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# Evaluation of Biotic Inducers on Sesame Growth and Defense Enzyme Activity

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**ABSTRACT: This study aimed to evaluate the efficacy of biotic inducers for assessing growth promotion in sesame, as well as chlorophyll, biochemical changes (phenol) and antioxidant enzyme activity. The experiments were developed with the biotic inducers Salicylic acid (SA) (50ppm, 100ppm, 150ppm), Methyl jasmonate (MeJA) (50ppm, 100ppm, 150ppm) and Beta amino butyric acid (BABA) (50ppm, 100ppm, 150ppm) as well as with the PGPR *Bacillus subtilis* and Methyl dematon 25EC as insecticidal control. Leaf samples were collected 30$^{th}$ day after the treatments to determine antioxidant assays. Concerning biotic inducers, primed sesame seedling of SA 50ppm showed maximum germination percentage (86%), and SA 150ppm promotes maximum shoot length (87.63cm/plant) and maximum number of capsules (54 capsules /plant) were observed in the primed seedlings of MeJA 150ppm. The defense enzyme activity was found to be higher in SA 50ppm primed seedlings followed by SA 100ppm. These results showed that SA pre-seed treatment and exogenous application at 30$^{th}$, 45$^{th}$ & 60$^{th}$ DAS resulted in higher biomass production of sesame plants and added significant value by increasing defense enzyme activity (PO, PPO, PAL and SOD).**

**Keywords:** Sesame, Salicylic acid, Jasmonic acid, Beta amino butyric acid.

## INTRODUCTION

Sesame (*Sesamum indicum* L.) is an ancient oil seed crop which is originated from Africa and grown in many parts of the world. Because of the superior qualities of the seed, oil, and meal, it is referred to as the "Queen of oil seeds". It also has the highest nutritional energy (6355 kcal/kg) and oil content (46–64%). Despite its economic and nutritional importance and high concentration of lipid-soluble lignans, mainly sesamol, sesamin, and sesamolin, which protect it from oxidative rancidity and lengthens its shelf life, sesame is regarded as a "orphan crop" because science has paid it very little attention (Rizki *et al*., 2015). Global use of sesame oil is predicted to reach 100 MMT by 2030 (Troncoso-Ponce *et al*., 2011). However, the exposure of the crops to multiple biotic and abiotic stresses is largely responsible for the current decline in sesame farming. Low yields from a lack of production techniques often place a limit on the amount of sesame that can be grown.

The plant defense inducers/biotic inducers used for this study are Salicylic acid (SA), Methyl jasmonate (MeJA), Beta amino butyric acid (BABA). Salicylic acid, a naturally occurring phenolic molecule found in many plants, is crucial for the signal transduction pathway and has a role in both local and systemic pathogen resistance (Delaney *et al*., 1995; Maleck *et al*., 2000). Nemeth *et al*. (2002) showed that foliar SA treatments may be responsible for the stimulating effect of plant growth and tomato yield. MeJA are a class of oxylipins that are produced naturally in a variety of

higher plants by the lipoxygenase-dependent oxidation of fatty acids (Creelman & Mullet 1997). Under the short soybean season field conditions. Mabood *et al.* (2006) showed that treatment with MeJA at 50 µM promoted growth, dry matter accumulation, and grain production. According to Jisha *et al*. (2016), BABA seed-priming promoted seedling growth in rice under both unstressed and stressed conditions. Many reports shows that the plant defense inducers induce resistance against many pathogens/biotic stress by activating some of the defense enzymes like Peroxidase (PO), polyphenol oxidase (PPO), superoxide dismutase (SOD), Phenylalanine ammonia lyase (PAL) etc. But, few reports were attempted in the growth promotion activities using biotic inducers. In this present study, we investigate the growth promotion, yield attributes of sesame plants and the activation of defense enzymes under glasshouse were studied.

## MATERIALS AND METHODS

### Source of seed material

The study was carried out using a sesame seed variety (CO 1). The seed was purchased from the Tamil Nadu Agricultural University (TNAU), Department of Oilseeds, Coimbatore. The seeds were cleaned, dried, and stored in airtight polythene bags with a moisture content of 6%.

### Source of biotic inducers

**Blotter paper method.** To study the growth promotion effect of biotic inducers. Sesame seeds (0.5g) were treated with required concentrations of Salicylic acid, Methyl jasmonate, beta amino butyric acid each at 50ppm, 100ppm, 150ppm respectively and the untreated as control. Further the seeds were subjected to hot water treatment for 55°C for 10min. The treated seeds were blot dried and placed in a petriplates. Incubate the plates at 25± 2°C for 3 days to analyze seed germination.

**Roll towel method.** Seed germination ability biotic inducers (Salicylic acid, Methyl jasmonate, beta amino butyric acid) were tested using roll paper towel method at different ppm concentrations *i.e*., 50ppm, 100ppm, 150ppm. Seeds were treated with required concentrations of biotic inducers and the seeds are subjected to hot water treatment and then 25 seeds were placed in germination paper and incubated at 25± 2°C for 10 days. Untreated seeds were used as control. At tenth day, germination percentage, root length, and shoot length, vigour index were measured for each treatment. The vigour index (VI) of sesame seedlings was calculated by using the below mentioned formula described by Agrawal and Agrawal (2013).

VI=Germination% × Mean total length of seedling (root length + shoot length)

**Study of efficacy of biotic inducers under glasshouse condition.** Seeds are primed with biotic inducers each at different concentrations (50ppm, 100ppm & 150ppm) for 30 mins. Further, primed seeds were subjected to soaking for 15 minutes which allowing the biotic inducers to absorb into the seeds and then the seeds were air dried (Fig. 1). The experiment were conducted at PL-480 glasshouse, TNAU, Coimbatore. Soaked sesame seeds were planted in pots containing red soil, sand and farm yard manure in the ratio of 2:1:1. The soil was autoclaved for two hours to sterilize it prior to seeding. Three uniform and healthy seedlings were kept in each pot after being thinned five days after germination. The Foliar application of SA (ppm of 50,100,150), MeJA (ppm of 50,100,150), BABA (ppm of 50,100,150), and combined (50ppm of SA+ MeJA+BABA) on 30$^{th}$, 45$^{th}$ & 60$^{th}$ days after planting. Methyl dematon 25 EC and *Bacillus subtilis* Bbv 57 were used as treatments as part of farmers' practice. Respective control was also maintained. The experiment was a completely randomized design (CRD) with 13 treatments with 3 replications. The treatment details are as follows (Table 1).

| S. No. | Biotic inducers name | Molecular weight (g/mol) | Nature | Purchased from | CAS number |
|---|---|---|---|---|---|
| 1. | Salicylic acid | 138.12 | Soluble in water | Hi-Media | 69-72-7 |
| 2. | Methyl jasmonate | 224.30 | Soluble in ethanol | Sigma–Aldrich | 39924-52-2 |
| 3. | Beta amino butyric acid | 103.12 | Soluble in water | Hi-Media | 56-12-2 |

**Growth parameters assessment under glasshouse condition.** After spraying under glasshouse conditions, Plant growth parameters including germination percentage, shoot length, number of branches, no. of capsules, girth and width were measured and calculated.

### Biochemical analysis

**Chlorophyll content.** Using a chlorophyll concentration meter, the amount of chlorophyll was determined. The fourth leaf from the top of the plant was chosen to measure chlorophyll. The chlorophyll content (mg/m$^2$) of the leaf was measured by simply placing it between the sensors of the chlorophyll concentration meter (SPAD 502 Plus Chlorophyll meter).

**Total phenol content.** The Folin Ciocalteu assay, with minor modifications, was used to gauge the amount of phenolic chemicals present in the plant extracts. In a summary, the extract was diluted to a concentration of 1 mg/ml, and aliquots of 100µl of a standard solution of gallic acid (20, 40, 60, 80, and 100 mg/l) were combined with 500µl of Folin Ciocalteu reagent (previously diluted 10-fold with distilled water), 400µl of $Na_2$ $CO_3$ and 400µl of (7 %). The absorbance at 760

nm was measured using a spectrophotometer against a blank sample following 40 min of incubation at room temperature (23 2°C). Using a calibration curve for gallic acid (R2 = 0.998), the total phenolic content was determined. Gallic acid equivalent per gram of dry weight of extract (mg of GAE/g of extract) was used to express the results. Each sample was examined three times (Abdelhakim *et al*., 2016).


**Fig. 1.** Priming of sesame seeds with biotic inducers (a) Pre-soaking of salicylic acid @ 50 ppm, 100ppm, 150ppm (b) Pre-soaking of Methyl jasmonate @ 50 ppm, 100ppm, 150ppm (c) Pre-soaking of beta amino butyric acid @ 50 ppm, 100ppm, 150ppm).

**Table 1: Treatments and their concentrations used for this study.**

| Sr. No. | Treatment details |
|---|---|
| T1 | ST+FS of Salicylic acid (SA) @ 50 ppm @ 30$^{th}$, 45$^{th}$,60$^{th}$ DAS |
| T2 | ST+FS of Salicylic acid (SA) @ 100 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T3 | ST+FS of Salicylic acid (SA) @ 150 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T4 | ST+FS of Methyl jasmonate (MeJA) @ 50 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T5 | ST+FS of Methyl jasmonate (MeJA) @ 100 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T6 | ST+FS of Methyl jasmonate (MeJA) @ 150 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T7 | ST+FS of Beta amino butyric acid (BABA) @ 50 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T8 | ST+FS of Beta amino butyric acid (BABA) @ 100 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T9 | ST+FS of Beta amino butyric acid (BABA) @ 150 ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T10 | ST+FS of Combination (SA+JA+BABA) each @ 50ppm @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T11 | ST+FS of Methyl dematon 25EC @ 10ml/kg @ 30$^{th}$, 45$^{th}$, 60$^{th}$ DAS |
| T12 | ST+FS of *Bacillus subtilis* Bbv57@ 10ml/kg @ 45$^{th}$, 60$^{th}$ DAS |
| T13 | Control |

**Assay for defense enzymes activities**

**Enzyme extraction.** Extract 1g of fresh plant tissue in 3ml of 0.1 M phosphate buffer pH 7.0 by grinding in a pre-cooled pestle and mortar. Centrifuge the homogenate for 15 minutes at 18,000 rpm at 5°C. Within two to four hours, use the supernatant as an enzyme source. Until the assay is completed, keep on ice.

**Peroxidase assay (PO).** Peroxidase activity was analyzed as described by (Hammerschmidt *et al*., 1982). The cuvette was filled with 1.5ml of 0.05 M pyrogallol and 0.1ml of enzyme extract. 1% hydrogen peroxide was added to 0.5 ml to start the reaction. After one second of incubation at room temperature, the change in absorbance was measured at 420 nm every 30 seconds for three minutes. Change in absorbance/min/g of fresh tissue was used to express the results.

**Polyphenol oxidase (PPO).** Polyphenol oxidase activity was analyzed as described by (Mayer *et al*., 1966). The reaction mixture consisted of 1.5ml of 0.1 M sodium phosphate buffer pH 6.5 with 0.1ml of enzyme extract. To start the reaction, 0.2ml of 0.01 M catechol was added. The results were expressed as change in absorbance /min/g of fresh tissue and the absorbance change was measured at 495 nm.

**Phenylalanine ammonia lyase (PAL).** The method outlined by (Ngadze *et al*., 2012) was used to measure phenylalanine ammonia lyase (PAL). 0.25g of the seedlings from the homogenized tissue were added to 5 ml of buffer containing 50 mM of 2-mercaptoethanol and 5% (w/v) polyvinylpyrrolidone. The homogenate was centrifuged at 13000 rpm for 4 minutes at 4°C after being filtered through four layers of cheesecloth. The sample was incubated at 30°C for an hour after 1 ml of the supernatant was added to a solution containing 2 ml of 0.05M borate buffer (pH 8.8) and 1 ml of 0.02M L phenylalanine. 0.2 ml of 6M trichloroacetic acid was added to the test tube to terminate the reaction. For spectrophotometer readings at 290 nm absorbance, this solution was divided into three sections.

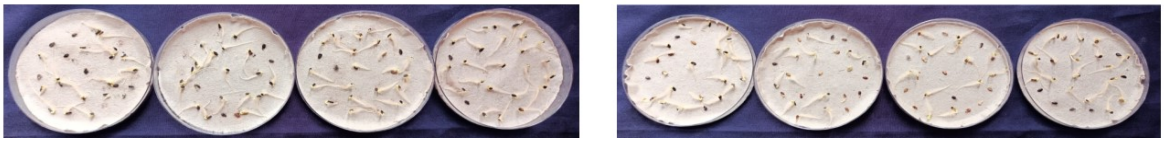
**Super oxide dismutase (SOD).** SOD activity was measured as its capacity to prevent the photochemical reduction by NBT using the supernatant as an enzyme source (Giannopolitis and Ries 1977). The assay mixture (3 ml) contains 100 ml of the enzyme extract, 50 mM sodium phosphate buffer (pH 7.8), 13 mM methionine, 75 mM NBT, 2 mM riboflavin, 0.1 mM EDTA, and the riboflavin was added at the end. Tubes were shaken and placed under a 40-W fluorescent lamp at 25°C. The reaction was initiated and terminated by turning the light on and off respectively. In parallel with the sample tubes for the blank, the absorbance at 560 nm was measured against identical, non-illuminated samples. The percentage inhibition of NBT photo-

reduction was calculated by subtracting each extract from the blank, dividing mathematical differences by the blank, and multiplying the result by 100.The SOD activity was expressed in SOD units mg/tissue (50% NBT inhibition=1unit).

**Statistical analysis.** The results of germination percentage and plant growth and yield parameters of sesame under glasshouse conditions were subjected to analysis of variance (ANOVA) using the SPSS programme, and the treatment means were compared using the Duncan's multiple range test (DMRT).

## RESULT AND DISCUSSION

**Growth promotion assay.** Growth promotion study of blotter paper assay (Fig. 2, Table 2) showed that SA 50ppm enhanced the germination percentage of sesame seedlings up to 85%, followed by SA 100ppm (83%), MeJA 150ppm (83%) and BABA 50ppm (82%) when compared to control (68%) and similar report has been accounted that, SA 50ppm enhanced the vigour index of sesame seedlings up to 1405.05, followed by BABA 50ppm (1291.50), MeJA 150ppm (1272.00) and SA 100ppm (1162.40) when compared to control (771.28) through roll towel method (Fig. 3, Table 3).



(a) (b)

**Fig. 2.** Blotter paper assay (a) Salicylic acid 50 ppm (b) Control).

**Table 2: Effect of growth peomotion of primed sesame seedlings by Blotter paper assay.**

| Sr. No. | Treatments | *Germination percentage (%) | | |
|---|---|---|---|---|
| | | Salicylic acid (SA) | Methyl jasmonate (MeJA) | Beta amino butyric acid (BABA) |
| 1. | 50 ppm | $85.2^{a}$ (67.34) | $76.3^{b}$ (60.71) | $82.6^{a}$ (64.99) |
| 2. | 100 ppm | $83.4^{ab}$ (65.75) | $74.5^{b}$ (59.37) | $77.2^{a}$ (61.39) |
| 3. | 150 ppm | $77.7^{bc}$ (61.39) | $83.3^{a}$ (65.75) | $81.6^{a}$ (64.23) |
| 4. | Control | $73.4^{c}$ (58.72) | $69.5^{b}$ (56.18) | $68.4^{b}$ (55.57) |
| | **CD (p < 0.05)** | 5.53 | 4.90 | 5.12 |
| | **SE(d)** | 2.39 | 2.12 | 2.22 |

*Values are the means of three replications. Means in a column followed by superscript letters are not significantly different according to Duncan's multiple range test at p < 0.05. Values in parentheses are arcsine transformed values.

**Table 3: Effect of growth peomotion of primed sesame seedlings by Roll towel assay.**

| Treatments | *Shoot length | *Root length | *Germination % | Vigour index |
|---|---|---|---|---|
| SA 50 ppm | $7.95^{a}$ (16.32) | $8.58^{a}$ (17.03) | $85.00^{a}$ (67.19) | 1405.05 |
| SA 100 ppm | $7.91^{ab}$ (16.26) | $7.87^{f}$ (16.95) | $80.00^{a}$ (64.53) | 1262.40 |
| SA 150 ppm | $7.15^{h}$ (16.19) | $7.30^{g}$ (16.79) | $75.00^{a}$ (61.28) | 1083.75 |
| MeJA 50 ppm | $7.57^{f}$ (16.18) | $8.50^{ab}$ (16.48) | $74.00^{b}$ (37.09) | 1189.18 |
| MeJA 100 ppm | $7.64^{e}$ (16.04) | $8.35^{c}$ (16.28) | $72.00^{c}$ (0.28) | 1151.28 |
| MeJA 150 ppm | $7.85^{c}$ (15.95) | $8.05^{d}$ (16.19) | $80.00^{c}$ (0.28) | 1272.00 |
| BABA 50 ppm | $7.77^{de}$ (15.50) | $7.98^{e}$ (15.95) | $82.00^{c}$ (0.28) | 1291.50 |
| BABA 100 ppm | $7.78^{d}$ (14.85) | $7.14^{h}$ (15.67) | $74.00^{c}$ (0.28) | 1104.08 |
| BABA 150 ppm | $7.37^{g}$ (14.61) | $7.05^{i}$ (14.61) | $80.00^{c}$ (0.28) | 1153.60 |
| Control | $6.37^{i}$ (14.11) | $6.07^{j}$ (14.26) | $62.00^{c}$ (0.28) | 771.28 |
| **CD (p < 0.05)** | 0.49 | 0.60 | 16.70 | |
| **SE(d)** | 0.05 | 0.08 | 0.09 | |

*Values are the means of three replications. Means in a column followed by superscript letters are not significantly different according to Duncan's multiple range test at p < 0.05. Values in parentheses are arcsine transformed values.

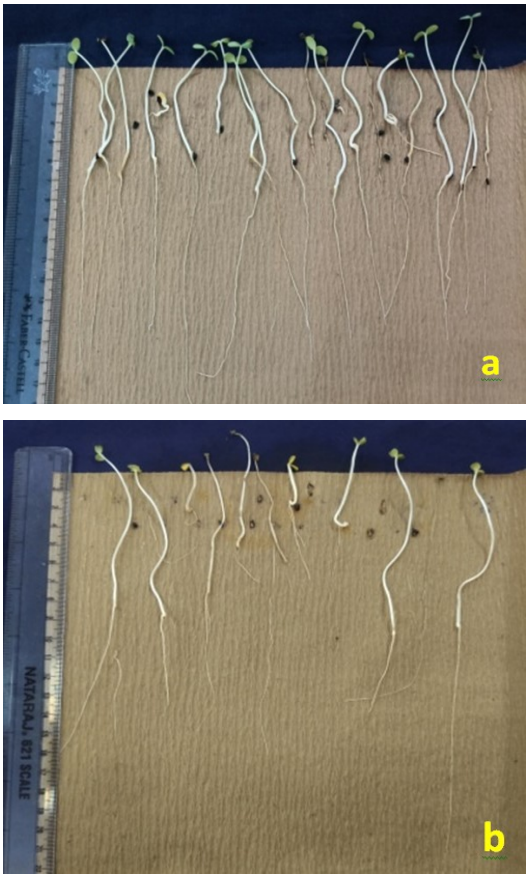



**Fig. 3.** Roll paper assay (a) Salicylic acid 50 ppm (b) Control.

The treatment SA 50ppm also recorded high germination percentage (85%) with increase root length (8.58 cm) and shoot length (7.95 cm) and the shoot length of (6.37 cm), root length of (6.07 cm) with low germination percentage (62%) has been recorded in the control. Although salicylic acid is a growth stimulator for plant germination, the dose that is utilized for priming must be restricted, and salicylic acid at large concentrations not only doesn't improve germination conditions, but also has negative impacts on them (Salehi *et al*., 2015).

**Effect of biotic inducers on plant growth under glasshouse conditions.** The measured growth attributes are shoot length, no. of branches and grith length (Table 4). In this study, Primed & foliar sprayed SA 150ppm seedlings showed maximum Shoot length (87.63 cm/plant), followed by SA 100ppm (83.93 cm/plant), *Bacillus subtilis* Bbv57 (73.7cm/plant), Methyl dematon 25 EC (66.3 cm/plant) compared with control (64cm/plant) on 90$^{th}$ day after planting (Fig. 4). Similar results showed that, In *C. officinalis*, exogenous SA (1 and 2 mM) treatment increased shoot, root, and total plant dry weight while promoted early blooming and a high number of floral buds per plant (Bayat *et al*., 2012). Other treatments were also had significant effect on shoot length of sesame plants. Same number of branches were observed in all treatments. The maximum grith length was observed in the treatment of SA 100ppm (3.87cm/plant) compared to control (3.14cm/plant).

**Table 4: Efficacy of growth and yield parameters of biotic inducers under glass house conditions.**

| Sr. No. | Treatments | *Germination % | *Shoot length (cm) | | | *Grith (cm) | *No. of branches/plant | *No. of capsules/plant | *Length of capsules/ plant |
|---|---|---|---|---|---|---|---|---|---|
| | | | 30th day | 60th day | 90th day | | | | |
| T1 | SA 50 ppm | 86.00$^{a}$ (68.06) | 26.00$^{ef}$ (30.64) | 67.00$^{a}$ (54.94) | 67.73$^{def}$ (55.38) | 3.50$^{bcd}$ (10.78) | 5.00 | 36.60de (37.22) | 3.10$^{cd}$ (10.13) |
| T2 | SA 100 ppm | 85.00$^{ab}$ (67.30) | 35.20$^{abc}$ (36.38) | 56.00$^{b}$ (48.45) | 83.93$^{ab}$ (66.56) | 3.87$^{a}$ (11.34) | 5.00 | 40.66cd (39.60) | 3.20$^{c}$ (10.30) |
| T3 | SA 150 ppm | 80.00$^{abc}$ (63.43) | 33.80$^{bc}$ (35.54) | 55.76$^{b}$ (48.31) | 87.63$^{a}$ (69.50) | 3.54$^{bcd}$ (10.84) | 5.00 | 42.33$^{bc}$ (40.58) | 3.33$^{bc}$ (10.51) |
| T4 | MeJA 50 ppm | 81.00$^{abcd}$ (64.18) | 35.20$^{abc}$ (36.90) | 54.96$^{b}$ (47.85) | 75.70$^{cd}$ (60.60) | 3.22$^{ef}$ (10.33) | 5.00 | 44.66$^{bc}$ (41.93) | 3.31$^{bc}$ (10.47) |
| T5 | MeJA 100 ppm | 81.00$^{abcde}$ (64.15) | 36.13$^{abc}$ (36.94) | 58.00$^{b}$ (45.60) | 77.13$^{cd}$ (61.51) | 3.66$^{abc}$ | 5.00 | 44.66$^{bc}$ (41.92) | 3.34$^{bc}$ (10.52) |
| T6 | MeJA 150 ppm | 83.00$^{abcde}$ (64.85) | 36.23$^{ab}$ (37.00) | 54.50$^{b}$ (47.58) | 74.53$^{cd}$ (59.75) | 3.37$^{de}$ (10.57) | 5.00 | 53.66$^{a}$ (47.10) | 3.21$^{c}$ (10.31) |
| T7 | BABA 50 ppm | 82.00$^{abcde}$ (64.90) | 34.86$^{abc}$ (36.18) | 55.53$^{b}$ (48.18) | 77.00$^{cd}$ (61.37) | 3.82$^{a}$ (11.27) | 5.00 | 46.33$^{b}$ (42.89) | 3.30$^{bc}$ (10.46) |
| T8 | BABA 100 ppm | 79.00$^{bcde}$ (62.76) | 33.50$^{c}$ (35.36) | 55.23$^{b}$ (48.17) | 74.33$^{cd}$ (58.35) | 3.66$^{abc}$ (11.02) | 5.00 | 42.33$^{bc}$ (40.58) | 3.64$^{a}$ (10.99) |
| T9 | BABA 150 ppm | 76.00$^{cde}$ (60.70) | 34.96$^{abc}$ (36.34) | 56.33$^{b}$ (48.64) | 77.40$^{bcd}$ (61.69) | 3.74$^{ab}$ (11.15) | 5.00 | 38.00$^{de}$ (38.05) | 3.36$^{bc}$ (10.55) |
| T10 | Combination (SA 50ppm+MeJA 50ppm+ BABA 50ppm) | 81.00$^{def}$ (64.17) | 36.80$^{a}$ (37.34) | 68.10$^{a}$ (55.64) | 81.1$^{6abc}$ (64.27) | 3.22$^{ef}$ (10.33) | 5.00 | 35.00$^{e}$ (36.27) | 3.54$^{ab}$ (10.84) |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| T11 | Methyl dematon 25 EC | 72.00$^{ef}$ (58.05) | 28.60$^{de}$ (32.32) | 47.93$^{e}$ (43.81) | 66.30$^{efg}$ (53.13) | 3.05$^{f}$ (10.05) | 5.00 | 27.66$^{f}$ (31.72) | 2.62$^{e}$ (9.31) |
| T12 | *Bacillus subtilis* Bbv57 | 77.00$^{fg}$ (61.51) | 28.93$^{ef}$ (32.53) | 53.80$^{bc}$ (47.17) | 73.70$^{de}$ (59.15) | 3.47$^{cd}$ (10.73) | 5.00 | 30.33$^{f}$ (33.41) | 2.83$^{de}$ (9.68) |
| T13 | Control | 70.00$^{g}$ (56.86) | 25.16$^{f}$ (30.10) | 48.40$^{cd}$ (44.08) | 64.00$^{g}$ (54.52) | 3.14$^{ef}$ (10.20) | 5.00 | 21.33$^{g}$ (27.49) | 2.77$^{e}$ (9.58) |
| | **CD (p < 0.05)** | 2.25 | 2.63 | 5.45 | 6.72 | 0.246 | | 4.08 | 0.27 |
| | **SE(d)** | 0.07 | 0.08 | 0.04 | 0.07 | 0.02 | | 0.03 | 0.07 |

*Values are the means of three replications. Means in a column followed by superscript letters are not significantly different according to Duncan's multiple range test at p < 0.05. Values in parentheses are arcsine transformed values.

(a) Salicylic acid (b) Control

**Fig. 4.** Growth promotion under glasshouse condition.

**Effect of biotic inducers on yield attributes.** The inducers primed plants enhance the yield parameters of sesame seedlings. The maximum number of capsules were observed in the plants sprayed with MeJA 150ppm (54 capsules /plant) followed by MeJA 50ppm & MeJA 100ppm (44.66 capsules/plant), *Bacillus subtilis* Bbv57 (30.33 capsules/plant), Methyl dematon 25 EC (27.66capsules/plant) while control has recorded minimum number of capsules (21.33 capsules/plant). Similarly, preharvest concentrations of 0.01 and 0.1 mmol $L^{-1}$ In the 'Magenta' and 'Crimson' table grape varieties, MeJA treatments enhanced berry size and overall yield (García-Pastor *et al*., 2019). All other treatments were also observed to significantly increasing the number of capsules per plant after 60 days when compared to control.

**Biochemical analysis**

**Chlorophyll content.** The assessment of chlorophyll content (Fig. 5) in primed sesame seedlings showed that SA 50ppm increased the chlorophyll activity of sesame seedlings up to 99.46, followed by MeJA 150ppm (98.22 mg/$m^2$), MeJA 100ppm (97.22 mg/$m^2$) and MeJA 50ppm (97.02 mg/$m^2$) when compared to control (64.68 mg/$m^2$) and there is an increased chlorophyll content was observed in treatments compared to control. Hence, our study correlates with the reports of SA application consistently improved the chlorophyll content of plant leaves, as reported by Moharekar *et al.* (2003) in wheat and Yildirim *et al.* (2008) in cucumber. However, their combination treatment (SA + JA) was unquestionably more effective in reducing the negative effects of salinity on total chlorophylls and carotenoids in lemon balm. Application of SA and JA against salt-stressed plant exhibited dramatically enhanced chlorophyll concentrations (Pazoki, 2015).

**Total phenol content.** Phenol content analysis (Fig. 6) in primed seedlings of sesame accounted that SA 50ppm increased the phenolic activity of sesame seedlings up to 1.93, followed by SA 100ppm (1.89 $min^{-1}$ $g^{-1}$), SA 150ppm (1.76 $min^{-1}$ $g^{-1}$) and BABA 50ppm (1.72 $min^{-1}$ $g^{-1}$) compared to control (1.31 $min^{-1}$ $g^{-1}$). Similar studies reported that, SA treatment significantly increased the amount of total phenols in broccoli sprouts (Balibrea *et al*., 2011).

**Plant defense enzyme assay.** The defence enzyme analysis of peroxidase (PO) showed that SA 100ppm increased the enzyme activity of sesame seedlings up to 2.87 $min^{-1}$ $g^{-1}$ at 52 hrs, followed by SA 150ppm (2.84 $min^{-1}$ $g^{-1}$), SA 50ppm (2.82 $min^{-1}$ $g^{-1}$) and MeJA 100ppm (2.78 $min^{-1}$ $g^{-1}$) when compared to control (2.39 $min^{-1}$ $g^{-1}$) and there is an decreased enzyme activity was observed after 76hrs in the primed sesame seedlings (Fig. 7). Similar reports were revealed that peroxidase enzyme activity is increased by the treatments with salicylate increased one peroxidase isoform's activity in the leaves of *Quercus rubra* L. seedlings (Steven and Jack 2004).

Polyphenol oxidase (PPO) analysis showed similar results like PO that SA 50ppm increased the enzyme activity of primed sesame seedlings up to 2.9 $min^{-1}$ $g^{-1}$ at 52 hrs, followed by SA 100ppm (2.42 $min^{-1}$ $g^{-1}$), BABA 50ppm (2.39 $min^{-1}$ $g^{-1}$) and BABA 150ppm (2.39 $min^{-1}$ $g^{-1}$) when compared to control (2.26 $min^{-1}$ $g^{-1}$) and the enzyme activity was decrease after 76hrs of spraying (Fig. 8). According to Szepsi *et al.* (2005), pre-treating tomato plants with SA prior to salt stress increased antioxidant enzyme activity (PPO), boosting the plants' capacity to withstand stress was reported similarly. In addition to, this methyl jasmonate-induced expression was also confirmed by Koussevitzky *et al.* (2004), showing that pre-treatment with methyl jasmonate enhanced tomato PPO import and processing into chloroplasts.

The results Phenylalanine ammonia lyase (PAL) showed that enzyme activity of primed sesame

seedlings was maximum in SA 100ppm (1.88 $min^{-1}$ $g^{-1}$) at 52 hrs after spraying (Fig. 9), followed by SA 50ppm (1.83 $min^{-1}$ $g^{-1}$), SA 150ppm (1.81 $min^{-1}$ $g^{-1}$) and MeJA 150ppm (1.68 $min^{-1}$ $g^{-1}$) when compared to control (1.42 $min^{-1}$ $g^{-1}$) and our study correlated with the reports of Zeng *et al.* (2006), treatment with salicylic acid (SA) can increase the activities of PAL which are crucial for the disease resistance of mango fruit.

The enzyme analysis of super oxide disumutase (SOD) showed that SA 50ppm increased the enzyme activity of primed sesame seedlings up to 1.98 $min^{-1}$ $g^{-1}$ at 52 hrs, followed by SA 100ppm (1.85 $min^{-1}$ $g^{-1}$), SA 150ppm (1.80 $min^{-1}$ $g^{-1}$) and MeJA 50ppm (1.74 $min^{-1}$ $g^{-1}$) when compared to control (1.30 $min^{-1}$ $g^{-1}$) and reduction in enzyme activity was observed after 76hrs of spraying (Fig. 10). similar reports shows that CAT, GR, and SOD activity are induced by salicylic acid have also been documented by Clark *et al*. (2002) and Molina *et al.* (2002). It was found that SA treatment of wheat plants cultivated in ideal temperature conditions increased SOD and APX activity (Agarwal *et al*., 2005).

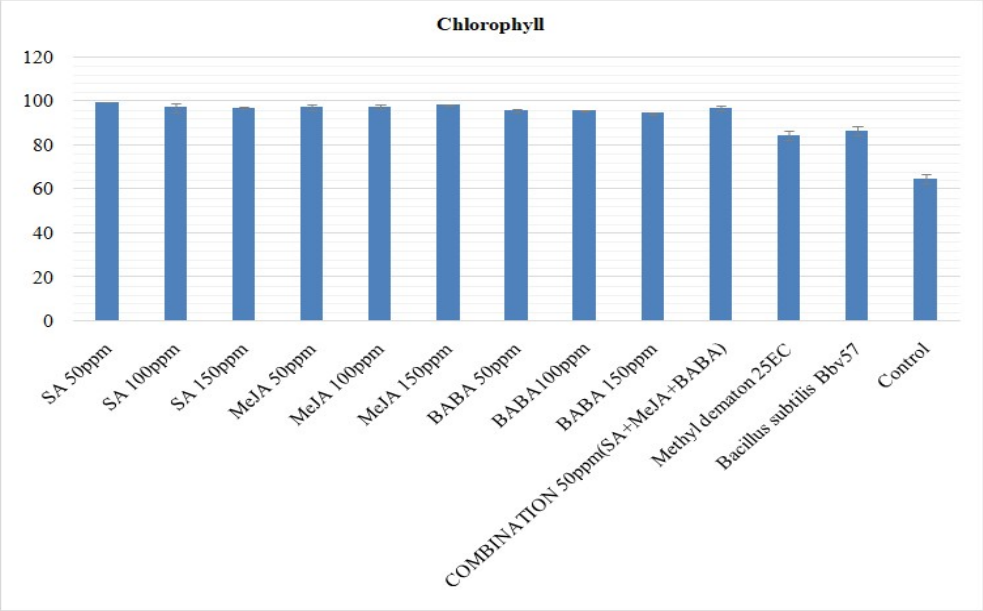



**Fig. 5.** Effects of biotic inducers at various concentration on chlorophyll content.

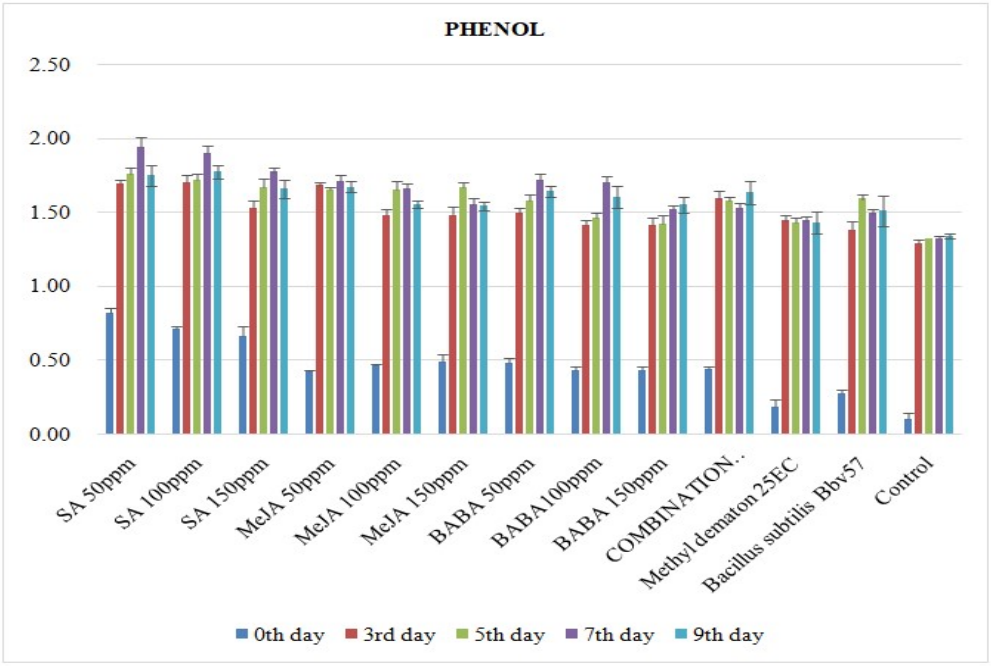



**Fig. 6.** Effects of biotic inducers at various concentration on phenol content.

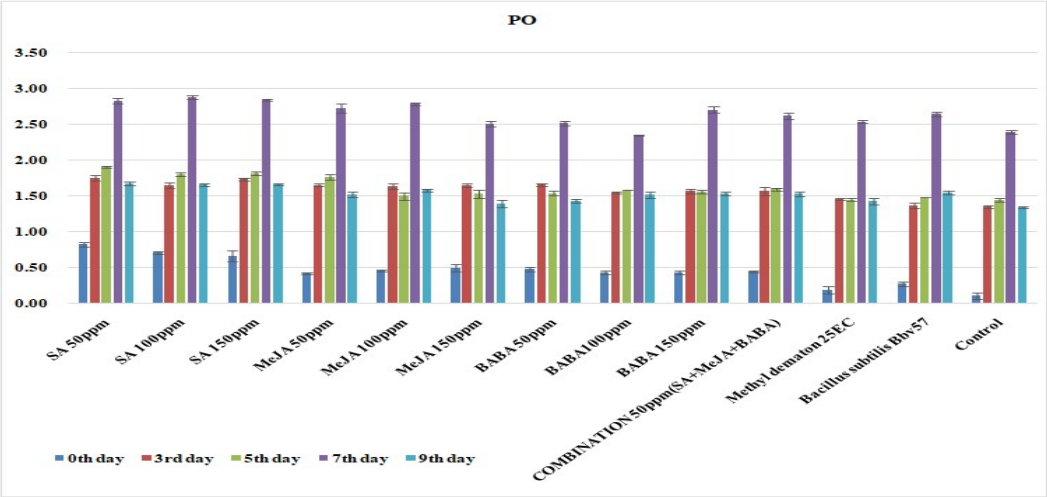



**Fig. 7.** Effects of biotic inducers at various concentration on peroxidase enzyme (PO).

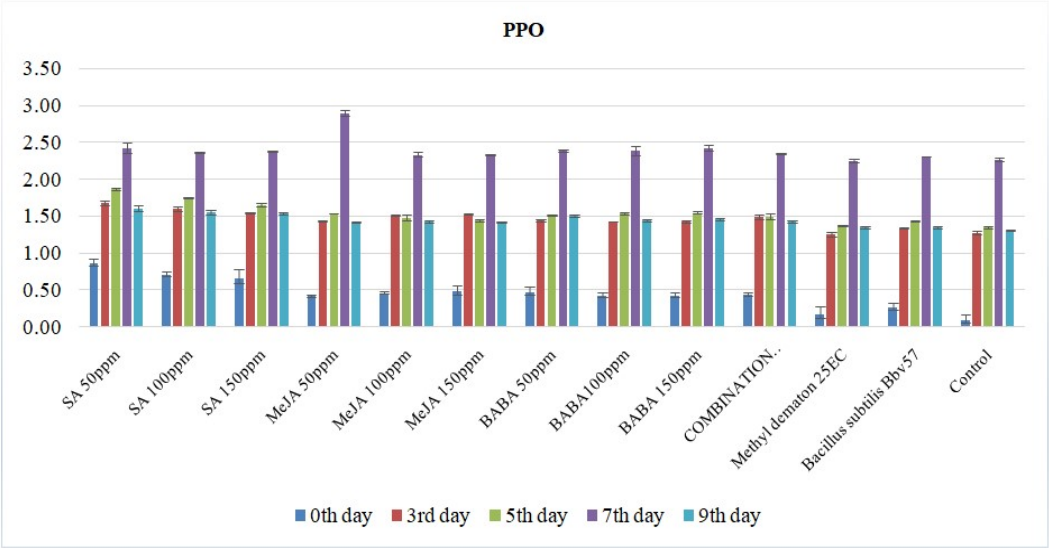



**Fig. 8.** Effects of biotic inducers at various concentration on polyphenol oxidase enzyme (PPO).

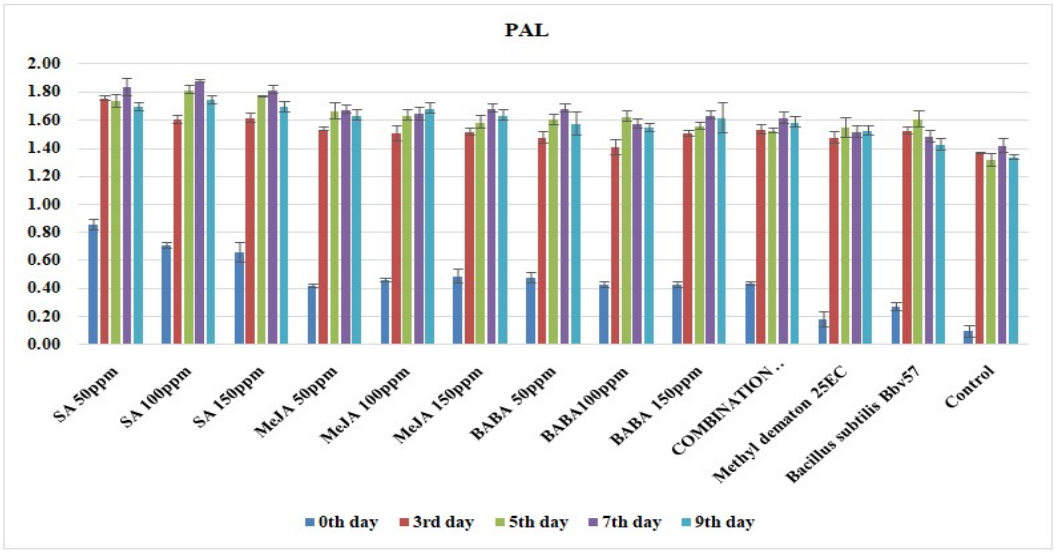



**Fig. 9.** Effects of biotic inducers at various concentration on Phenylalanine ammonia lyase enzyme (PAL).

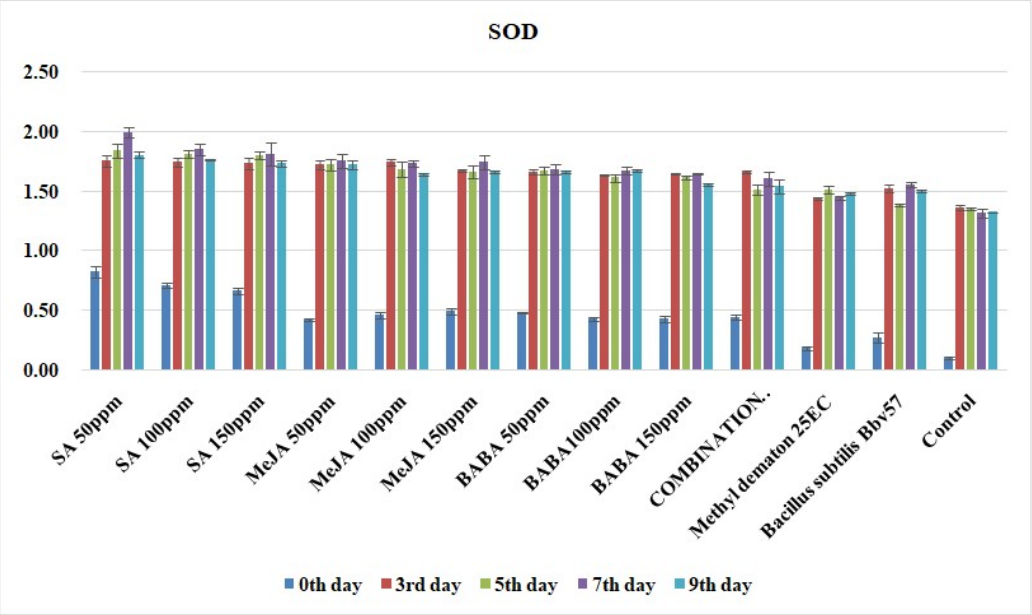



**Fig. 10.** Effects of biotic inducers at various concentration on Super oxide dismutase enzyme (SOD).

## CONCLUSION

Based on the above results, it could be concluded that the application of biotic inducers could be a useful technique to enhance the growth promotion of sesame. As a result, the SA 50ppm treatment had a more effect on growth attributes and activating defense enzymes. On the other hand, maximum number of capsules were observed in MeJA 150ppm primed sesame seedlings when compared to other treatments and control. Thus, this study represents the SA 50ppm is comparably best among the treatments to increase plant growth and activation of defense enzymes.

## FUTURE SCOPE

Many chemicals are used to promote growth and yield promotion in sesame crops but they are Pest resistance, pest resurgence, residue in sprayed crops, and environmental pollution are all consequences of this. It was necessary to find some products that were environmentally safe. Most of the people were unaware about the usage of biotic inducers for plant growth and yield promotion and it can able to activate defense enzymes also. So, In order to promote growth and yield in sesame, biotic inducers such as Salicylic acid, Methyl Jasmote and Beta amino butyric acid could be an alternative choice.

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**Conflict of Interest.** None.

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