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Effect of Chemical Inducers of Systemic Acquired Resistance (SAR) for the Management of Late Blight Disease of Potato

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Authors' contributions

This work was carried out in collaboration among all authors. Author SHP designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors HSV and KAB both managed the analyses of the study and literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Late blight of potato is one of the most devastating diseases with great economic importance. Among several disease management strategies, ISR (Induced systemic resistance) plays an important role in checking the disease spread. So, in the present study, nine different SAR chemical inducers *viz*. Phosphoric acid, Salicylic acid, Naphthalene acetic acid(NAA), Benzoic acid, Benzothiadiazole, Dichloro-iso nicotinic acid (INA) and Kinetin were evaluated at 0.05% (C1), 0.10% (C2) and 0.15% (C3) concentrations, whereas Calcium chloride and Ascorbic Acid at 3.5% (C1), 5.0% (C2), 6.5% (C3) and 1.5% (C1), 2.0% (C2), 2.5% (C3) respectively at three different concentrations each, along with positive and negative checks of standard fungicide metalaxyl 8+ macozeb 64 WP @0.25% and control(water) respectively by spraying them at 2-3 compound leaf stage of plants which were challenged by *P. infestans* inoculations five days later in pots to assess disease incidence and intensity which were recorded at weekly intervals of four stages (stage I-IV). It was found that the foliar spray with benzothiadiazole (BTH) @ 0.10 to 0.15% was best in delaying the appearance of first symptoms of the disease by 11 days followed by salicylic acid@

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0.10 to 0.15% and ascorbic acid @ 2.5% which delayed the symptom appearance by 9 days, whereas the standard fungicidal spray of metalaxyl 8 + mancozeb 64 WP at the recommended concentration of 0.25% delayed the symptom appearance by only 8 days when compared to water sprayed check. It was also found that on an average, BTH sprays completely arrested the late blight development (0% intensity) followed by salicylic acid, INA and ascorbic acid sprays exhibiting 0.46-0.74% mean blight intensity compared to 2.00 and 6.28 percent intensity recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants one week after first appearance of disease in control pots (stage I). These treatments with chemical inducers showed an increase in the activity of peroxidases and polyphenol oxidases in the infected potato leaves as a result of SAR activity at all stages (stage I-IV). Average maximum peroxidase activity of 19.01-20.66 mg and polyphenol oxidase activity of 2.70-2.89 mg were recorded in the potato leaves during stage-I, sprayed with either BTH, benzoic acid, phosphoric acid or salicylic acid as compared to only (16.78 mg and 2.28 mg), (4.66 mg and 1.36 mg) recorded in (metalaxyl 8 + mancozeb 64 WP) and water-sprayed check respectively. The highest concentrations of all the test chemicals, in general, showed increased biochemical activity thereby yielding lesser blight intensity compared to their lower concentrations.

Keywords: The late blight of potato; Phytophthora infestans; SAR inducers; disease incidence and intensity.

1. INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the most important food crops worldwide which represents a valuable source of nutrients in a balanced diet. In terms of human consumption, the potato is the third most important food crop in the world, following only rice and wheat.

Late blight of potato is one of the deadly diseases causing devastation to the crop which leads to severe yield losses. For more than a decade, controlling late blight has become increasingly demanding because of the emergence of new strains of the pathogen. These new strains are known to be more aggressive and resistant to the novel fungicides. Moreover, indiscriminate use of fungicides poses a great threat to the environment and it was reported that consumption of these accumulated fungicidal toxic residues which are carcinogenic when entered into the food chain leads to many severe health complications [1].

Disease resistance in plants is dependent on both pre-existing physical or chemical barriers (such as thick cell walls or high quantities of lignin or tannins) and inducible defence mechanisms. Upon recognition of the attacker, inducible defences are activated at the site of infection as well as in distant uninfected tissues. Depending on the type of attack, the plant activates different signalling pathways to synthesize a specific set of defensive compounds. The resistance process, mediated by the accumulation of endogenous salicylic acid (SA), a metabolite downstream the biosynthetic pathway initiated by phenylalanine ammonia lyase (PAL), is called systemic acquired resistance (SAR) and is based on the induction of secondary metabolic pathways and the increased synthesis of products, phenolic compounds among them, by this metabolism as a response to pathogen attack.

In the absence of any attack, these defence mechanisms may be induced by physical or chemical elicitation. Application of chemical inducers or elicitors of defense signalling molecules is a promising way of disease management. Some chemicals were reported as inducers or elicitors of defense against several plant diseases. Chemical elicitors, such as Salicylic acid (SA), benzothiadiazole (BTH) (functional analogue of SA), harpin and 1-methylcyclopropane, among others, are agrochemicals that can mimic the action of the signaling molecules SA and JA and their derivates, or simulate the attack of a pathogen. These molecules may interact with receptors in the plant, activating defence responses and triggering. Some other chemicals like Ascorbic acid, Phosphoric acid, calcium chloride, INA, NAA were reported for inducing resistance in many plants against various fungal, bacterial and viral diseases [2].

Although chemical inducers/elicitors were first used to increase plant resistance to pathogens, it was found that the mechanism involved increased polyphenol levels. Consequently, elicitors can be regarded as an interesting

alternative for obtaining plants with higher polyphenol content. In addition to phenolics, these elicitors were found to stimulate the accumulation of signal molecules like hydrogen peroxide, reactive oxygen species, protein kinase, all of which play a crucial role in intracellular signalling pathways. Peroxidase enzymes are important in the production of reactive oxygen species, such as hydrogen peroxide and superoxide which have been linked to increased disease resistance in plants. Peroxidase also shows affinity to substrates involved in cellular lignification and the products of its activity have direct antimicrobial activity in the presence of hydrogen peroxide [3].

So the present study was focused upon the application of foliar sprays of different SAR chemical inducers *viz.* Phosphoric acid, Salicylic acid, Naphthalene acetic acid (NAA), Benzoic acid, Benzothiadiazole (BTH), Dichloro-iso nicotinic acid (INA), Kinetin, Calcium chloride and Ascorbic Acid which were tested at three different concentrations to elicit a systemic immune response in potato plant and thereby protecting the plant against the late blight pathogen by assessing the number of days for symptom appearance, incidence and intensity of late blight disease. SAR activity of these treatments was analyzed by the estimation of biochemical changes *viz.* peroxidases and polyphenol oxidases in the infected leaf tissues.

2. MATERIALS AND METHODS

2.1 Isolation of Pathogen and Production of Inoculums

Blighted samples were brought to the laboratory and the isolation of pure culture was made on V-8 agar medium using standard plant pathological techniques. A pure culture of *P. infestans* was transferred onto the V8 medium amended with rose Bengal in 90 mm diameter Petri dishes and incubated for about 14 days at 18°C in darkness for sporangia production. The inoculum was harvested from 10 to 14 days old cultures by adding 10 ml sterile distilled water to each plate and scraping the surface lightly with the edge of a glass rod to dislodge the sporangia. These sporangial suspensions were filtered through a double layer of cheesecloth to remove mycelial fragments and diluted to an appropriate concentration with the aid of Haemocytometer. The sporangia were chilled at 4°C for 2 hours to induce zoospore liberation.

2.2 Use of SAR Activators

2.2.1 Chemical inducers

A total of nine SAR chemical inducers viz Phosphoric acid, Salicylic acid, Naphthalene acetic acid (NAA), Benzoic acid, Benzothiadiazole (BTH), Dichloro-iso nicotinic acid (INA), Kinetin, Calcium chloride and Ascorbic Acid were evaluated at three different concentrations. Treatments with Phosphoric acid, Salicylic acid, Naphthalene acetic acid, Benzoic acid, Benzothiadiazole (BTH), Dichloro-iso nicotinic acid and Kinetin at 0.05% (C1), 0.10% (C2) and 0.15% (C3) concentrations, whereas Calcium chloride and Ascorbic Acid at 3.5% (C1), 5.0% (C2), 6.5% (C3) and 1.5% (C1), 2.0% (C2), 2.5% (C3) respectively, along with a positive check of metalaxyl 8+ mancozeb 64 WP @ 0.25% and a negative check of water sprayed plants (control) were evaluated in pots (30 cm dia) arranged in completely randomized design in a pot-house at experimental Farm of Division of Plant Pathology of the University. Each pot (30 cm dia), filled with pre-sterilized sandy loam field soil (autoclaved at 1.05 kg cm^{-2} atmospheric pressure for two hours) and planted with three equidistantly placed healthy whole seed tubers of potato cv. Kufri jyoti, represented a treatment and replicated thrice. Each SAR inducer was applied separately at three different concentrations at 2-3 compound leaf stage maintaining metalaxyl 8+ mancozeb 64 WP (0.25%) and water applied as positive and negative checks respectively. Spore/sporangia suspension of the pathogen *P. infestans* (2×10^4 spores/sporangia per ml) prepared from one-week-old culture actively growing on V-8 agar medium, was sprayed at the rate of 10 ml suspension per pot after 5 days of application of SAR inducers. All other practices for raising the crop were as per the recommended package. Observations on late blight incidence and intensity were recorded at weekly intervals after the first appearance of the disease in control pots. The estimation of chemical changes in plant tissue infected with the pathogen and also as a consequence of the application of SAR inducers was assessed seven days after symptom appearance in control pots (stage 1), 7 days later when these spots had enlarged (stage 2), further 7 days later when there were extensive leaf spots (stage 3) and again after 7 days from 3rd stage when spots had coalesced together forming necrotic patches (stage 4). The biochemical changes assessed include the changes in the levels of enzymes such as

polyphenol oxidases and peroxidases as described by Mahadevan and Sridhar using standard methods [4].

2.2.2 Estimation of polyphenol oxidase and peroxidase enzymes for SAR activity in the infected plant tissues

The selected samples of infected leaves were washed first under running tap water and then with double distilled water. Thereafter, estimation of peroxidases and polyphenol oxidases was done for these infected leaf samples.

For extraction of peroxidase enzyme 100 mg of each infected leaf tissue from each sample was ground with a pinch of neutral sand in a mortar at 0°C (kept in the deep-freeze); the extract was squeezed through double layer clean cloth and the filtrate centrifuged at 3000 rpm for 15 minutes in a refrigerated centrifuge (2-4°C). The supernatants were recovered as enzyme extract and kept separately in tubes in ice bath for assaying the per oxidase activity, 5 ml of freshly prepared pyrogallol reagent (prepared by mixing 10 ml of 0.5 m pyrogallol solution and 12.5 ml of 0.66 m phosphate buffer and volume made to 100 ml with distilled water) and 1.5 ml of enzyme extract were mixed in a spectrophotometer tube and the mixture immediately adjusted to zero absorbance of a spectrophotometer. 0.5 ml of 1% H_2O_2 solution was added to it and the contents mixed by inverting the tube. The change in the absorbance was recorded at 430 nm wave length immediately after addition of substrate at per minute interval up to 3 minutes to give an estimation of the peroxidase activity.

Extraction of polyphenol oxidase was done by homogenizing 100 mg of infected leaf tissue with pinch of sand in 6 ml phosphate buffer of 0.1 M at pH 7.0 at 0°C. The extract was filtered with a clean cloth, centrifuged at 3000 rpm for 15 minutes and stored in ice bath until used. The polyphenol oxidase activity was estimated by the method given by Sadasivam and Manickam. 2 ml of enzyme extract and 3 ml of distilled water were mixed together in a spectrophotometer tube and adjusted to zero absorbance of a spectrophotometer. 1 ml of catechol ($C_6H_6O_2$) solution (0.4 mg/ml) was added to the above mixture and the reactants were quickly mixed. The enzyme activity was measured as the change in absorbance per minute at 490 nm immediately after the addition of catechol solution which initiated the reaction up to 3 minutes [5].

2.2.3 Disease incidence

A total number of diseased and healthy plants were recorded at weekly intervals. Mathematically, the incidence was calculated as

$$\text{Late blight incidence (\%)} = \frac{n}{N} \times 100$$

Where n is the number of plants showing blight symptoms and N the total number of plants examined.

Table 1. Disease rating scale

Disease score	Score description in terms of foliage infected (%)
0	No visible symptoms
1	1-10
2	11-25
3	26-50
4	51-75
5	>75

2.2.4 Disease intensity

The observation on the extent of the foliage blighted was recorded at weekly intervals using the disease rating scale given by Mohan and Thind [6].

The disease intensity was calculated by using the following formula

$$\text{Late blight intensity (\%)} = \frac{\text{Summation of numerical rating}}{\text{No. of plants examined} \times \text{Maximum disease score}} \times 100$$

2.2.5 Statistical analysis

Statistical analysis was carried out using SPSS 11.0 design and the data was evaluated by ANOVA using the least significant differences test (LSD) at 5% ($P \leq 0.05$) probability level.

3. RESULTS AND DISCUSSION

3.1 Use of SAR Inducers

Various SAR inducers were sprayed at different concentrations on potato cv. Kufri Jyoti at 2-3 compound leaf stage and challenged by *P. infestans* inoculations five days later to assess the usefulness of these inducers in checking late blight development.

3.2 Effect on Disease Development

3.2.1 Disease incidence

The results (Table 1) revealed that foliar spray with benzothiadiazole (BTH) @ 0.10 to 0.15% was the best treatment in delaying the appearance of first symptoms of the disease by 11 days followed by salicylic acid @ 0.10 to 0.15% and ascorbic acid @ 2.5% which delayed the symptom appearance by about 9 days, whereas the standard fungicidal spray of metalaxyl 8 + mancozeb 64 WP at the recommended concentration of 0.25% delayed the symptom appearance by only 8 days compared to water sprayed check. The observation on late blight incidence recorded seven days after the initial symptom appearance in check pots (stage I) revealed a significant decrease in disease incidence compared to check by the application of SAR inducer. On an average, BTH sprays completely arrested the late blight development (0% incidence); salicylic acid, INA and ascorbic acid sprays were the next best SAR inducers exhibiting 3.7% mean blight incidence compared to 5.55 and 24.06 percent incidence recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The highest concentration of all the test chemicals, in general, yielded lesser blight incidence compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight incidence also existed.

Observations recorded seven days after initial symptom appearance in check (stage I), followed by three more observations (stage II to stage IV) at intervals of seven days; Phosphoric acid, Salicylic acid, Naphthalene acetic acid, Benzoic acid, Benzothiadiazole, Dichloro-iso nicotinic acid, and Kinetin evaluated at 0.05(C1),0.10(C2) and0.15(C3) concentration, Calcium chloride, Ascorbic Acid at 3.5 (C1), 5.0 (C2), 6.5 (C3) and 1.5 (C1), 2.0 (C2), 2.5 (C3) respectively, and metalaxyl 8+ macozeb 64 WP @0.25%. Figures in parenthesis are arc sine transformed values and mean of three replications.

No blight incidence was recorded in treatments sprayed with either BTH (C1-C2 concentration), salicylic acid or INA (both at C2-C3 concentrations). Ascorbic acid at C1-C2 concentrations and metalaxyl 8 + mancozeb 64 WP at 0.25% concentration were the next best treatments showing blight incidence of 5.55% compared to 22.22 -27.77% incidence recorded on water sprayed plants. The observation on late blight incidence recorded seven days after (stage I) revealed again a significant decrease in disease incidence compared to check by the application of SAR inducers. BTH, INA and Ascorbic acid depicted 14.81% incidence followed by those of salicylic acid, NAA and Benzoic acid sprays exhibiting 20.36-25.92% mean blight incidence compared to 27.77 and 39.62 percent incidence recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The highest concentration of all the test chemicals, in general, yielded lesser blight incidence compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight incidence also existed. Lesser blight incidence of 11.11 percent was recorded at C2-C3 concentration in treatments sprayed with BTH, INA, or ascorbic acid. Salicylic acid or phosphoric acid at C3 concentrations were the next best treatment showing blight incidence of 16.66 compared to 27.77 and 41.11% incidence recorded on water sprayed plants. A similar trend was recorded seven days after stage II which again revealed a significant decrease in disease incidence compared to check by the application of SAR inducers, BTH and ascorbic acid showed 14.81% incidence followed by those of salicylic acid and INA sprays exhibiting 18.51-25.92% mean blight incidence compared to 55.55 and 68.88 percent incidence recorded, respectively on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The highest concentration of all the test chemicals, in general, yielded lesser blight incidence compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight incidence also existed. Lesser blight incidence of 11.11 percent was recorded at (C3 concentration) in treatments sprayed with BTH, INA and ascorbic acid. Salicylic acid at C3 and BTH and INA at C2 concentrations were the next best treatments showing blight incidence of 22.22% compared to 55.55 and 66.66-68.88% incidence recorded, respectively, on metalaxyl 8

+ mancozeb 64 WP and water sprayed plants. The same trend was recorded seven days after stage III which revealed a significant decrease in disease incidence compared to check by the application of SAR inducers, BTH and ascorbic acid showed 24.06% incidence followed by those of salicylic acid and INA sprays exhibiting 29.62-31-47% mean blight incidence compared to 70.73 and 79.99 percent incidence recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The highest concentration of all the test chemicals, in general, yielded lesser blight incidence compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight incidence also existed. Lesser blight incidence of 16.66 percent was recorded at C3 concentration in treatments sprayed with BTH and ascorbic acid. Salicylic acid, ascorbic acid and INA at C2 and C3 concentrations were the next best treatment showing blight incidence of 22.22% compared to 63.33- 68.88 and 79.99-82.22% incidence recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water sprayed plants. On the overall basis, it was observed that foliar sprays with BTH showed the least disease incidence of 13.88% followed by ascorbic acid with 14.34%. Incidence as compared to 39.90 and 53.14 recorded on metalaxyl 8 + mancozeb 64 WP and water sprayed plants.

Observations recorded seven days after initial symptom appearance in control pots (stage I), followed by three more observations (stage II to stage IV) at intervals of seven days; Phosphoric acid, Salicylic acid, Naphthalene acid, Benzoic acid, Benzothiadiazole, Dichloro-iso nicotinic acid and Kinetin evaluated at 0.05 (C1), 0.10 (C2) and 0.15 (C3) concentration, calcium chloride, ascorbic acid at 3.5 (C1), 5.0 (C2), 6.5 (C3) and 1.5 (C1), 2.0 (C2), 2.5 (C3) respectively, and metalaxyl 8 + mancozeb 64 WP @ 0.25%. Figures in parenthesis are arc sine transformed values and mean of three replications.

An insight into the data (Figs. 1 & 2) revealed that the leaf blight incidence was not allowed to exceed beyond 20% up to 4th stage about a month after spraying with BTH and ascorbic acid compared to standard fungicide metalaxyl 8 + mancozeb 64 WP and water foliar spray which exhibited 71 and 80% disease incidence after the same period.

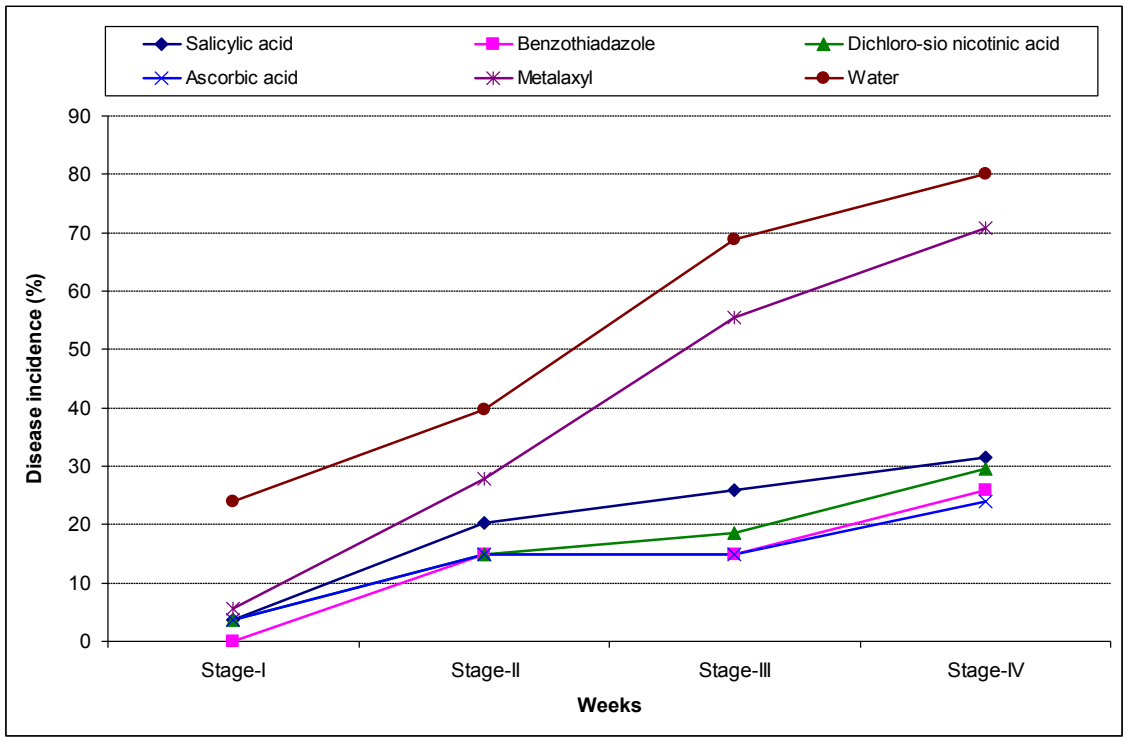

Fig. 1. Effect of foliar spray with SAR inducers on the incidence of late blight of potato cv. Kufri Jyoti planted in pots

Fig. 2. Evaluation of different treatments of SAR chemical inducers in pots

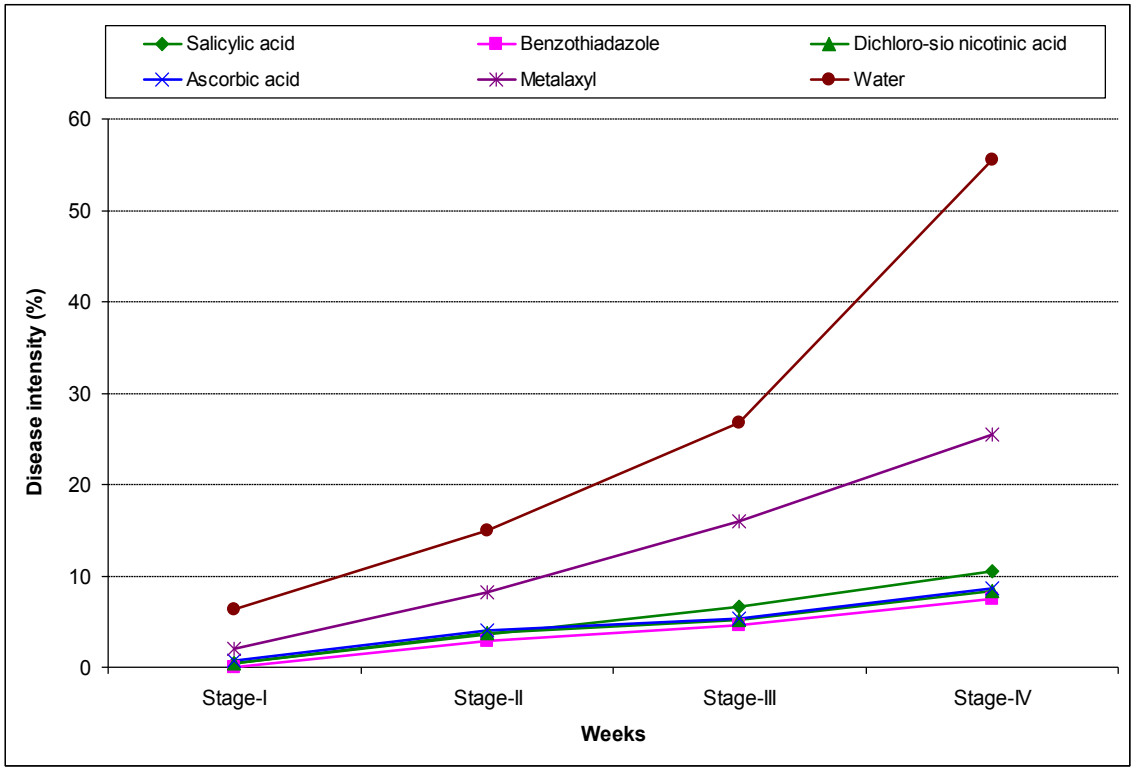

Fig. 3. Effect of foliar spray with SAR inducers on the intensity of late blight of potato cv. Kufri Jyoti planted in a pot

Table 2. Effect of foliar spray of SAR chemicals at different concentrations on the incidence of late blight of potato cv. Kufri Jyoti planted in pots

Chemical	Days to symptoms appearance	Late blight incidence (%)																Mean
		Stage I				Stage II				Stage III				Stage IV				
		C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	
Phosphoric acid	5-8	11.11 (16.06)	11.11 (16.06)	5.55 (8.03)	9.25 (13.38)	33.33 (35.26)	22.21 (27.81)	16.66 (19.78)	24.07 (27.62)	49.99 (44.99)	38.88 (38.50)	38.88 (38.50)	42.59 (40.67)	55.55 (48.24)	44.44 (41.75)	38.88 (38.50)	46.29 (42.83)	30.53 (31.12)
Salicylic acid	5-7	11.11 (16.06)	0.00 (0.00)	0.00(-0.00)	3.70 (5.35)	22.22 (27.81)	22.22 (27.81)	16.66 (19.78)	20.36 (25.13)	33.33 (34.78)	22.22 (27.81)	22.22 (27.81)	25.92 (30.13)	38.88 (38.50)	27.77 (31.53)	27.77 (31.53)	31.47 (33.86)	20.36 (23.62)
Naphthalene Acetic acid (NAA)	5-6	11.11 (16.06)	11.11 (16.06)	11.11 (16.06)	11.11 (16.06)	33.33 (34.78)	22.22 (27.81)	22.22 (23.50)	25.92 (28.70)	38.88 (38.50)	38.88 (38.50)	38.88 (38.50)	38.88 (38.50)	44.44 (41.75)	50.00 (45.00)	38.88 (38.50)	44.44 (41.75)	30.09 (31.25)
Benzoic acid	7-8	16.66 (19.61)	11.11 (16.06)	11.11 (16.06)	12.88 (17.24)	27.77 (31.35)	22.22 (27.81)	22.22 (27.81)	24.06 (29.05)	38.88 (38.50)	38.88 (38.50)	38.88 (38.50)	38.88 (38.50)	50.00 (45.00)	44.44 (41.75)	38.88 (38.50)	44.44 (41.75)	30.07 (31.64)
Benzothiadiazole (BTH)	8-11	0.00(-0.00)	0.00(-0.00)	0.00(-0.00)	0.00 (-0.00)	22.22 (27.81)	11.11 (16.06)	11.11 (16.06)	14.81 (19.97)	22.22 (27.81)	11.11 (16.06)	11.11 (16.06)	14.81 (19.97)	27.77 (31.53)	27.77 (31.53)	22.22 (27.81)	24.06 (30.29)	13.88 (17.56)
Calcium chloride	5-7	27.77 (31.53)	22.22 (27.81)	22.22 (23.50)	24.07 (27.62)	38.33 (38.50)	33.33 (35.26)	27.77 (31.53)	33.33 (35.10)	44.33 (41.75)	38.88 (38.50)	38.88 (38.50)	40.73 (39.59)	55.55 (48.24)	50.00 (45.00)	38.88 (38.50)	48.14 (43.91)	36.57 (36.55)
Dichloro-iso nicotinic acid (INA)	5-8	11.11 (11.75)	0.00 (-0.00)	0.00 (-0.00)	3.70 (3.91)	22.22 (27.81)	11.11 (16.06)	11.11 (11.75)	14.81 (18.54)	33.33 (34.77)	11.11 (16.06)	11.11 (16.06)	18.51 (22.29)	44.44 (41.75)	22.22 (27.81)	22.22 (27.81)	29.62 (32.46)	16.66 (19.30)
Ascorbic acid	6-9	5.55 (8.03)	5.55 (8.03)	0.00 (-0.00)	3.70 (5.35)	22.22 (23.50)	11.11 (!6.06)	11.11 (16.06)	14.81 (18.54)	22.22 (27.81)	11.11 (16.06)	11.11 (16.06)	14.81 (19.97)	33.33 (34.78)	22.20 (27.80)	16.66 (24.09)	24.06 (28.89)	14.34 (18.19)
Kinetin	6-8	22.22 (27.81)	16.66 (24.09)	16.66 (19.78)	18.51 (23.89)	38.88 (38.50)	27.77 (31.53)	27.77 (31.53)	31.47 (33.86)	44.44 (41.75)	38.88 (38.50)	38.88 (38.50)	40.73 (39.59)	55.55 (48.24)	49.99 (44.99)	38.88 (38.50)	48.14 (43.91)	34.71 (35.31)
Metalaxyl 8 + mancozeb 64 WP	5-7	5.55 (8.03)	5.55 (8.03)	5.55 (8.03)	5.55 (8.03)	27.77 (31.53)	27.77 (31.53)	27.77 (31.53)	27.77 (31.53)	55.55 (48.24)	55.55 (48.24)	55.55 (48.24)	55.55 (48.24)	79.99 (67.88)	68.88 (56.12)	63.33 (52.88)	70.73 (58.96)	39.90 (36.69)
Water	4-5	22.22 (27.81)	27.77 (31.53)	22.22 27.81)	24.06 (29.05)	38.88 (38.05)	38.88 (38.05)	41.11 (39.83)	39.62 (38.94)	66.66 (54.73)	68.88 (56.12)	71.11 (57.51)	68.88 (56.12)	77.77 (66.48)	82.22 (69.27)	79.99 (67.88)	79.99 (67.88)	53.14 (48.00)
Mean		13.10 (16.61)	10.09 (13.42)	8.58 (10.84)		29.79 (32.32)	22.72 (26.93)	21.41 (24.47)		40.90 (39.42)	34.03 (33.90)	34.23 (34.02)		51.21 (46.58)	44.54 (42.05)	38.78 (38.59)		
		S.Em±	CD (0.05)			S.Em±	CD (0.05)			S.Em±	CD (0.05)			S.Em±	CD (0.05)			
SAR		0.26	O.73			0.22	0.81			0.20	0.81			0.28	0.80			
Concentration		0.33	I.O5			0.28	1.22			0.31	1.02			0.42	1.32			
F x C		0.54	1.84			0.49	1.83			0.46	1.67			0.55	1.88			

3.2.2 Disease intensity

The observation on late blight intensity (Table 2) recorded seven days after the initial symptom appearance in check pots (stage I), revealed a significant decrease in disease intensity compared to check by the application of SAR inducer.

On an average BTH spray completely arrested the late blight development (0% intensity); salicylic acid, INA, and ascorbic acid sprays were the next best SAR inducers exhibiting 0.46-0.74% mean blight intensity compared to 2.00 and 6.28 percent intensity recorded, respectively, on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The highest concentrations of all the test chemicals, in general, yielded lesser blight intensity compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight intensity also existed. No blight intensity was recorded in treatments sprayed with either BTH (C1-C3 concentration), salicylic acid, or INA (both at C2 and C3 concentration) or ascorbic acid (C3 concentration). Benzoic acid, ascorbic acid at C2 concentration, and metalaxyl 8 + mancozeb 64 WP at 0.25% concentration were the next best treatments showing blight incidence of 1.02-1.38 and 2.01-2.10 percent compared to 6.28% disease intensity recorded on the water- sprayed plants.

The disease intensity recorded, a week after the stage 1 observation again revealed a significant decrease in disease intensity compared to check; BTH and salicylic acid sprays exhibited 2.83-3.61 percent blight intensity. Ascorbic acid and INA were the next best SAR inducers, exhibiting 3.77 and 3.96% mean blight intensity compared to 8.16 and 14.94 percent intensity recorded in on metalaxyl 8 + mancozeb 64 WP and water-sprayed plants. The higher concentrations of all the test chemicals, in general, yielded lesser blight intensity compared to their lower concentrations. A significant interaction was observed between the SAR inducers and their concentrations in reducing blight intensity. A blight incidence of 2.48% was recorded in treatments sprayed with either BTH. Salicylic acid, INA, or ascorbic acid at C3 concentration exhibited 3.51- 3.61 percent disease intensity as compared to 7.98-8.29% and 14.10-16.01% recorded on metalaxyl 8 + mancozeb 64 WP and water- sprayed check plants. A similar trend was recorded seven days after stage II revealed, a significant decrease in disease intensity compared to check. BTH sprays exhibited 4.67 percent blight intensity. INA and ascorbic acid were the next best SAR inducers exhibiting 5.20-5.38 mean blight intensity compared to 15.95 and 26.78 percent incidence recorded on metalaxyl 8 + mancozeb 64 WP and water sprayed plants. The higher concentrations of all the test chemicals, in general, yielded lesser blight intensity compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight intensity existed at this stage too. Blight intensity of 3.88- 3.95% was recorded in treatments sprayed with BTH at C2-C3 concentration followed by ascorbic acid at the same concentration as compared to 15.48-16.30% and 26.02-28.16% disease intensity recorded on metalaxyl 8 + mancozeb 64 WP and water- sprayed plants.

The disease intensity recorded a week after the stage III, revealed a significant decrease in disease intensity compared to check. BTH exhibited a 7.46 percent blight intensity. INA and ascorbic acid were the next best SAR inducers exhibiting 8.31 and 8.61 percent disease intensity. The higher concentrations of all the test chemicals, in general, yielded lesser blight intensity compared to their lower concentrations. A significant interaction between the SAR inducers and their concentrations in reducing blight intensity existed at this stage too. Blight intensity of 6.20-6.32% was recorded in treatments sprayed with BTH at C2-C3 concentration followed by ascorbic acid and INA 7.51-7.79% at the same concentration as compared to 24.76-25.72% and 54.83-55.05% disease intensity recorded on metalaxyl 8 + mancozeb 64 WP and water- sprayed plants.

An insight into the data (Fig. 3) revealed that the leaf blight intensity was not allowed to exceed beyond 7% about a month after the spraying SAR inducers stage IV compared to standard fungicide metalaxyl 8 + mancozeb 64 WP and water foliar spray which exhibited 13 and 26% disease intensity after the same period.

3.2.3 Induction of Biochemical changes by SAR chemical inducers in the infected potato leaves peroxidase

The peroxidases in potato leaves of infected plant were estimated at weekly intervals after spraying SAR chemicals followed by the inoculation of the late blight pathogen *P. infestans*, maintaining metalaxyl 8 + mancozeb 64 WP and water sprays as positive and negative checks respectively.

Table 3. Effect of foliar spray of SAR chemicals at different concentrations on the intensity of late blight of potato cv. Kufri Jyoti planted in pots

Chemical	Late blight intensity (%)																Mean
	Stage I				Stage II				Stage III				Stage IV				
	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	
Phosphoric acid	1.80 (6.26)	1.60 (5.87)	1.60 (4.21)	**1.60 (5.54)**	4.27 (11.73)	4.15 (11.57)	4.15 (9.54)	**4.19 (10.95)**	8.64 (17.02)	6.38 (14.51)	6.30 (14.50)	**7.10 (15.34)**	13.82 (21.55)	10.20 (18.36)	10.08 (18.33)	**11.36 (19.42)**	**6.08 (12.79**
Salicylic acid	1.48 (5.62)	0.00 (0.00)	0.00 (0.00)	**0.49 (1.87)**	3.78 (11.11)	3.45 (10.67)	3.61 (8.96)	**3.61 (10.25)**	7.98 (16.35)	5.89 (13.88)	5.78 (13.88)	**6.55 (14.70)**	12.76 (20.85)	9.42 (17.56)	9.24 (17.12)	**10.47 (18.51)**	**5.28 (11.33)**
Naphthalene Acetic acid (NAA)	1.78 (6.22)	1.64 (5.99)	1.64 (6.00)	**1.68 (6.07)**	4.98 (12.77)	4.84 (12.70)	4.64 (12.42)	**4.82 (12.63)**	7.66 (16.03)	6.50 (14.75)	6.37 (14.60)	**6.84 (15.13)**	12.25 (20.09)	10.40 (18.35)	10.17 (18.32)	**10.94 (18.92)**	**6.07 (13.19)**
Benzoic acid	1.82 (6.31)	1.37 (5.40)	1.33 (5.35)	**1.50 (5.71)**	4.82 (12.65)	4.64 (12.43)	2.84 (9.69)	**4.10 (11.59)**	7.83 (16.21)	6.20 (14.33)	6.13 (14.30)	**6.72 (14.97)**	10.19 (17.90)	9.65 (17.83)	9.80 (17.94)	**9.88 (17.90)**	**5.55 (12.54)**
Benzothiadiazole (BTH)	0.00 (-0.00)	0.00 (0.00)	0.00 (0.00)	**0.00 (-0.00)**	3.44 (10.63)	2.57 (7.54)	2.48 (7.40)	**2.83 (8.52)**	6.18 (14.34)	3.95 (11.42)	3.88 (11.35)	**4.67 (12.73)**	9.86 (17.67)	6.32 (14.37)	6.20 (14.33)	**7.46 (15.45)**	**3.74 (9.09)**
Calcium chloride	1.78 (7.60)	1.66 (7.35)	1.64 (6.01)	**1.69 (6.99)**	5.33 (13.32)	5.24 (13.18)	5.25 (13.22)	**5.27 (13.24)**	9.67 (18.04)	7.48 (15.82)	7.40 (15.73)	**8.18 (16.53)**	15.47 (22.92)	11.95 (20.14)	11.84 (19.80)	**13.08 (20.95)**	**7.05 (14.34)**
Dichloro-iso nicotinic acid (INA)	1.38 (3.91)	0.00 (0.00)	0.00 (0.00)	**0.46 (1.30)**	4.12 (11.68)	3.83 (9.18)	3.60 (6.17)	**3.77 (9.01)**	6.01 (14.15)	4.87 (10.42)	4.72 (10.28)	**5.20 (11.62)**	9.61 (17.65)	7.79 (15.02)	7.55 (15.67)	**8.31 (16.11)**	**4.43 (9.51)**
Ascorbic acid	1.21 (3.66)	1.02 (3.35)	0.00 (0.00)	**0.74 (2.34)**	4.81 (12.65)	3.58 (10.81)	3.51 (8.81)	**3.96 (10.75)**	6.67 (14.93)	4.78 (12.60)	4.70 (10.17)	**5.38 (12.56)**	10.67 (18.73)	7.64 (15.82)	7.52 15.61)	**8.61 (16.72)**	**4.67 (10.59)**
Kinetin	1.89 (7.89)	1.49 (6.78)	1.53 (5.79)	**1.63 (6.82)**	4.01 (11.48)	4.70 (12.45)	4.70 (12.39)	**4.47 (12.11)**	7.71 (16.05)	5.90 (5.90)	5.87 (13.93	**6.49 (14.65)**	11.47 (19.61)	9.44 (17.47)	9.29 (17.27)	**10.06 (18.11)**	**5.66 (12.92)**
Metalaxyl 8 + mancozeb 64 WP	2.01 (4.73)	1.90 (4.60)	2.10 (4.84)	**2.00 (4.72)**	8.29 (16.43)	7.98 (16.30)	8.21 (16.62)	**8.16 (16.45)**	16.30 (23.76)	15.48 (23.11)	16.08 (23.55)	**15.95 (23.47)**	26.08 (30.28)	24.76 (29.48)	25.72 (30.18)	**25.52 (29.98)**	**12.90 (18.66)**
Water	6.39 (14.51)	6.12 (14.20)	6.35 (14.55)	**6.28 (14.42)**	14.10 (21.96)	14.72 (22.51)	16.01 (23.47)	**14.94 (22.65)**	26.02 (30.57)	26.02 (30.57)	28.16 (32.00)	**26.78 (31.09)**	56.68 (48.94)	54.83 (47.92)	55.05 (48.05)	**55.52 (48.30)**	**25.88 (29.11)**
Mean	**1.95 (6.06)**	**1.52 (4.87)**	**1.47 (4.25)**	**1.65 (5.06)**	**5.63 (13.31)**	**5.42 (12.67)**	**5.34 (11.70)**	**5.46 (12.56**	**10.74 (17.96)**	**8.49 (15.95)**	**8.67 (15.85)**	**9.08 (16.59)**	**17.16 (23.29)**	**14.76 (21.12)**	1.08	**15.56 (21.85**	
	S.Em±	**CD (0.05)**			**S.Em±**	**CD (0.05)**			**S.Em±**	**CD (0.05)**			**S.Em±**	**CD (0.05)**			
SAR	0.08	0.47			0.11	0.48			0.15	0.68			0.14	0.44			
Concentration	0.41	1.09			0.23	0.97			0.23	1.27			0.21	1.87			
F x C	0.66	1.97			0.63	1.69			0.70	2.09			0.63	1.91			

Table 4. Estimation of peroxidase in potato leaves after application of SAR inducer chemicals and inoculated with *P. infestans*

Chemical	Peroxidase activity (mg)															
	Stage I				Stage II				Stage III				Stage IV			
	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean
Phosphoric acid	17.40	19.00	21.15	**19.18**	19.70	22.90	24.35	**22.23**	19.00	22.30	24.15	**21.81**	18.20	21.85	23.95	**21.31**
Salicylic acid	17.60	21.20	23.20	**20.66**	19.90	24.70	27.40	**24.00**	19.30	24.10	27.00	**23.57**	19.00	24.75	26.20	**23.22**
Naphthalene Acetic acid (NAA)	14.50	18.00	20.45	**17.65**	16.90	20.65	24.82	**20.79**	16.50	20.18	23.60	**20.09**	16.20	19.90	23.20	**19.83**
Benzoic acid	17.00	17.30	22.75	**19.01**	18.50	22.80	26.95	**22.75**	18.05	22.45	26.03	**22.21**	16.90	22.30	25.15	**21.67**
Benzothiadiazole (BTH)	14.40	18.00	20.15	**17.51**	17.80	20.30	25.65	**21.25**	17.20	20.10	24.85	**20.71**	16.95	19.85	26.85	**20.77**
Calcium chloride	16.30	16.65	21.55	**18.16**	20.60	20.70	28.45	**23.25**	19.20	20.15	27.95	**22.43**	19.00	19.90	26.85	**21.91**
Dichloro –iso nicotinic acid (INA)	16.20	17.80	21.73	**18.57**	17.95	20.10	24.75	**20.93**	17.55	19.88	24.03	**20.48**	17.05	19.35	23.45	**19.95**
Ascorbic acid	15.90	17.30	22.75	**18.65**	17.65	20.22	28.15	**22.00**	17.32	19.80	27.50	**21.54**	17.20	19.40	27.15	**21.25**
Kinetin	19.95	17.76	22.90	**18.87**	19.84	20.45	27.95	**22.74**	16.35	20.25	27.15	**21.25**	15.95	19.15	26.85	**20.65**
Metalaxyl 8 + mancozeb 64 WP	16.15	16.20	18.00	**16.78**	18.20	19.90	23.66	**20.55**	17.90	19.10	19.10	**18.70**	17.20	18.75	23.15	**19.70**
Water	4.35	4.75	4.88	**4.66**	7.42	7.10	7.70	**7.40**	7.01	7.92	7.50	**7.47**	6.80	7.70	7.10	**7.50**
Mean	**15.06**	**16.72**	**19.95**		**17.67**	**19.97**	**24.50**		**16.86**	**19.65**	**23.56**		**16.47**	**19.41**	**23.50**	
	SEm±	**CD(0.05)**			**SEm±**	**CD (0.05)**			**SEm±**	**CD(0.05)**			**SEm±**	**CD (0.05)**		
Chemical	0.02	0.21			0.11	0.26			0.16	0.20			0.01	0.27		
Concentration	0.07	0.43			0.06	0.47			0.05	0.47			0.15	0.49		
Chemical x concentration	0.05	0.47			0.04	0.79			0.09	0.89			0.31	0.81		

Table 5. Estimation of polyphenol oxidase in potato leaves after application of SAR inducer chemicals and inoculated with *P. infestans*

Treatment	Polyphenol oxidase activity (mg)															
	Stage I				Stage II				Stage III				Stage IV			
	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean	C1	C2	C3	Mean
Phosphoric acid	2.28	2.72	3.11	**2.70**	2.37	2.78	3.27	**2.80**	2.28	2.53	3.04	**2.61**	2.25	2.44	3.15	**2.61**
Salicylic acid	2.35	2.70	3.48	**2.84**	2.53	3.06	3.85	**3.14**	2.49	2.46	3.78	**2.91**	2.35	2.62	2.53	**2.50**
Naphthalene Acetic acid (NAA)	2.32	2.40	2.97	**2.56**	2.37	2.51	3.47	**2.78**	2.30	2.46	3.38	**2.71**	2.22	2.32	3.19	**2.57**
Benzoic acid	2.45	2.89	3.35	**2.89**	2.52	3.01	3.81	**3.11**	2.31	2.65	3.51	**2.80**	2.29	2.52	2.39	**2.40**
Benzothiadiazole (BTH)	2.41	2.98	3.11	**2.83**	2.41	3.23	3.58	**3.07**	2.36	2.19	3.45	**3.00**	2.22	3.12	3.18	**2.84**
Calcium chloride	2.28	2.32	2.87	**2.49**	2.28	2.55	3.08	**2.63**	2.22	2.50	2.91	**2.54**	2.19	2.42	2.77	**2.46**
Dichloro-iso nicotinic acid (INA)	2.40	2.65	2.76	**2.60**	2.42	2.80	3.08	**2.76**	2.33	2.76	3.02	**2.70**	2.30	2.64	2.89	**2.60**
Ascorbic acid	2.28	2.88	3.10	**2.78**	2.44	2.98	3.18	**2.75**	2.41	2.91	3.17	**2.83**	2.38	2.73	3.10	**2.69**
Kinetin	2.28	2.42	3.15	**2.61**	2.28	2.57	3.36	**2.73**	2.22	2.40	2.90	**2.50**	2.16	2.20	3.23	**2.53**
Metalaxyl 8 + mancozeb 64 WP	2.33	2.32	2.20	**2.28**	2.21	2.27	2.31	**2.25**	2.27	2.43	2.41	**2.35**	2.23	2.25	2.27	**2.24**
Water	1.31	1.44	1.38	**1.36**	1.47	1.35	1.65	**1.49**	1.58	1.70	1.66	**1.54**	1.38	1.44	1.37	**1.39**
Mean	**2.25**	**2.52**	**2.86**		**2.30**	**2.61**	**3.14**		**2.24**	**2.51**	**3.06**		**2.17**	**2.42**	**2.72**	
	SEm±		**CD (0.05)**		**SEm±**		**CD(0.05)**		**SEm±**		**CD(0.05)**		**SEm±**		**CD(0.05)**	
Chemical	0.03		0.22		0.12		0.29		0.07		0.12		0.12		0.10	
Concentration	0.09		0.41		0.05		0.44		0.09		0.26		0.15		0.25	
Chemical x concentration	0.15		0.69		0.19		0.73		0.11		0.59		0.31		0.51	

The results on the peroxidase activity in potato leaves after the spray of test chemicals followed by inoculation with *P. infestans* estimated after 7 days of symptom appearance in control pots (stage I) (Table 4) revealed an average maximum peroxidase of 19.01-20.66 mg in the potato leaves sprayed with either BTH, benzoic acid, phosphoric acid or salicylic acid as compared to only 16.78 mg and 4.66mg recorded in metalaxyl 8 + mancozeb 64 WP and water- sprayed checks, respectively. On an average, higher concentration (C2-C3) exhibited significantly greater peroxidase activity compared to lower concentration (C1). A significant interaction between the chemicals and their concentrations in increasing the peroxidase activity in potato leaves also existed. Benzoic acid, Phosphoric acid, BTH, INA and salicylic acid at C1-C2 concentration and benzoic acid, ascorbic acid, Kinetic and salicylic acid sprayed at only C3 concentration resulted in maximum peroxidases of 17.00-21.20 mg compared to 16.15-18.00 and 4.35-4.88 mg obtained respectively in metalaxyl 8 + mancozeb 64 WP and water sprayed plants. Seven days later (Stage II), the peroxidase activity was again measured and found maximum on an average in potato leaves sprayed with either ascorbic acid, phosphoric acid, and salicylic acid (22.80-24.00 mg) compared to metalaxyl 8 + mancozeb 64 WP (16.20 mg) and water sprays (4.75 mg). Higher concentration, on an average, was superior to lower concentrations in increasing the peroxidases. A significant interaction was recorded at this stage also between chemicals and their concentrations. Ascorbic acid, kinetin, benzoic acid, calcium chloride and salicylic acid at C3 concentration resulted in maximum peroxidase activity of 26.95-28.45mg followed by benzoic acid, phosphoric acid and salicylic acid at C2 concentration showing peroxidase of 22.8-24.70 mg compared to 19.90-23.66 mg and 7.70-7.10 mg activity obtained in metalaxyl 8 + mancozeb 64 WP and water- sprayed check plants. Similarly, at crop stage III, an average maximum peroxidase activity of 21.81- 23.57 mg was recorded in leaves sprayed with either phosphoric acid, ascorbic acid, benzoic acid, calcium chloride or salicylic acid, whereas phosphoric acid, ascorbic acid, benzoic acid, calcium chloride and salicylic exhibited maximum peroxidase of 21.25-23.22 mg at stage IV. The higher concentrations were found superior to lower (C1 and C2) concentrations at both the stages III and IV with kinetin showing least estimates of peroxidase (15.95 mg) in C1 concentration at stage IV.

Observation recorded seven days after initial symptom appearance in control pots (stage I), followed by three more observations (stage II to stage IV) at intervals of seven days ; phosphoric acid, salicylic acid, naphthalene acetic acid, Benzoic acid, benzothiadiazole, dichloro-iso nicotinic acid and kinetin evaluated at 0.05 (c1), 0.10 (c2) and 0.15 (c3) concentration, calcium chloride, ascorbic Acid at 3.5 (c1), 5.0 (c2), 6.5 (c3) and 1.5 (c1), 2.0 (c2), 2.5 (c3) respectively, and metalaxyl 8 + mancozeb 64 WP @ 0.25%. Figures in parenthesis are arc sine transformed values and mean of three replications Polyphenol oxidase.

3.2.4 Polyphenol oxidase

The polyphenol oxidases were estimated in potato leaves at weekly intervals after spraying SAR chemicals followed by the inoculation of the late blight pathogen, *P. infestans,* maintaining metalaxyl 8 + mancozeb 64 WP and water sprays treatments as positive and negative checks respectively.

Results on the polyphenol oxidase activity in potato leaves after the spray of test chemicals followed by inoculation with *P. infestans* estimated after 7 days of symptom appearance in control pot (stage I) (Table 5), revealed an average maximum polyphenol oxidase activity of 2.70-2. 89 mg in the potato leaves sprayed with either benzoic acid, BTH, salicylic acid, ascorbic acid or phosphoric acid as compared to only 1.36 and 2.28 mg recorded in water and metalaxyl 8 + mancozeb 64 WP sprayed leaves. On an average, the higher concentration (C2 and C3) exhibited significantly more polyphenol oxidase estimates compared to lower concentration (C1). A significant interaction between the chemicals and their concentrations for the increased levels of polyphenol oxidase also existed. Benzoic acid, BTH, and ascorbic acid, phosphoric acid and kinetin at C3 concentration resulted maximum polyphenol oxidase activity of 3.10-3.48 mg compared to 2.20 -2.33 and 1.31 -1.44 mg activity obtained, respectively, in metalaxyl 8 + mancozeb 64 WP and water sprayed plants. Seven days later (stage II), the polyphenol oxidase activity in potato leaves estimated after spraying with the test chemicals revealed an average maximum polyphenol oxidase of 3.07-3.14 mg in the potato leaves sprayed with either benzoic acid, salicylic acid, and BTH, as compared to 2.25 and 1.49 mg recorded in metalaxyl 8 + mancozeb 64 WP and water sprayed leaves respectively. On an average, the

higher concentration (C2-C3) exhibited significantly more polyphenol oxidases compared to lower concentration (C1). A significant interaction between the chemicals and their concentrations in increasing the polyphenol oxidase also existed. Benzoic acid, BTH, salicylic and ascorbic acid at C3 concentration exhibited maximum polyphenol oxidase of 3.47-3.85 mg compared to 1.65 and 2.31 mg activity obtained, respectively, in metalaxyl 8 + mancozeb 64 WP and water sprayed plants.

Observations recorded seven days after initial symptom appearance in control pots (stage I), followed by three more observations (stage II to stage IV) at intervals of seven days ; Phosphoric acid, Salicylic acid, Naphthalene acetic acid, Benzoic acid, Benzothiadiazole, Dichloro-iso nicotinic acid and Kinetin evaluated at 0.05 (C1), 0.10 (C2) and 0.15 (C3) concentration, Calcium chloride, Ascorbic Acid at 3.5 (C1), 5.0 (C2), 6.5 (C3) and 1.5 (C1), 2.0 (C2), 2.5 (C3) respectively, a Metalaxyl 8 + mancozeb 64 WP @ 0.25%. Figures in parenthesis are arc sine tranformed values and mean of three replications. Similarly, at stage III, the polyphenol oxidase was again estimated and found maximum on an average in potato leaves sprayed with either salicylic acid, benzoic acid ascorbic acid or BTH (2.80-3.00) compared to metalaxyl MZ (2.35 mg) and water (1.54 mg) sprays. The C3 concentration on an average was superior to lower concentration in increasing the polyphenol oxidase. A significant interaction existed at this stage also between chemicals and their concentration. Salicylic acid, Benzoic acid, BTH, and NAA at C3 concentration resulted in maximum polyphenol oxidase of 3.38 -3.78 mg compared to 2.41 and 1.54 mg obtained in metalaxyl 8 + mancozeb 64 WP and water-sprayed check plants. Similarly, at crop stage IV an average maximum polyphenol oxidase was found in potato leaves sprayed with either ascorbic acid or BTH (2.69-2.80) followed by that with ascorbic acid, phosphoric acid, salicylic acid, NAA, INA or kinetin (2. 50-2. 61) compared to metalaxyl 8 + mancozeb 64 WP (2.24 mg) and water (1.39 mg) sprays. The C3-concentration on an average was superior to lower concentrations in increasing the polyphenol oxidase. A significant interaction was recorded at this stage also between chemicals and their concentration. Salicylic acid, Benzoic acid, BTH, NAA and phosphoric acid at C3 concentration resulted in maximum polyphenol oxidase of (3.10-3.19 mg) compared to 2.27 and 1.37mg obtained in metalaxyl 8 + mancozeb 64 WP and water-sprayed check plants.

The use of SAR inducers has been gaining significant importance since a long time ago due to their eco-friendly nature with no adverse effects on human health and ecology. Foliar sprays with SAR inducers in the present study have been found to result in a significant decrease in the incidence and intensity of potato late blight. Foliar spray with BTH caused the maximum delay (8-11 days) in initial symptom appearance followed by salicylic acid (6-9 days) compared to check. BTH, INA, or salicylic acid sprays further exhibited reduced blight intensity compared to check, indicating thereby that a spray or two with these chemicals can help contain the disease without any deleterious effects on ecology and environment. Several direct and indirect mechanisms of the action of SAR chemical inducers were studied where they were reported to associate with the production of increased levels of phenolic compounds and phenol related enzymes and also act as signalling molecules by interacting with receptors in the plant thereby activating or triggering defence responses. Biochemical changes triggered by the application of these chemicals were studied in many host pathogen combinations [7,8]. Attempts were made during the present investigation to unveil the changes in the peroxidases and polyphenol oxidases as a result of application of SAR inducers challenged with the pathogen. Our present studies revealed increased peroxidase and polyphenol oxidase in potato leaves applied with the test SAR inducers challenged with the test pathogen proved that increased activity of peroxidase and polyphenol oxidase enzymes is an indication of induction of defence mechanism operating in the plant system resulting ultimately in reduced infection and prolonged incubation periods ultimately resulting in reduced rate of disease development. The increased activity of these enzymes was also demonstrated by several other workers for many host species [9,10]. Kumar et al. [11] reported that SAR inducer salicylic acid caused 52.4 and 22.4% increase in peroxidase and polyphenol oxidase activity.

The use of SAR inducers for controlling many plant diseases has been made by several other workers. Astha and Sekhon [12] reported that SAR elicitors, especially SA and BTH, stimulated defence reactions in potato against late blight and muskmelon against downy mildew disease and thereby decreasing the disease severities in

those plants. They reported that this effect might be due to the impact of these substances on enzymatic activity in potato and muskmelon plants which is in agreement with our findings where we obtained the increase in peroxidase enzymes. Their increased amount leads to the production of more reactive oxygen species, such as hydrogen peroxide and superoxide which have been linked to increased disease resistance in plants. Kazemi [13] studied foliar application with salicylic acid on growth, flowering, yield, fruit quality, and resistance against diseases in tomato and demonstrated its positive correlation with those attributes. The reduction of disease severity in potato and many other crops by SA, BTH, and INA application may be attributed to the role of elicitors in the induction of local and/or systemic acquired resistance (SAR) in treated plants against the invasion of the pathogen as they stimulate the accumulation of signal molecules such as jasmonic acid, SA, hydrogen peroxide, reactive oxygen species and protein kinase, all of which play a crucial role in intracellular signalling pathways [14]. Nadia et al. [15] reported that foliar spray treatments with Ascorbic acid (AA), Dichloro-isonicotinic acid (INA) and calcium chloride reduced late and early blight severities more than 75.0- 82.1% under greenhouse and even field conditions by increasing the activity of *ß*-1,3-glucanase and chitinase enzymes. Our findings were also in agreement with Farouk et al. [16], Buonaurio et al. [17], Ghandi and Anfoka [18], Mur et al. [19], Cole [20], Lawton et al. [21] and Cohen et al. [22] who also demonstrated that BTH and other SAR inducers control various Phytophthora diseases of different crops.

4. CONCLUSION

The efficiency of these SAR elicitors over the prescribed conventional methods of controlling the disease in addition to their safety to the environment, make them an excellent alternative to the chemical treatment. Also, from our present findings it can be concluded that SAR elicitors can play a very important role in defence response against various pathogens. Therefore, treatment with only a few of these SAR elicitors brings an added advantage of reducing the cost on different chemicals which are aimed at controlling specific pathogens. In addition to this they can as well be included as one of the several practices of Integrated Disease Management, thus helping in efficient control of the disease.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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