



A Review : Role of Elicitors in Plant Defense Mechanism

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Abstract

Disease means disturb the normal physiological processes in plants caused by microorganisms and an abiotic factor. Pathogen means an agent that causes infection. The attribution of the hosts that reduce the chances of infection and the further development of the pathogen are considered to be defense mechanism. Defense mechanism in plants can be done by Elicitors. Elicitors are the chemicals and bio factors from various sources that can trigger physiological and morphological responses and also phytoalexin accumulation in the target living organisms. These elicitors may be present in plants and also produced synthetically. Elicitors are compounds, which activate chemical defense in plants. Various biosynthetic pathways are activated in treated plants depending on the compound used. Commonly tested chemical elicitors are salicylic acid, methyl salicylate, benzothiadiazole, benzoic acid, chitosan and so forth which affect production of phenolic compounds and activation of various defense-related enzymes in plants. Elicitors are used in agricultural practice that could minimize the scope of chemical control, thus contributing to the development of sustainable agriculture.

Keywords: Disease, pathogen, elicitors, synthetic elicitors

Introduction

Disease means disfunction of normal physiological processes in plants caused by microorganisms or an abiotic factor. Pathogen means an agent that causes infection or disease, especially microorganism, such as a bacterium; protozoan and virus. A pathogen that causes diseases is termed virulent. A pathogen that does not cause diseases is termed avirulent. Plant pathogens are three types, they are:-

Necrotrophic pathogen	Bio trophic pathogen	Hemibiotrophic pathogen
Necrotrophic fungal pathogens infect and kill host tissue and extract nutrients from the dead host cells	Bio trophic fungal pathogens colonize living plant tissue and obtain nutrients from living host cells	Hemibiotrophic fungi are successful groups of plant pathogens that require living plant tissue to survive and complete their life cycle
Ex: Grey mould fungus -- <i>Botrytis cinerea</i> Bacterial pathogen – <i>Erwinia caratovora</i> (Soft rot of potato)	Ex: <i>Erysiphe cruciferarum</i> (Powdery mildew on cabbage)	Ex: <i>Phytophthora infestans</i> (Late blight of potato) <i>Pseudomonas syringae</i> (Bacterial speck disease in tomato)

Defense mechanism in plants

The attribution of the hosts that reduce the infection and the further development of the pathogen are opined to be defense mechanism. The defense may be used against pathogen at any stage of infection during pre-penetration, penetration and post penetration. Two types of defense are :-

Constitutive defense	Induced defense
The defense is always present in the plant	The defenses that are produced when a plant is injured and detects foreign pathogen
Ex: 1. Cell wall - prevents the pathogen from entering the cell 2. Waxy epidermal cuticle - prevents water loss and micro bacterial enter into epidermal cuticle 3. Bark -- Keeps pathogens and insects from going to the living cells underneath the bark 4. Thorns – Reduce feeding by large herbivores by limiting the feeding rate	Ex: Toxic chemicals Pathogen degrading enzymes Deliberate cell suicide Systemic resistance

Disease control is largely based on the use of fungicides, bactericides and insecticides. These chemical compounds are toxic to plant invaders, causative agents and vectors of plant diseases. However, the hazardous effect of these chemicals and their degradation products on the environment and human health strongly necessitates the search for new, harmless means of disease control. There must be some natural phenomenon of induced resistance to protect plants from disease.

Elicitors

Elicitors are the chemicals and bio-factors from various sources that can trigger physiological and morphological responses and also phytoalexin accumulation in the target living organisms. Originally the term elicitor was used for molecules capable of inducing the production of phytoalexins, but it is now commonly used for compounds stimulating any type of plant defense (Thakur and Sohal, 2013). Elicitor molecules attach to the special receptor proteins located on plant cell membranes. Receptor is a protein molecule that receives chemical signals from outside a cell. A molecule that binds to a receptor is called a Ligand. The receptors are able to recognize the molecular pattern of elicitors, which trigger the intracellular defense signaling and results in the increased synthesis of secondary metabolites, phytoalexins which reduce the damage and increase the resistance to pests, diseases and environmental stresses. This is known as Pattern Triggered Immunity (PTI). Receptors are two types, they are

Transmembrane receptors	Intracellular receptors
They include ion channel linked (ionotropic), G – proteins, enzyme linked hormone receptors	They include cytoplasmic receptors, nuclear receptors

Use of elicitors in defense mechanism: Minimizing these losses is a major area of concern for all nations to cope with increasing food requirements. In India, losses due to diseases is 20% and insect pests is 25%, and abiotic stresses reduce an average yield of crops by upto 50% (Verma and Deepthi, 2016). There are various options available for the farmers to protect their crop. Some options include development of resistant cultivars, biological control, crop rotation, tillage and agro-chemicals. Nearly all chemical pesticides and fungicides have a direct antibiotic principle. But their use at commercial level is uneconomical, application is cumbersome, and some are proved to be carcinogenic.

Classification of elicitors

Endogenous elicitors:- Elicitors are produced within plant cells

Exogenous elicitors :- Elicitors are produced by microorganisms

Depending upon their origin, they are classified as **biotic** and **abiotic** elicitors. Biotic elicitors are either pathogen (or) host origin that can stimulate defense responses (such as phytoalexin accumulation) in plant tissues. Abiotic elicitors are of non-biological origin. (Baenas *et al.*, 2014).

Biotic Elicitors	
Polysaccharides	Pectin and Cellulose (Cell wall); Chitosan, Chitin and Glucans, alginate
Oligosaccharides	Galacturonides, guluronate, mannan, mannuronate
Proteins	Cellulase, Cryptogein, Glycoproteins, Oligandrin, Pectolyase, Lactoferrin
Pathogen toxin	Coronatine

Abiotic Elicitors	
Chemical	Physical
Acetic acid	Altered gas composition
Benzothiadiazole (BTH)	Chilling
Silicon	CO ₂
Prohexadione	Drought
Ethanol, Ethene	Extreme temperature shock
Inorganic salts: HgCl ₂ , CuSO ₄ , CaCl ₂ , VSO ₄	High pressure
Metal ions: Co ²⁺ , Fe ²⁺ , Al ³⁺ , Ag ²⁺ , Mn ²⁺ , Zn ²⁺ , Cu ²⁺ , Pb ²⁺ and Cd ²⁺	High (or) low osmolarity
	UV irradiation
	Saline stress
	Wounding

Plant Hormones
Jasmonic acid, Methyl jasmonate, Salicylic acid, Methyl salicylate, Ethylene, ABA, Polyamines, Brassinosteroids, Tricontinol

Elicitor classification based on their interaction with the host plant:-

General Elicitors	Specific Elicitors
They are able to trigger defense both in host and non-host plants	They induce defense responses leading to disease resistance only in specific host cultivars
Ex: Carbohydrates, Cell wall proteins, Oligosaccharides	Ex: (<i>avr</i> gene products) from fungal, bacterial, viral (or) plant origin

General elicitors: They do not significantly differ in their effect on different cultivars within a plant species and are involved in primary innate immunity. They include chemicals, Microbes – Associated Molecular Patterns (MAMPs) from non- pathogenic microorganisms, Damage Associated Molecular Patterns (DAMPs) from pathogenic microorganisms, (PAMPs---Pathogen Associated Molecular Patterns). Even if perception of elicitors is often described as being receptor mediated, only few binding sites have been characterized to date.

Specific elicitors (Effectors): They are formed by specialized pathogens and function only in plant cultivars carrying the corresponding disease resistance gene. Effectors typically lead to the secondary innate immunity after an intracellular receptor mediated perception

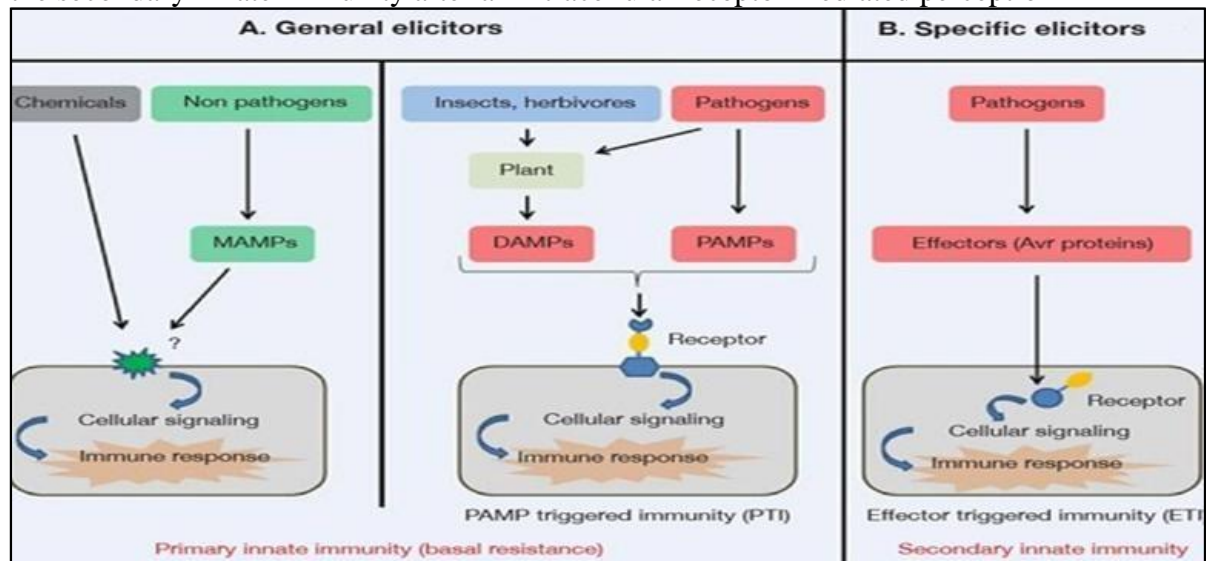


Fig 1: Primary innate and Secondary innate immunity

Elicitor- induced effects in plant cells: Different types of effects are induced in plant cells by the use of elicitors, which is reflected by the influenced cell metabolism. Increase in production of secondary metabolites, Phytoalexin biosynthesis, Increase in the activity of defense enzymes, and Induces SAR and LAR by accumulation of pathogenesis-related (PR) proteins, differential gene expression, consequently forming enzymes concerned in the synthesis of polysaccharides as callose, Hydroxyproline Rich Glyco Proteins (HRGP) in cell walls *via* induction of proline hydroxylase, lignin and polyphenolics (deposited in cell walls). When elicitors bind to the receptor on the cell wall signal transduction pathways gets activated which results in the production of defense genes. These defense genes releases several gene products *viz.*, secondary metabolites (abiotic stress), pathogenesis related (PR) proteins, Phytoalexins and chitinases (biotic stress) which help in primary immune response of plants (Fig.2). Elicitors like salicylic acid (SA) and Jasmonic acid (JA) activates their respective dependent pathways *i.e.*, SA-dependent pathway and JA-dependent pathway (Fig. 3). SA results in systemic acquired resistance (SAR) *i.e.*, when there is a pathogen attack in one part of the plant then the remaining parts of the plant become resistant to further pathogen attack. SA activates NPR1 (Non-expressor of pathogenesis related genes) which releases PR-1, 2, 5 (pathogenesis related proteins) that causes SAR. JA results in localized acquired resistance (LAR) *i.e.*, when there is a pathogen attack JA helps in the death of the affected tissue thus preventing further attack of the pathogen. JA activates MYC2 (mycelocytomatosis-2) a transcription factor which releases PR-3, 4 and PDF 1.2 (Protodermal factor 2) that causes LAR (Fig. 2). (Enoki and Suzuki, 2016).

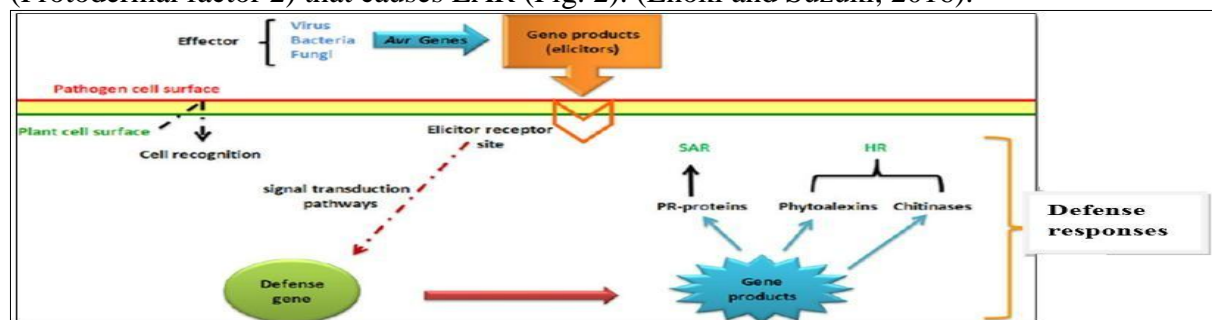
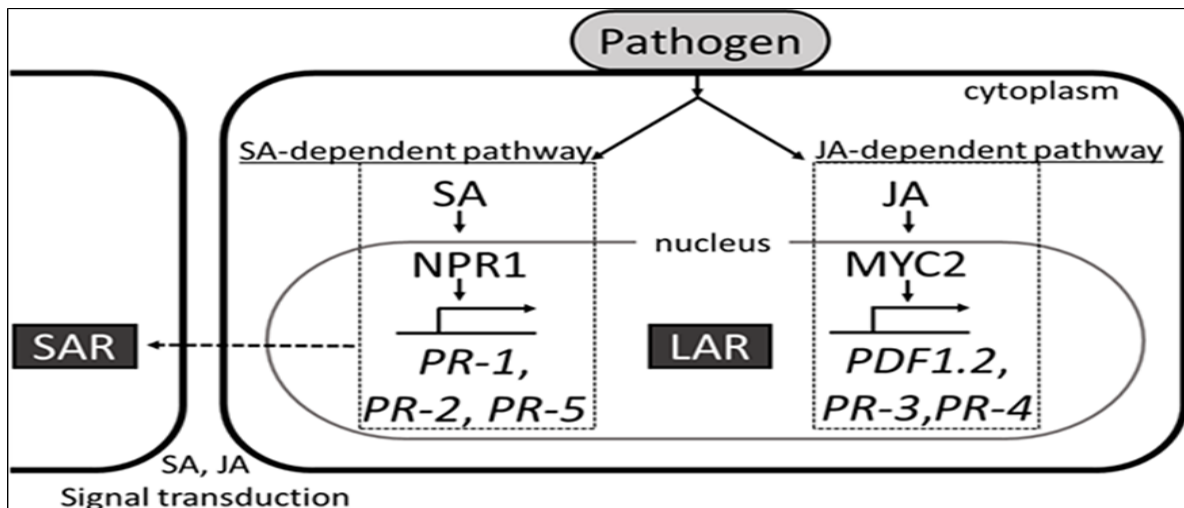


Fig 2: Elicitor induced defense responses in plant



NPR1 - Non-expressor of pathogenesis-related genes

MYC2 - Mycelocytomatosis-2

PDF 1.2 - Plant defensin 1.2 PR- 2, 3, 4, 5- Pathogenesis related proteins

Fig 3: SA and JA dependent pathway leading to SAR and LAR upon pathogen attack

Systemic acquired resistance (SAR)

Induced resistance in plants is generally systemic. Thus, defence responses to pathogenic infection occur not only at the initial infection site, but also in spatially separated tissues. SAR is a mechanism of induced defense that confers potentially long-lasting protection against a broad spectrum of microorganisms and found across a wide range of plant species. Salicylic acid is a key signal compound in the SAR response to pathogen infection. After pathogen recognition by the host, a cascade of early responses is induced including ion fluxes, phosphorylation, which precedes the accumulation of ROS, nitric oxide, SA and the transcriptional activation of defense-related genes.

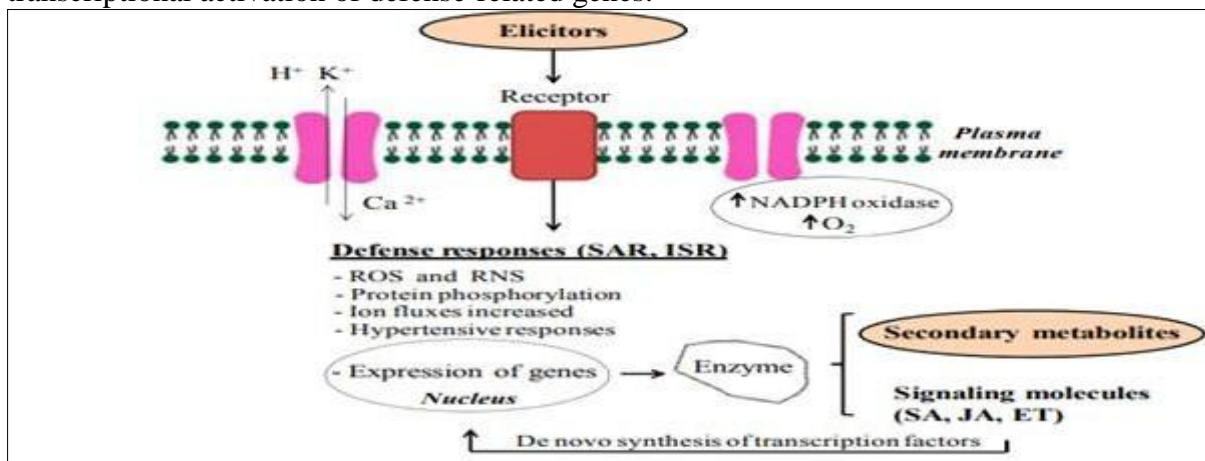


Fig 4: General mechanism after elicitor perception

Pr-proteins

Pathogenesis related proteins are called PR proteins - A group of plant coded proteins. They have low molecular weights and are extractable at low pH. Most of them are located in the apoplast. They produced under stress. They are widely distributed in plants in trace amounts but are produced in high concentration following pathogen attack.

Significance of PR-proteins: They show strong antifungal and other antimicrobial activity. These are associated with strengthening of the host cell wall, it's out growths and papillae. Some of the PR-proteins, for Ex: β -1,3- glucanase and chitinase have the ability to hydrolyze

fungal cell wall components and also release elicitor of oligosaccharides from the cell walls of pathogens.

Classification of PR-proteins in grape

Family	Property	Function/Target site
PR -1	Antifungal	Unknown
PR -2	β - 1,3 -Glucanase	Cell wall (β - 1,3 glucan)
PR -3	Chitinase (types I, II, IV, V, VI, and VII)	Cell wall (chitin)
PR -4	Chitinase (types I and II)	Cell wall (chitin)
PR -5	Thaumatococin-like	Plasma membrane
PR -1 0	Ribonuclease (like)	RNA
PR -1 4	Lipid-transfer protein	Involvement in defense signaling pathway
PR -1 5	Oxalate oxidase	Production of H ₂ O ₂ with
PR -1 6	Oxalate oxidase-like protein	Antimicrobial activity

Local Acquired Resistance (LAR)

Induced resistance is at first localized around the point of plant necrosis caused by infection by the pathogen (or) by the chemical and then it is called Local acquired resistance. LAR is characterised by elevated levels of antifungal compounds and defence enzymes. LAR is effective against a broad range of pathogens.

● Synthetic elicitors

Synthetic elicitors are small drug-like molecules that induce plant defense responses, but are distinct from known natural elicitors of plant immunity. Some synthetic elicitors are Polyamines, Salicylic acid, Benzothiadiazole, Jasmonic acid, Chitosan, Brassinosteroids, Tricentrol, 2,6-dichloro- isonicotinic acid, Probenazole, Isotianil, Sulfanilamide, Clopamide, Butamide (Bektas and Euglem, 2015).

A) **Polyamines:** Polyamines are organic polycations having variable hydrocarbon chains and two or more primary amino groups. These are widespread in living organisms, found in high concentration in actively proliferating cells. Ex. Putrescine, Spermidine, Spermine and Cadaverine

Role of polyamines

In plants polyamines are responsible for the performance of wide range of functions like growth and development because of its effect on cell division, flowering and fruit ripening. These polyamines are reported to have the regulatory effect on promoting the productivity of the plants. They are found to be anti-senescent agents and effective for delaying softening in several fruits (mango, papaya), as they help in the integrity of cell membrane. Polyamines play important role in abiotic and biotic stress management. Polyamines helps in eliminating the chilling injury, heat & salinity stress, drought and disease control.

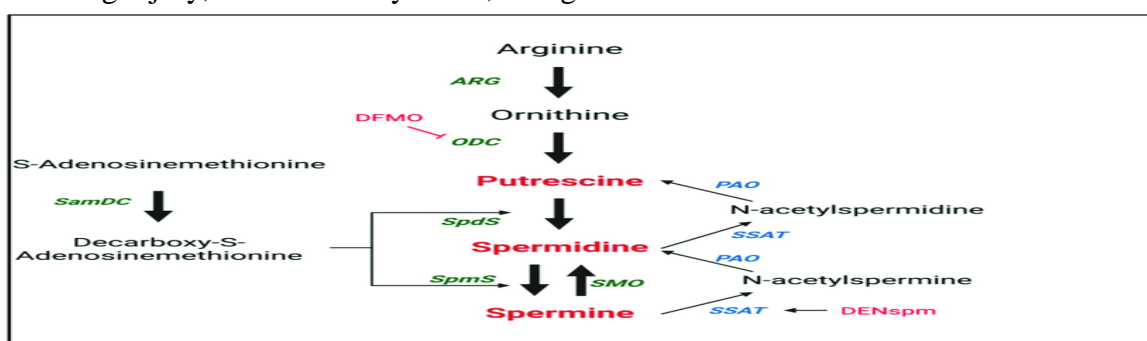


Fig 5: Polyamines biosynthesis

B) Salicylic acid: Salicylic acid is a phenolic derivative, distributed in a wide range of plant species. It is biosynthesized from the amino acid Phenylalanine. Synthetic SA analogs are 2,6 dichloro iso nicotinic acid and BTH. Decarboxylation of transcinnamic acid to benzoic acid and its subsequent 2- hydroxylation results to salicylic acid.

Role of salicylic acid

Phenolic compounds exert their influence on physiological and biochemical processes including, photosynthesis, ion uptake, membrane permeability, enzyme activities, flowering, growth and development of plants. Bion and active ingredient is Benzothiadiazole (Actigard) is a commercially released plant health promoter which induces SA- dependent defense pathway. SA is involved in endogenous signaling, mediating in plant defense against pathogens. It plays a role in the resistance to pathogens by inducing the production of pathogenesis-related proteins. It is involved in the SAR in which a pathogenic attack on one part of the plant induces resistance in other parts. The signal can also move to nearby plants by salicylic acid being converted to the volatile ester, methyl salicylate

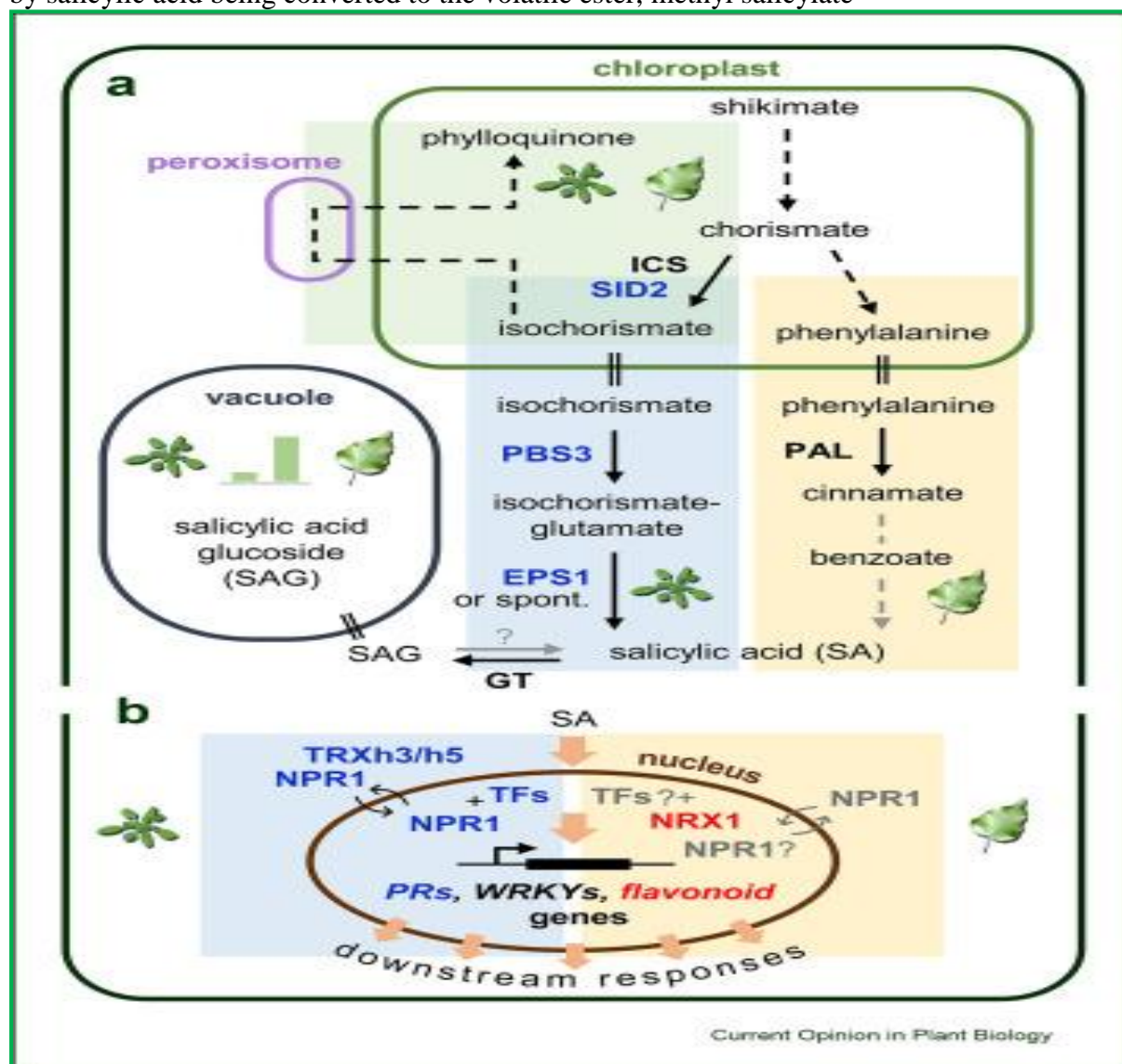


Fig 6: Salicylic acid biosynthesis

C) Jasmonic acid: Jasmonic acid is derived from the fatty acid linolenic acid. Methyl jasmonate is a volatile organic compound, derived from jasmonic acid.

Role of jasmonic acid: Regulated plant growth and development processes include growth inhibition, senescence, flower development and leaf abscission. Jasmonic acid is responsible

for tuber formation in potatoes, yams and onions. It has an important role in response to wounding of plants and SAR. Levels of jasmonic acid rise in response to damage. The action of jasmonic acid induces the transcription of many genes involved in plant defense.

D) **Chitosan:** Chitosan, deacetylated chitin, is currently obtained from the outer shell of crustaceans such as crabs, krills and shrimps. Chitin and chitosan are polysaccharides, chemically similar to cellulose differing only by the presence (or) absence of nitrogen

Role of chitosan: Chitosan as a carbon sources may stimulate the growth of beneficial microbes in the soil. Accelerating the transferring processes of inorganic matter into organic matter. Facilitating the root system of plants to absorb more nutrients from soil hence stimulating plant growth. Chitosan is used in agriculture as a seed treatment and biopesticide, helping plants to fight off fungal infections.

E) **Brassinosteroids:** Brassinosteroids are class of plant polyhydroxysteroids that recognized as new kind of phytohormones. The occurrence of brassinosteroids has been demonstrated in almost every part of plants. Pollen and immature seeds are the richest sources of BR with a range of 1–100 µg/kg (fresh tissue), whereas shoots and leaves usually possess lower amounts of 0.01–0.1 µg/kg (fresh tissue). About 70 brassinosteroids have been isolated from plants.

Role of brassinosteroids: At cellular levels, brassinosteroids can regulate cell elongation, cell division, cell differentiation. At whole-plant levels, brassinosteroids can regulate hypocotyl elongation, root and shoot development, leaf development, male fertility, senescence and responses to biotic and abiotic stresses

F) **Tricontinol:** 1-Triacontanol is a fatty alcohol of the general formula C₃₀H₆₂O, also known as melissyl alcohol (or) myricyl alcohol. It is found in plant cuticle waxes and in beeswax.

Role of Tricontinol: TRIA shows improvement in growth, yield, photosynthesis, protein synthesis, uptake of water and nutrients, nitrogen- fixation, enzymes activities (malate dehydrogenase – respiratory enzyme) and contents of free amino acids, reducing sugars, soluble protein and active constituents of essential oil in various crops. Triacontanol is a growth stimulant for many plants. Ex: In Roses, Tricontinol increased the number of basal breaks

Types of elicitors used in different horticulture crops

S. No.	Plant	Type of Elicitor used	Effects	Reference
1	Gladiolus	Salicylic acid	Delays petal senescence and extended quality of cut spikes	Hatamzadeh <i>et al.</i> , 2012
2	Lilium	Benzoic acid	Modified the growth, and induced stress tolerance, anatomy and morphology of eatable and ornamental species	Ding <i>et al.</i> , 2002
3	<i>Citrus sinensis</i>	B-amino butyric acid	Involved induced resistance against <i>Penicillium italicum</i> by inhibiting spore germination <i>in vitro</i>	Tavallai <i>et al.</i> 2008
4	Tomato	Avr gene products, Ex: AVR 4 and AVR 9 from <i>Cladosporium fulvum</i>	Activation of hypersensitive response	Leach and White, 1996

5	Bhendi	Salicylic acid	Accumulation of phenolics and increased activity of enzyme PAL leading to resistance against <i>Erysiphe cichoracearum</i>	Vimala and Suriachandrasel van, 2009
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Conclusion

Extensive use of agrochemicals leads to the problems of pest resistance, resurgence, residues, destruction of beneficial fauna and environmental pollution. Pesticides and fungicides possess inherent toxicities that endanger the health of the farmers, consumers and the environment. Elicitors cause no adverse effects on human health and leave no residues. Hence, use of elicitors in disease management is an environmental friendly strategy and reduces environmental and health hazards.

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