



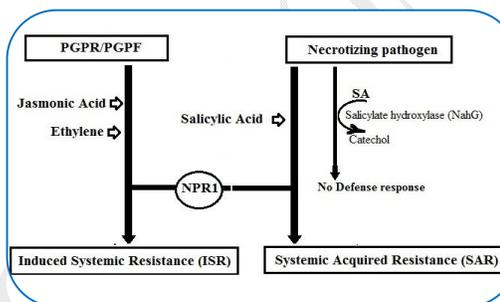
## AN OVERVIEW OF PGPR/PGPF MEDIATED INDUCED SYSTEMIC RESISTANCE (ISR) IN PLANT DEFENSE

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### ABSTRACT

Plants are exposed to various pathogens such as fungi, bacteria, virus, insect, etc which may cause some short of physiological and morphological disorder in plant, so called diseases. To overcome this disease incidence, plants have some defense mechanisms such as induced systemic resistance (ISR) and systemic acquired resistance (SAR). ISR is mediated by root colonizing plant growth promoting rhizobacteria (PGPR) or plant growth promoting fungi (PGPF) in response of Jasmonic acid (JA) and Ethylene (ET) which is subjected to the expression of defense related enzymes and defense chemicals in order to structural and chemical barrier against pathogen, while SAR is responsible for expression of pathogenesis-related proteins (PR-proteins) in response to a different endogenous signaling molecule, salicylic acid (SA) against necrotizing pathogen. PGPR and PGPF play dual roles in plant growth such as induction of resistance and promoting of growth. In this present work JA- ET dependent ISR is focused for protection of crops against pathogenic agents.



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**KEYWORDS:** Induced Systemic Resistance (ISR), Jasmonic acid (JA) and Ethylene (ET), Defense mechanism

### INTRODUCTION

All higher plants have the ability to express various defense mechanisms when they are exposed to various pathogens such as fungi, bacteria, virus, insect, etc. These mechanisms do not allow pathogen to cause disease in plant, if the reaction occurs in a timely manner. However, if the defense reactions start at too late or suppressed, infection of pathogen occurs successfully which may causes disease in plant (Somssich and Hahlbrock, 1998). There are two different signal transduction pathways in plant such as systemic acquired resistance (SAR) and induced systemic resistance (ISR). ISR is called as Jasmonic acid-Ethylene (JA-ET) dependant- plant growth promoting rhizobacteria (PGPR) or plant growth promoting fungi (PGPF) mediated induced systemic resistance (ISR) which is subjected to the expression of defense related enzymes and defense chemicals for structural and chemical barrier in host plant against pathogen. While, SAR is called as salicylic acid (SA) dependant- necrotizing pathogen mediated systemic acquired resistance which is responsible for expression of pathogenesis related proteins (PR-proteins) playing direct defensive role against pathogen attract.

A variety of biotic and abiotic inducers/elicitors are reported by several workers for induction of induced systemic resistance (ISR) and systemic acquired resistance (SAR) in different crops. Among the abiotic inducers, Meena *et al.* (2001) used salicylic acid in groundnut, Higa *et al.* (2001) used active oxygen radicals in rice, O'Donnell *et al.* (1996) used ethylene in tomato, Smith-Beaker *et al.* (1998) used SA and 4-

hydroxybenzoic acid in cucumber, Cohen *et al.* (1993) used jasmonic acid and methyl jasmonate in potato and tomato, Siegrist *et al.* (2000) used  $\beta$  - aminobutyric acid in tobacco, Kaur and Kolte (2001) used benzothiadiazole in mustard and wheat plant, Brederode *et al.* (1991) used UV-light in tobacco, Ernst *et al.* (1992) used ozone in tobacco, Klessig *et al.* (2000) used nitric oxide and Kaku *et al.* (1997) used N-acetylchitooligosaccharide in barley. Similarly, some biotic inducers have also been used to enhance in plant defense reactions such as leaf extract of *Azadirachta indica* in barley (Paul and Sharma, 2002), *Acalypha indica* in ginger (Ghosh and Purkayastha, 2003), *Reynoutria sachaliensis* in cucumber (Daayf *et al.*, 1995). Thus it is important for plants for early detection of pathogen and early delivering signal information for ISR (intracellularly/ intercellularly) to plant for activation of defense machinery such as phytoalexins, antimicrobial proteins, defense enzymes, reactive oxygen species etc. against pathogen (Shibuya and Minami, 2001) for management of diseases.

### INDUCED SYSTEMIC RESISTANCE (ISR)

The plant root colonizing microorganisms such as plant growth promoting fungi (PGPF) or plant growth-promoting rhizobacteria (PGPR) may suppress plant disease incidence by triggering defense mechanism in plant has been reported by several workers (Meera *et al.*, 1995; Munoz *et al.*, 2008). PGPF-mediated induced systemic resistance (ISR) been studied in great detail on downy mildew disease incidence in pearl melle using root colonizing fungi, *Penicillium sp.*, *Trichoderma sp.*, *Rhizoctonia sp.*, and *Pythium sp.* (Murali *et al.*, 2012). Plant growth promoting fungi (PGPF) or Plant growth promoting rhizobacteria (PGPR) can suppress the disease in plant by triggering induced systemic resistance (ISR). Induction of ISR by PGPR or non-pathogenic fungi PGPF differ from SAR for their signal transduction pathways, it is designated by a separate term ISR proposed by Kloepper *et al.* (1992) and latter supported by Pieterse *et al.* (1996). ISR requires essential endogenous signal molecules, Jasmonic acid (JA) and Ethylene (ET) for its expression in order to accumulation of defense related enzymes and defense related substances for structural and chemical barrier against pathogen (such as Peroxidase, Polyphenol oxidase, Chalcone synthase, Phytoalexin, Phenolic compounds, etc), rather than PR-proteins (Van Loon, 1999). Interestingly, simultaneous activation of both the JA/ethylene-dependent ISR pathway and the SA-dependent SAR pathway results in an enhanced level of disease protection. Thus combining both types of induced resistance provides an attractive tool for the improvement of disease management.

### PLANT GROWTH PROMOTING RHIZOBACTERIA (PGPR)

Root colonizing non pathogenic bacteria are generally called as plant growth promoting rhizobacteria (PGPR) which can be classified into extracellular plant growth promoting rhizobacteria (ePGPR) and intracellular plant growth promoting rhizobacteria (iPGPR) [Viveros *et al.*, 2010]. The ePGPRs may exist in the rhizosphere, on the rhizoplane and in the spaces between the cells of root cortex, while, (iPGPRs) are living within a specialized nodular structure in plant. The bacterial genera (such as *Agrobacterium*, *Arthrobacter*, *Azotobacter*, *Azospirillum*, *Bacillus*, *Burkholderia*, *Caulobacter*, *Chromobacterium*, *Erwinia*, *Flavobacterium*, *Micrococcous*, *Pseudomonas* and *Serratia*) are belongs to ePGPR (Ahemad and Kibret, 2014). The bacterial species such as *Allorhizobium*, *Bradyrhizobium*, *Mesorhizobium* and *Rhizobium*, and *Frankia* are belongs to iPGPR, which can symbiotically fix atmospheric nitrogen with the higher plants (Bhattacharyya and Jha, 2012). Some common reported PGPR genera exhibit plant growth promoting activities are *Pseudomonas*, *Azospirillum*, *Azotobacter*, *Bacillus*, *Burkholdaria*, *Enterobacter*, *Rhizobium*, *Erwinia*, *Mycobacterium*, *Mesorhizobium*, *Flavobacterium*, *Pseudomonas fluorescens*, *Pseudomonas syringae* (Maurhofer *et al.*, 1994, Chen *et al.*, 2000; Liu *et al.*, 1995; Wei *et al.*, 1991, Rasmussen *et al.*, 1991, Singh, 2013).

### PLANT GROWTH PROMOTING FUNGI (PGPF)

Root colonizing non pathogenic fungi are generally called as plant growth promoting fungi (PGPF) which are reported to be suppressed disease incidence by triggering induced systemic resistance in

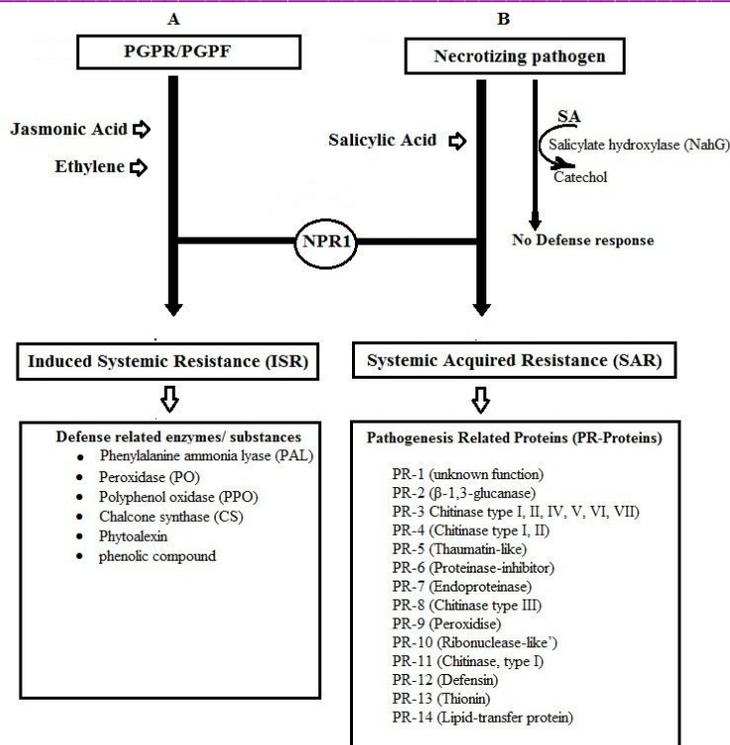
cucumber and effectively control soil-borne diseases like damping-off caused by *Fusarium*, *Rhizoctonia* and *Sclerotium* and take-all caused by *Gaeumannomyces graminis* of a number of crop plants (Narita and Suzuki, 1991; Meera *et al.*, 1994; Hyakumachi *et al.*, 1993). Hence PGPF has dual roles in plant protection which may trigger induced systemic resistance (ISR) as well as promoting growth of plants (Murali *et al.*, 2012). Some common plant growth-promoting fungi (PGPF) are reported as *Fusarium*, *Penicillium*, *Phoma*, *Trichoderma* (Meera *et al.*, 1994).

### SIGNAL TRANSDUCTION PATHWAY OF ISR

PGPR-mediated defense resistance mechanism, so called induced systemic resistance (ISR) is associated with signal molecules, Jasmonic acid (JA) and Ethylene (ET) response. ISR is responsible for induction of defense related enzymes and defense related substances rather than PR-Proteins (**Figure-1**). Jasmonic acid (JA) plays an important role in plant defense response (Creelman *et al.*, 1992) and its level is increased under wounding and treatment with pathogen-elicitors that induce genes encoding enzyme for structural and chemical barrier in plant cell against pathogenic agent. A mutant, *mpk4* shows elevated accumulation of salicylic acid (SA) in the absence of spontaneous necrotizing lesions in *Arabidopsis* (Petersen *et al.*, 2000). This mutation in the Mitogen Activated Protein Kinase 4 (MAPK4) gene results in the constitutive expression of SAR and PR-proteins. While, the wild-type MAPK4 is characteristic to be a negative regulator of SAR gene expression and a positive regulator of ISR. Constitutive expression of active MAPK kinase (NtMEK2) in tobacco plant results in the activation of two MAPKs: salicylic-acid-induced protein kinase (SIPK) and wound-induced protein kinase (WIPK) that lead to the expression of phenylalanine ammonia lyase (PAL), the first enzyme in the phenylpropanoid pathway that ultimately cause cell death (Bent *et al.*, 2001).

### DEFENSE ENZYMES AND COMPOUNDS

Many defense-related enzymes are involved in ISR gene expression. These includes oxidative enzymes such as peroxidase (PO) and polyphenol oxidase (PPO) which catalyse the formation of lignin and other oxidative phenols that contribute to the formation of defense barriers in plant cell structure against pathogen (Avdiushko *et al.*, 1993). Other enzyme such as tyrosine ammonia lyase (TAL) and phenylalanine ammonia lyase (Gundlach *et al.*, 1992), chalcone synthase (Creelman *et al.*, 1992) are involved in phytoalexin or phenolic compound biosynthesis. It is reported that Phenylalanine ammonia lyase (PAL) is a key enzyme of phenylpropanoid pathway which leads to the deposition of lignin, phytoalexins and phenolic compounds and form structural and chemical barriers of the plants to the pathogens (Ramamoorthy *et al.*, 2002). The defense related enzyme, Phenylalanine ammonia lyase (PAL) frequently increases in plants in respond to pathogen invasion. Maher *et al.* (1994) reported the increased disease susceptibility of tobacco plants to *Cercospora nicotiana* in which PAL activity was suppressed, but over expressed PAL, exhibited reduction of lesion areas caused by two compatible, necrotrophic pathogens in transgenic tobacco plants. Elicitor treatment and wounding in parsley and sweet potato increased the activity of PAL. About a 3-fold increase in phenolic content was observed 4 days after challenge inoculation with *C. personatum* following pretreatment with SA in groundnut (Meena *et al.*, 2001). Polyphenol oxidase (PPO) catalyses the synthesis of defense substances like tannin which is toxic to pathogenic microorganisms (Mahadevan and Sridhar, 1996; Chen *et al.*, 2000) and formation of oxidative phenols that contribute to the inhibition of pathogen to the plant cell (Avdiushko *et al.*, 1993). Peroxidase activity is changed under various environmental stresses such as heavy metals, salts, temperature (Kiwani and Lee, 2003), air pollution (Lee *et al.*, 2000). It is related with the defense reaction in plants that lead to the detoxification of the reactive oxygen species (Higa *et al.*, 2001).



**Figure-1: Molecular level plant defense related signal transduction pathway:** A- Plant Growth promoting rhizobacteria (PGPR)/ Plant Growth promoting fungi (PGPF) mediated signalling pathway, Induced Systemic Resistance (ISR) which require Jasmonic acid (JA) and Ethylene as elicitor signalling molecules and produce a diverse array of defense related enzymes (such as peroxidase, polyphenol oxidase, chalcone synthase etc.) and defense related substances (such as phytoalexin, anti-microbial phenolic compounds, etc); B- Necrotizing pathogen mediated signalling pathway, Systemic Acquired Resistance (SAR) which require endogenous salicylic acid (SA) signalling and produce Pathogenesis-related proteins (PR-proteins).

**Table 2. Differentiation between the mechanisms of SAR and ISR**

	SAR	ISR
<b>Differences</b>	<ol style="list-style-type: none"> <li>SAR is mediated by necrotizing pathogen</li> <li>It is switch on in response of an endogenous signalling molecule- Salicylic acid (SA)</li> <li>It is subjected to the expression of Pathogenesis related proteins (PR-proteins)</li> <li>It has direct inhibitory role against pathogenic agents.</li> </ol>	<ol style="list-style-type: none"> <li>ISR is mediated by non pathogenic microorganism such as Plant growth promoting rhizobacteria (PGPR) or Plant growth promoting fungi (PGPF)</li> <li>It is switch on in response of two different signalling molecules Jasmonic acid (JA) and Ethylene (ET).</li> <li>It is subjected to the expression of diverse range of defense enzymes and defense chemicals such as Phenylalanine ammonia lyase (PAL), Peroxidase (PO), Polyphenol oxidase (PPO), Chalcone synthase phytoalexin, phenolic compound etc.)</li> <li>It has indirect inhibitory activity such as involve in structural and chemical barrier</li> </ol>

	<p>5. SAR is regulated by NahG and NPR1</p> <p>6. It is used the term Systemic acquired resistance (SAR)</p> <p>7. MAPK4 is characteristic to be a negative regulator of SAR gene expression</p>	<p>against pathogenic agents</p> <p>5. SAR is not regulated by NahG and NPR1</p> <p>6. It is used the term Induced Systemic resistance (ISR)</p> <p>7. MAPK4 is characteristic to be a positive regulator of ISR</p>
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### CONCLUSION:

ISR is effective against a broad range of pathogenic microorganism. It requires JA and ET as essential signalling molecules, rather than SA. The wild-type MAPK4 is a negative regulator of SAR gene expression and a positive regulator of ISR.

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