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# Effect of foliar application of different elicitors on occurrence of pest and diseases during chia (*Salvia hispanica* L.) cultivation

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## Abstract

**Chia is an important medicinal crop with lot of nutritional value in seeds and there is lot of demand to chia seeds grown without any chemical application. In this context, present study was concentrated on foliar application of different elicitors at 25 and 50 days after sowing as effective plant protectants by inducing plant defense response. Both black and white chia plants sprayed with chitosan at 200 ppm were un-affected by any pest and diseases. The plants sprayed with other elicitors like 100 ppm of salicylic acid, methyl jasmonic acid, potassium silicate, 200 ppm of boric acid, humic acid and 5000 ppm of dry yeast and PGPR are less prone to pest and disease attack. Whereas, the maximum pest infestation and disease incidence were noticed in plants sprayed with gibberellic acid (pest - 2.38 and 1.93%; disease - 0.89% in both) and in control (pest - 1.34 and 1.04%; disease - 1.34 and 0.74%) in black and white chia respectively.**

**Key words:** ***Chia, Chitosan, Elicitor, $GA_3$, Methyl Jasmonic Acid, Salicylic Acid***

## Introduction

Elicitors are the compounds which triggers/stimulates defense response in plants. Whenever elicitors are sprayed on the plant, the plant assumes that an external invader has come. Hence, plants want to ensure that it strengthened its cell wall. As a result a "signal" is created due to a metabolic stimulus and possible movement intra-cellularly as well as inter-cellularly and systemically caused by interaction with the elicitor (Baenas *et al*., 2014). Plants sprayed with elicitors respond quickly through plant membrane receptors by which it induces local resistance and subsequently generate plant molecular response (Aziz *et al.,* 2003). Many authors have described that plants activates defence related reaction in response to elicitor action through induction of proteins and enzymes which are involved in oxidative stress protection and also cause rapid death of cells by hypertensive response at the immediate point of exposure to external invaders (Garcia-Brugger *et al*., 2006). During these processes, reactive oxygen species and reactive nitrogen species are produced and they alerts the plasma membrane of a cell and inflate the ion fluxes i.e, $Cl^-$, $K^+$ efflux and $Ca^{2+}$ influx which acts as structural defence barriers by activation and *de novo* biosynthesis of transcription factors which regulate and induce the genes responsible for secondary metabolite production (Smetanska, 2005, Zhao *et al*., 2005 and Ferrari, 2010). These secondary metabolites are the key components which deter, repel or fight against external invaders. Chia is a new crop to India introduced recently by Central Food Technological Research Institute, (CFTRI) Mysuru. There were no severe pests and diseases recorded in India as for now, but, as the cultivation progresses new pest and diseases may emerge and therefore, use of elicitation technique may be a promising strategy to avoid use of plant protection chemicals since, the people are demanding for residue free food.

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## Materials and Methods

The present study was conducted for both black and white chia in a randomized complete block design with eleven treatments and 3 replications during *rabi* season of 2018-19 and 2019-20 at Department of Plantation, Spices, Medicinal and Aromatic Crops, College of Horticulture, University of Horticultural Sciences campus, Gandhi Krishi Vignana Kendra, Bengaluru.

The elicitors were applied twice at 25 and 50 days after sowing as foliar spray. The various treatments includes $T_1$: Chitosan 200 ppm, $T_2$: Salicylic acid 100 ppm, $T_3$: Dry yeast 5000 ppm, $T_4$: Methyl jasmonic acid 100 ppm, $T_5$: Potassium silicate 100 ppm, $T_6$: Gibberellic acid 100 ppm, $T_7$: Kinetin 100 ppm, $T_8$: Humic acid 200 ppm, $T_9$: Boric acid 200 ppm, $T_{10:}$ Plant growth promoting rhizobacteria (PGPR) 5000 ppm and $T_{11}$: Control. The chitosan was added to distilled water three days prior to application for complete solubilization. Similarly dry yeast was added to distilled water five days prior to application for the multiplication of beneficial microbes. The PGPR consortia contain *Azospirllum*, P-solubilizer (*Bacillus megaterium*) and *Pseudomonas fluorescens* in the proportion of 1:1:1. The major pests infested the chia were termites (Fig. 1) and ants (Fig. 2). While, the major diseases noticed during the study was begomo virus infection (Fig. 3). The number of plants infested by diseases and pests during the entire cropping period were recorded and percentage infestation was worked out for both the years as well as for pooled data. The data analysis was done by using WASP 2.0 programme developed by Ashok and Pranjali of ICAR Research Complex for Goa (https://ccari.res.in/wasp2.0/index.php) and data was subjected to arcsine transformation.

## Results and Discussion

In black chia, the plants which were sprayed with $GA_3$ at 100 ppm (3.27%) were much affected and no plants were infested by any pest in plants sprayed with 200 ppm of chitosan ($T_1$) and 100 ppm of methyl jasmonic acid ($T_4$) during first year (Table 1). In the second year, more pest infestation (1.79%) was seen in plants sprayed with kinetin at 100 ppm ($T_5$) which was *on par* with $GA_3$ (100 ppm), control and boric acid (200 ppm) treatments while, the infestation was nil in case of $T_1$, $T_2$ (Salicylic acid at 100 ppm) and $T_4$. Similarly, in case of pooled mean data, the infestation was seen maximum (2.38%) in $GA_3$ treatment and nil in case of chitosan and methyl jasmonic acid treatment. With respect to disease incidence, the maximum disease occurrence (1.49, 1.19 and 1.34% respectively) was recorded in the control and no infection was seen in $T_1$ and $T_2$ in 2018-19, 2019-20 ($T_4$ and $T_5$ also) and in pooled mean data. The plants in control maintained statistical parity with plants sprayed with gibberellic acid at 100 ppm (1.19%) for maximum disease incidence in second season (Table 1).

Whereas in white chia also, the plants treated with $GA_3$ at 100 ppm ($T_6$) recorded maximum pest infestation of 2.38, 1.49 and 1.93 per cent during first, second (*at par* with control) and in average data over two years respectively (Table 2). Whereas, the plants sprayed with of chitosan (200 ppm) and salicylic acid (100 ppm) recorded zero pest infestation in all the cropping periods. Similarly, the plant sprayed with dry yeast (5000 ppm) and methyl jasmonic acid (100 ppm) also recorded zero pest infestation during 2018-19 and 2019-20 respectively. The maximum disease incidence (0.89%) was also recorded in plants treated with 100 ppm of $GA_3$ during all the cropping periods (Table 2). Whereas, $T_6$ was *on par* with $T_{11}$ and the disease incidence level in control was same as $T_6$ during second year *i.e*. 0.89 per cent. However, disease incidence was nil in plants elicited with chitosan (200 ppm), dry yeast (5000 ppm), methyl jasmonic acid (100 ppm) and kinetin (100 ppm) throughout the study. Zero disease incidences were also observed in plants treated with salicylic acid (100 ppm) and PGPR (5000 ppm) during 2018-19 and in potassium silicate (100 ppm) and boric acid (200 ppm) during 2019-20. In both black and white chia the pest occurrence and disease incidence was nil in plants sprayed with 200 ppm of chitosan in both the seasons. While, the incidence was very less in case of plants sprayed other elicitors except $GA_3$ and control treatments. Gibberellic acid spray caused the apical dominance (Brian, 1959), further resulted in production of less number of tertiary spikes which caused the sparse canopy which frees up the movement of pests. Whereas the disease incidence was maximum in control since there was no stimuli for the induction of defence response.

**Table 1: Pest infestation and disease incidence recorded during the crop period of black chia**

| Treatments | Pest infestation (%) | | | Disease incidence (%) | | |
|---|---|---|---|---|---|---|
| | 2018-19 | 2019-20 | Pooled | 2018-19 | 2019-20 | Pooled |
| $T_1$:Chitosan 200 ppm | $0.00^{c}$ (0.00) | $0.00^{e}$ (0.00) | $0.00^{c}$ (0.00) | $0.00^{b}$ (0.00) | $0.00^{b}$ (0.00) | $0.00^{c}$ (0.00) |
| $T_2$: Salicylic acid 100 ppm | $0.30^{bc}$ (3.13) | $0.00^{e}$ (0.00) | $0.15^{c}$ (2.21) | $0.00^{b}$ (0.00) | $0.00^{b}$ (0.00) | $0.00^{c}$ (0.00) |
| $T_3$: Dry yeast 5000 ppm | $0.30^{bc}$ (3.13) | $0.30^{de}$ (3.13) | $0.30^{c}$ (3.13) | $0.00^{b}$ (0.00) | $0.30^{b}$ (3.13) | $0.15^{c}$ (2.21) |
| $T_4$: Methyl Jasmonic acid 100 ppm | $0.00^{c}$ (0.00) | $0.00^{e}$ (0.00) | $0.00^{c}$ (0.00) | $0.30^{b}$ (3.13) | $0.00^{b}$ (0.00) | $0.15^{c}$ (2.21) |
| $T_5$: Potassium silicate 100 ppm | $0.30^{bc}$ (3.13) | $0.60^{cde}$ (4.42) | $0.45^{c}$ (3.83) | $0.60^{b}$ (4.42) | $0.00^{b}$ (0.00) | $0.30^{c}$ (3.13) |
| $T_6$: Gibberellic acid 100 ppm | $3.27^{a}$ (10.42) | $1.49^{ab}$ (7.01) | $2.38^{a}$ (8.88) | $0.60^{b}$ (4.42) | $1.19^{a}$ (6.26) | $0.89^{ab}$ (5.42) |
| $T_7$: Kinetin 100 ppm | $0.89^{bc}$ (5.42) | $1.79^{a}$ (7.68) | $1.34^{b}$ (6.65) | $0.60^{b}$ (4.42) | $0.30^{b}$ (3.13) | $0.45^{bc}$ (3.83) |
| $T_8$: Humic acid 200 ppm | $0.89^{bc}$ (5.42) | $0.60^{cde}$ (4.42) | $0.74^{b}$ (4.95) | $0.60^{b}$ (4.42) | $0.30^{b}$ (3.13) | $0.45^{bc}$ (3.83) |
| $T_9$: Boric acid 200 ppm | $0.60^{bc}$ (4.42) | $0.89^{abcd}$ (5.42) | $0.74^{b}$ (4.95) | $0.60^{b}$ (4.42) | $0.30^{b}$ (3.13) | $0.45^{bc}$ (3.83) |
| $T_{10}$: PGPR 5000 ppm | $0.89^{bc}$ (5.42) | $0.60^{cde}$ (4.42) | $0.74^{b}$ (4.95) | $0.30^{b}$ (3.13) | $0.30^{b}$ (3.13) | $0.30^{c}$ (3.13) |
| $T_{11}$: Control | $1.49^{b}$ (7.01) | $1.19^{abc}$ (6.26) | $1.34^{b}$ (6.65) | $1.49^{a}$ (7.01) | $1.19^{a}$ (6.26) | $1.34^{a}$ (6.65) |
| **SE m±** | 0.36 | 0.24 | 0.24 | 0.26 | 0.23 | 0.17 |
| **CD at 5%** | 1.07 | 0.71 | 0.69 | 0.75 | 0.67 | 0.48 |
| **CV** | 77.07 | 61.23 | 80.04 | 96.17 | 111.79 | 102.62 |

**Note: Figures in parentheses are arcsine transformed values**
**Means followed by same letter in the column do not differ significantly at 5 per cent level by DMRT**

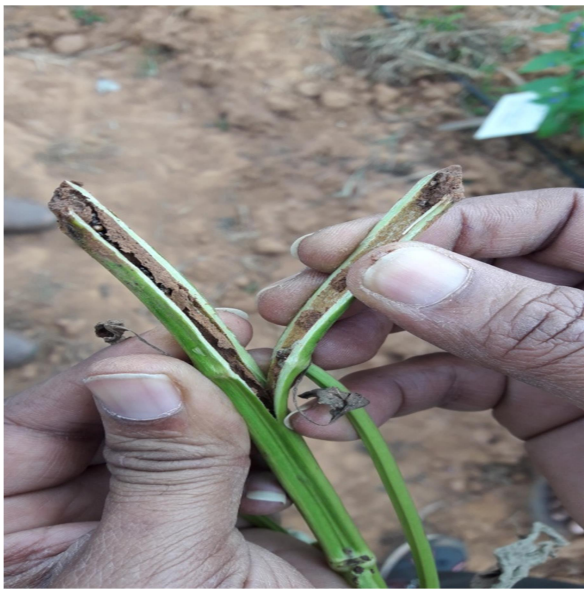

**Figure 1: Termite's damage**

Elicitors induce the plant immune response by activation of signal cascade (Patel *et al*., 2020). The secondary metabolites produced in plants due to various signaling molecules in response to elicitor application stimulate the defense action (Dzhavakhiya and Shcherbakova 2007). Hence, elicitation is one promising approach to enhance the resistance of plants against various diseases and insect attack. Chitosan activates the various enzymes to detoxify reactive oxygen species in which hydrogen peroxide and nitric oxide are also said to be involved in signalling of chitosan which helped in boosting the plant immunity (Hadwiger, 2013). It is also suspected to interact with chromatin and directly affect the gene expression (Hadwiger, 2015). Salicylic acid is a stress messenger induces hypersensitive responses and helps the plant to escape from external invaders (Rodas-Junco *et al*., 2020). The phenylalanine

**Table 2: Pest infestation and disease incidence recorded during white chia growing period**

| Treatments | Pest infestation (%) | | | Disease incidence (%) | | |
|---|---|---|---|---|---|---|
| | 2018-19 | 2019-20 | Pooled | 2018-19 | 2019-20 | Pooled |
| $T_1$:Chitosan 200 ppm | 0.00$^{c}$ (0.00) | 0.00$^{c}$ (0.00) | 0.00$^{c}$ (0.00) | 0.00$^{b}$ (0.00) | 0.00$^{b}$ (0.00) | 0.00$^{c}$ (0.00) |
| $T_2$: Salicylic acid 100 ppm | 0.00$^{c}$ (0.00) | 0.00$^{c}$ (0.00) | 0.00$^{c}$ (0.00) | 0.00$^{b}$ (0.00) | 0.30$^{b}$ (3.13) | 0.15$^{c}$ (2.21) |
| $T_3$: Dry yeast 5000 ppm | 0.00$^{c}$ (0.00) | 0.30$^{bc}$ (3.13) | 0.15$^{c}$ (2.21) | 0.00$^{b}$ (0.00) | 0.00$^{b}$ (0.00) | 0.00$^{c}$ (0.00) |
| $T_4$: Methyl Jasmonic acid 100 ppm | 0.30$^{c}$ (3.13) | 0.00$^{c}$ (0.00) | 0.15$^{c}$ (2.21) | 0.00$^{b}$ (0.00) | 0.00$^{b}$ (0.00) | 0.00$^{c}$ (0.00) |
| $T_5$: Potassium silicate 100 ppm | 0.30$^{c}$ (3.13) | 0.60$^{bc}$ (4.42) | 0.45$^{c}$ (3.83) | 0.30$^{b}$ (3.13) | 0.00$^{b}$ (0.00) | 0.15$^{c}$ (2.21) |
| $T_6$: Gibberellic acid 100 ppm | 2.38$^{a}$ (8.88) | 1.49$^{a}$ (7.01) | 1.93$^{a}$ (8.00) | 0.89$^{a}$ (5.41) | 0.89$^{a}$ (5.41) | 0.89$^{a}$ (5.41) |
| $T_7$: Kinetin 100 ppm | 0.60$^{bc}$ (4.42) | 0.60$^{bc}$ (4.42) | 0.60$^{bc}$ (4.42) | 0.00$^{b}$ (0.00) | 0.00$^{b}$ (0.00) | 0.00$^{c}$ (0.00) |
| $T_8$: Humic acid 200 ppm | 0.30$^{c}$ (3.13) | 0.60$^{bc}$ (4.42) | 0.45$^{c}$ (3.83) | 0.30$^{b}$ (3.13) | 0.30$^{b}$ (3.13) | 0.30$^{c}$ (3.13) |
| $T_9$: Boric acid 200 ppm | 0.60$^{bc}$ (4.42) | 0.30$^{bc}$ (3.13) | 0.45$^{c}$ (3.83) | 0.30$^{b}$ (3.13) | 0.00$^{b}$ (0.00) | 0.15$^{c}$ (2.21) |
| $T_{10}$: PGPR 5000 ppm | 0.60$^{bc}$ (4.42) | 0.30$^{bc}$ (3.13) | 0.45$^{c}$ (3.83) | 0.00$^{b}$ (0.00) | 0.30$^{b}$ (3.13) | 0.15$^{c}$ (2.21) |
| $T_{11}$: Control | 1.19$^{b}$ (6.26) | 0.89$^{ab}$ (5.41) | 1.04$^{b}$ (5.86) | 0.60$^{ab}$ (4.42) | 0.89$^{a}$ (5.41) | 0.74$^{b}$ (4.93) |
| **SE m ±** | 0.25 | 0.24 | 0.17 | 0.19 | 0.16 | 0.12 |
| **CD** | 0.73 | 0.72 | 0.50 | 0.55 | 0.46 | 0.34 |
| **CV** | 75.41 | 91.30 | 83.13 | 148.90 | 110.55 | 125.71 |

**Note: Figures in parentheses are arcsine transformed values**
**Means followed by same letter in the column do not differ significantly at 5 per cent level by DMRT**

**Figure 2: Ant's damage**

mmonia lyase is known to be involved in defense reaction of a plant due to *de novo* transcription of genes which can be activated by methyl jasmonate (Chen *et al*., 2017). Potassium silicate provides strength to plant and also resist the external invaders and minimizes the pest and disease attack. Some of the studies related to effect of elicitors on plant defence are listed here. Chitosan in tomato (Cho *et al*., 2011 and Tavallali *et al*., 2008) and in

**Figure 3: Begomo virus affected plant.**

paddy, radish, tobacco, groundnut and Pea (Maksimov *et al*., 2003). Salicylic acid in tomato (Esmailzadeh and Soleimani, 2008; Garcia-Magallon *et al*., 2002; Peng *et al*., 2004; Tavallali *et al*., 2008), pea (Katoch, 2005) and Bhendi (Vimala and Suriachandraselvan, 2009). Methyl jasmonic acid in *Brassica* (Xu *et al.,* 2018), Brinjal (Mandal, 2010) and cabbage (Perez-Balibrea *et al*., 2011). Humic acid in soyabean (Abdel-Monaim *et al*., 2011). Potassium silicate in wheat (Mehdi *et al*., 2007), strawberry (Kanto *et al.,* 2006) and in chilli etc. (Dogramaci *et al.,* 2013).

## Conclusion

Elicitation is an important strategy to induce resistance in chia which reduces the use of plant protection chemicals by reducing the pest and pathogen populations. In both black chia, foliar spray with chitosan at 200 ppm showed 100 per cent resistance to any pest and diseases and the plants sprayed with 100 ppm of salicylic acid, methyl jasmonic acid and potassium silicate was less to prone pest and disease attack. However for pooled mean, maximum pest infestation was recorded in plants sprayed with 100 ppm of $GA_3$ while the maximum disease occurrence was seen in untreated plants. In white chia, the plants sprayed with 200 ppm of chitosan and 100 ppm of salicylic acid showed 100 per cent resistance to pest infestation in all cropping periods while, the plats sprayed with $GA_3$ recorded maximum pest infestation. Whereas, plants sprayed with 200 ppm of chitosan, 100 ppm of methyl jasmonic acid, kinetin and 5000 ppm of dry yeast recorded zero disease incidence while, the plants treated with $GA_3$ (100 ppm) and in control recorded maximum disease occurrence in all the cropping periods.

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