoot development, leaf development, male fertility, senescence, and responses to both biotic and abiotic stresses. The contribution of brassinosteroids to enhanced yield is mediated through the regulation of cellular functions like cell elongation and division. They induce processes such as cell division, elongation, and differentiation, thereby stimulating photosynthetic activity by accelerating CO_2 fixation and increasing protein biosynthesis. Additionally, brassinosteroids are recognized for their ability to promote nucleic acid levels, nitrogen fixation, enhance soluble protein content, and elevate concentrations of DNA and RNA. Beyond these physiological responses, brassinosteroids demonstrate growth-promoting effects comparable to auxins and gibberellins, emphasizing their promising potential for overall yield improvement in crops (Rajni Rajan *et al*., 2017) [11]."

(F) Tricontinol

1-Triacontanol, with the chemical formula C30H62O, is a fatty alcohol also known as melissyl alcohol or myricyl alcohol. It is naturally present in plant cuticle waxes and is a component of beeswax.

Role of Tricontinol

"Triacontanol (TRIA) has shown positive effects on various aspects of plant physiology and performance. Its beneficial impacts include improvements in growth, yield, photosynthesis, protein synthesis, water and nutrient uptake, nitrogen fixation, enzyme activities, as well as the contents of free amino acids, reducing sugars, soluble protein, and active constituents of essential oil in different crops. Triacontanol acts as a growth stimulant for many plants, and in specific instances, such as in roses, Tricontinol has been observed to increase the number of basal breaks."

"According to the findings of S. K. Momin *et al*. (2016) [7], the application of Triacontanol at a concentration of 750 mg/litre yielded positive results in terms of fruit setting at the pea and marble-sized stage of fruits per panicle. Additionally, there was an increase in fruit retention per panicle at the harvesting stage, with minimal fruit drop observed during harvesting. The application of Triacontanol also resulted in a higher number of fruits on trees. These beneficial effects are attributed to the application of Triacontanol, which is thought to enhance the efficient utilization of nutrients for reproductive growth, flowering, and fruit setting. Other contributing factors include improved photosynthetic efficiency, an enhanced source-to-sink relationship in plants, increased nutrient and water uptake, reduced transpiration and respiration, as well as enhanced translocation and accumulation of sugars and other metabolites."

Conclusion

Defense modulators, such as elicitors, have the potential to enhance plant productivity and quality by influencing various metabolic processes. One of the advantages of using elicitors is the reduced environmental hazards compared to traditional pesticides, as they directly affect crop plants with lower acute toxicity to other organisms. Elicitors play a significant role in plant defense mechanisms, and their application can lead to improvements in fruit quality, color, appearance, shelf life, firmness, and resistance to spots. This can be particularly beneficial for farmers. Despite the promising potential, the use of elicitors in crop protection and pest management is still in its early stages, and there is a need for large-scale agricultural and horticultural experiments to validate their role in plant protection.

Future thrust

Utilization of next-generation elicitors and bio-molecules for improving fruit quality and disease management in fruit crops. The use of elicitors in crop protection and pest management is still in the very early stages of use as a new control method and hence, there is a need to conduct large scale agricultural/horticultural experiments to validate their role in plant protection. Improved quality fruit with high phenolic and anti-oxidant is a basic need to fight against several disease, COVID-19 is one of them.

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