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Integrated effect of *Trichoderma harzianum* with selected botanical extracts on *Alternaria* leaf spot of broccoli (*Brassica oleracea* var. *italica*) caused by *Alternaria brassicae* (Berk.) Sacc

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ABSTRACT

Broccoli (*Brassica oleracea* var. *italica*) is an important cole crop vegetable, belong to family Brassicaceae. *Alternaria* leaf spot caused by *Alternaria brassicae* is one of the serious diseases in broccoli. *In vitro* and *in vivo* experiment were conducted at the research plot of the Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, Uttar Pradesh in *Rabi* season 2023-2024, to evaluate *Trichoderma harzianum* and botanicals on *Alternaria* leaf spot of broccoli caused by *A. brassicae*. *In vitro* condition, five botanical extracts were taken at the concentration 10% and *T. harzianum* at 10% and Mancozeb at 0.2% (treated check). *In vivo* condition, eight treatments were taken and all seedlings were treated with *T. harzianum* at 10%, Mancozeb at 0.2% and foliar spray of botanical extracts @ 10% except T_0 – control (untreated check) and (treated check). *In vitro* condition, among the treatments Neem leaf extract at 10% were observed effective in the per cent disease inhibition (%) (79.77) followed by Eucalyptus leaf extract at 10% (75.69). Among the treatments, under *In vivo* condition, T_3 – *T. harzianum* (S.T.) + Neem leaf extract (F.S.) was observed minimum disease intensity (16.59, 19.85 and 23.28 %) at 45, 60 and 75 DAT, respectively. Maximum plant height (9.78, 23.84 and 36.66 cm) at 30, 60 and 90 DAT, number of leaves per plant (9.36, 17.26 and 28.00), head weight (0.38 kg) and yield (9.68 t/ha) were found in T_3 – *T. harzianum* (S.T.) and Neem leaf extract (F.S.) when compared to treated T_7 – mancozeb (treated check) and T_0 – Control (untreated check).

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Keywords: *Alternaria* leaf spot, *Alternaria brassicae*, botanicals plant extracts, broccoli, *Trichoderma harzianum*.

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26 **1. INTRODUCTION**

27 "Broccoli (*Brassica oleracea* var. *italica*) is an important cole crop vegetable, belong to
 28 Brassicaceae family originated from Italy about more than 2000 years ago (Buck, 1956).
 29 Broccoli is native to the eastern Mediterranean and Asia minor, broccoli was cultivated in Italy
 30 in ancient times. It was introduced to England in the 18th century and became popular
 31 in the United States in the 20th century" (Moreno *et al.*, 2007). "The common English name
 32 broccoli is derived from the Italian word 'broccolo', meaning 'the flowering crest of a cabbage'.
 33 Fresh broccoli is dark green in colour with firm stalks and compact bud clusters. As a
 34 vegetable, it is grown for its edible flower buds and stalk" (Das *et al.*, 2021).

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36 "Broccoli is a biennial plant belonging to the Brassicaceae family that is eaten as a vegetable
 37 throughout the world. The edible plant parts are the stalk and large flowering head. Broccoli is
 38 a rich source of vitamin C and vitamin K. Broccoli became one of the favourite foods due to its
 39 high nutrient and fiber content. Broccoli also contains numerous phytochemicals, such as
 40 polyphenols, namely kaempferol, quercetin glucosides, isorhamnetin, glucosinolates, and
 41 their derivatives. These are responsible for its antioxidant and anticancer properties and other
 42 health benefits". (Nagraj *et al.*, 2020). 43

44 "Broccoli is considered a cool season crop, which has now been distributed to both tropical and
 45 subtropical areas" (Shivakumar *et al.*, 2022). "This crop can be cultivated in various parts of
 46 India during the winter season, especially in regions where rainfall is not excessive. Areas with
 47 moderate rainfall and cooler temperatures are generally suitable for broccoli farming. Being a
 48 cool season crop, it requires 15-20°C temperature for head creation. Temperature above 25°
 49 C is not favourable for its development and can cause slacking and darting of heads" (Kumar
 50 *et al.*, 2021).

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52 "The global production of broccoli and cauliflower was 25,531,274 tons from an area of
 53 1,357,186 ha, in which Europe ranks third (after China and India) and contributes 9.5 and
 54 10.5% in production and area, respectively" (Siomos *et al.*, 2022). "This vegetable, closely
 55 resembling cauliflower but usually green in color, introduced in India many years after cabbage
 56 and cauliflower and has gained popularity in short span of time. Now, India stands at second
 57 position for cauliflower and broccoli production with an annual production of 6.7 million tonnes"
 58 (Tejaswini *et al.*, 2018). "It is mostly cultivated in hilly areas of Himachal Pradesh, Uttar
 59 Pradesh, Jammu and Kashmir, Nilgiri Hills and Northern plains of India" (Meena *et al.*, 2020). 60
 61 "There are many diseases and pests collectively that lower the yield and quality of broccoli
 62 crops. Broccoli is infected by various types of pathogens including bacteria, fungi, and viruses,
 63 which lead to reduction of yield and quality" (Zhan *et al.*, 2022). "Among all the diseases
 64 Alternaria leaf spot disease caused by *Alternaria* sp. is the most important major disease which
 65 resulted in significant decline in the yield of all cultivars" (Lafi *et al.*, 2022).

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67 "Among the different diseases caused by the genus *Alternaria*, Alternaria leaf spot disease
 68 is most dominant one that causes average yield loss in the range of 32-57%" (Singh *et al.*,
 69 2015). "During 2017 and 2018, severe symptoms of dark spots on leaves were observed on
 70 broccoli plants (*Brassica oleracea* L. var. *italica*) cultivar with 45 to 37% disease incidence and
 71 70% yield loss" (De Britto *et al.*, 2020).

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73 For the management of this disease fungicides have been recommended to manage the
 74 disease, but present-day farmer perceptions and environmental hazards are compelling to
 75 search for alternative eco-friendly disease management strategies (Sakhare *et al.*, 2021) So
 76 this situation compels to focus on disease management by utilizing biological agents, plant
 77 extracts and fungicides in lowest concentration. Application of biological agents and extracts

78 eco-friendly and a sustainable approach apart from being a promising alternative of fungicide
79 application. In the present study among the plant extracts, *Lantana camara* was found to be
80 effective in managing the *Alternaria* blight infection in the field (71.92% reduction in disease
81 severity and 68.18% increase in yield) in comparison to bio-agent (*Trichoderma viride*) 36.27%
82 reduction in disease severity respectively. (Ahmad and Ashraf, 2016).

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84 2. Materials and Methods

85 The experiment was conducted at the research plot of the Department of Plant Pathology,
86 Central Research Field, Sam Higginbottom University of Agriculture, Technology and
87 Sciences, Prayagraj during the *Rabi* season of 2023-2024. Field experiment was laid out in
88 Randomized Block Design with eight treatments having three replications. 89

90 2.1 Isolation and identification of pathogen

91 The leaves were collected from infected plants bearing characteristic symptoms of *Alternaria*
92 leaf spot of broccoli. The leaves were thoroughly washed under running tap water. The
93 symptoms on leaves after mounting on slides were examined under microscope to confirm the
94 presence of *Alternaria* sp. The infected leaf parts along with the healthy portion was cut into
95 small pieces under aseptic conditions and surface sterilized with 1% sodium hypochlorite
96 (NaOCl) solution for 1 minute and washed three times with sterile distilled water to remove
97 any traces of sodium hypochlorite (NaOCl) adhered with leaf bits. Then they were placed on
98 filter papers so that extra water gets absorbed. After that five leaf bits were transferred on PDA
99 media (Tuite, 1969) contained in sterilized petriplates with the help of forceps. To avoid
100 bacterial contamination streptomycin @ 100 ppm, was added in the medium at lukewarm
101 stage before pouring PDA into Petriplates. Then Petriplates were wrapped and incubated at
102 27±2°C in BOD, after 3 days mycelial growth was observed around leaf bits. With the help of
103 cork borer a portion from the periphery having single hyphal tip from this colony growth was
104 separated and transferred to other petriplates having medium to get pure culture and
105 identification of the pathogen was recorded by observing the morphological features of colony,
106 spore characteristics and referring the relevant literature (Barnett and Hunter, 1972).

107 2.2 Evaluation of botanical extracts and *Trichoderma harzianum* against 108 *Alternaria* leaf spot of broccoli

109 2.2.1 In vivo evaluation

110 Botanical extracts were prepared using method of standard procedure given by Mahapatra
111 and Das (2013). Matured leaves and other botanicals were collected and sterilized with distilled
112 water, the leaves were homogenized in a pestle and mortar, sterilized distilled water. Aqueous
113 extract of this botanical (1% w/v) were prepared by mixing 100 g fresh leaves/botanicals of
114 plant with 100 ml of sterile distilled water and crushing in a warring blender. The extract was
115 filtered through four layers of moist muslin cloth. The filtrate thus obtained was considered
116 as stock solution. The phyto-extracts were sprayed @ 10% prepared from standard solution,
117 Mancozeb at ratio 0.2%, fungal bio-agent *T. harzianum* were also sprayed @ 10%
118 after the appearance of the first symptoms in the field at thirty days after transplanting (DAT),
119 the foliar spray of botanicals, mancozeb and *T. harzianum* was applied three times
120 at fifteen days interval. Unsprayed plots were kept as control.

121 2.2.2 Disease Intensity

122 The intensity of disease was visually assessed in all the plots at weekly interval from first
123 appearance of disease for each treatment. Disease intensity was calculated using the formula
124 given by Wheeler (1969).

$$\text{Disease Intensity (\%)} = \frac{\text{Sum of all disease ratings}}{\text{Total no of ratings} \times \text{Maximum disease grade}} \times 100$$

125 2.2.3 In vitro evaluation

126 Culture of *T.harzianum*

127 One of product was taken and mixed in nine ml of sterilized distilled water in a clean and
 128 sterilized test tube to make 10^{-1} dilution (1:10) shaken well and one ml of the suspension of nine
 129 ml of sterilized water was taken in a tube to make 10^{-2} dilution (1:100). Similarly, four more
 130 serial dilutions in the same way was done to get 10^{-6} dilution and transferred one ml of this
 131 suspension to sterile petri plate containing 15-20 ml of sterilized melted and cooled PDA media
 132 the plates were rotated gently and allowed to solidify. The petri plates were incubated in BOD
 133 incubator at $25 \pm 2^\circ\text{C}$ for five to seven days. The development of typical colony of
 134 *T.harzianum* was observed.

135 Preparation of plant extracts

136 The botanicals used for the experiment were Neem, Black nightshade, Eucalyptus, Datura,
 137 Lantana. The botanical parts used for the treatment were Neem leaf extract 10%, Black night
 138 shade leaf extract 10%, Eucalyptus leaf extract 10%, Datura leaf extract 10%, Lantana leaf
 139 extract 10%. Aqueous plant extracts were obtained as per the method described by **Bhatti,**
 140 **(1988)**. A 100-gram sample of each fresh leaves were gently washed under running tap water
 141 and again washed in sterile distilled water. Then each sample was ground separately by using
 142 sterile pestle and mortar with 100 ml sterile distilled water. The extract of each sample
 143 thus obtained was filtered separately through sterilized double layered muslin cloth to
 144 remove the bits of plant material as filtrate. The extract was again filtered through a filter paper
 145 (Whatman No. 1). This formed the standard plant extract solution (100%). All the glassware
 146 used in the study were sterilized before use. All the plant extracts were tested at 10 percent
 147 concentration against the test pathogen using PDA as a basal medium. To obtain 10 percent
 148 concentration of plant extracts, 100 ml of lukewarm PDA was mixed with 10 ml of standard
 149 plant extracts in 250 ml conical flask, separately and then it was stirred well to obtain
 150 homogenized mixture.

151 2.2.4 Dual Culture Technique

152 Twenty ml of sterilized and cooled potato dextrose agar medium was poured into sterilized
 153 petri plates. Fungal antagonists were evaluated by inoculating a pathogen at one side of the
 154 petri plate and the antagonist at exactly opposite side of the same plate by leaving a space of
 155 4 cm. After required period of incubation i.e., when the growth in control plate records 90 mm
 156 in diameter, the radial growth of the pathogen was measured (**Vincent, 1947**).

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158 Percentage inhibition of mycelial growth of test pathogen was calculated using the formula:

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$$I = \frac{C - T}{C} \times 100$$

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167 Where,

168 I = Percent reduction in growth of test pathogen

169 C = Radial growth (mm) in control

170 T = Radial growth (mm) in treatment

171 2.2.5 Poisoned food technique

172 Five mm diameter of cultured disc of *A.brassicae* was kept at the center of each petri
 173 plate containing the botanicals of required concentration dissolved in PDA. Three replications
 174 were maintained. The plates were incubated at $27 \pm 2^\circ\text{C}$ for seven days and colony diameter
 175 was recorded (**Vincent, 1947**).

176 Percent inhibition of mycelial growth was calculated by using the formula:

$$I = \frac{C-T}{C} \times 100$$

171 Where,

172 I=Percent inhibition

173 C=Growth(mm)of test fungus in untreated control plate

174 T=Growth(mm)of test fungus in treated plate

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176 3.RESULTS AND DISCUSSION

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178 3.1 Effect of plant extract on *A. brassicae* by poison food technique

179 Radial growth (mm) of *A. brassicae*

180 The data presented in table 1, depicted reveals significant reduction in T₃- Neem leaf extract
 181 (18.20mm) followed by T₂-Eucalyptus leaf extract (21.87mm), T₅-Black nightshade leaf
 182 extract (23.83mm), T₄ -Lantanaleaf extract (26.00mm), T₁-Daturaleaf extract
 183 (26.98mm) as compared to mancozeb (treated check)-(2.50mm) and untreated check
 184 control T₀-(90mm).

185 Percent inhibition of mycelial growth

186 The data presented in table 1 and figure 1 reveal that there was significant increase
 187 in T₃-Neem leaf extract at 10% (79.77%) followed by T₂-Eucalyptus leaf extract at 10%
 188 (75.7%), T₅-Black nightshade leaf extract at 10% (73.52%), T₄-Lantanaleaf extract at 10%
 189 (71.11%), T₁-Daturaleaf extract at 10% (70.02%) as compared to T₇-mancozeb (treated
 190 check) (97.21%) and untreated check control T₀-(0%). The probable reason for such findings
 191 may be due to the fact that *Neem* leaf contains active compounds such as azadirachtin as well
 192 as salannin, nimbim. Constituents such as nimbim, nimbidin, limonoids which disrupt the cell
 193 of the pathogen and restrict the performance of some enzymes that are important to proliferate
 194 fungi in nature and lead to their death. The current findings are consistent with the research
 195 conducted by [Vijaykumar et al. \(2022\)](#) and [Kumar et al. \(2007\)](#) who tested the effectiveness
 196 of different phytoextracts against *A. brassicae* and found that the antifungal activity
 197 and potentiality of nimbicidin and *Neem* leaf extract have been proved successfully against
 198 the causal

fungus..

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202 3.2 Effect of treatments on disease intensity of *Alternaria* leaf spot of broccoli

203 caused by *A. brassicae*

204 The data presented in the [Table \(2\)](#) reveal that the disease intensity (%) on broccoli is significantly
 205 reduced by T₃- *T. harzianum*+Neem leaf extract at 10% (23.28%) followed by T₆-
 206 (*T. harzianum* at 10% (26.31%), T₅- (*T. harzianum*+Black nightshade
 207 leaf extract at 10% (30.26%), T₄- (*T. harzianum*+Lantanaleaf extract at 10%
 208 (32.56%), T₂- *T. harzianum*+Eucalyptus leaf extract at 10% (36.83%), T₁-
 209 (*T. harzianum*+Daturaleaf extract at 10% (37.28%) as compared to the T₇-
 210 mancozeb at 0.2% (treated check) (18.96%) as well as (T₀)-control (untreated check)
 211 (40.64%). Similar findings have been reported by [Anshika and Zacharia \(2023\)](#) and [Ravella](#)
 212 [et al. \(2023\)](#) The probable reason for this result may be due to the *Neem* having anti-microbial
 213 activity. *Neem* has reported to exhibit antifungal, antibacterial and insecticidal properties. The
 214 ethanolic extract of *Neem* leaves stops the growth of fungi. *T. harzianum* has shown
 215 significant results on disease intensity and yield attributes, whereas yield has been
 216 significantly increased and decreased disease intensity which may be due to the mycoparasite

217 nature and ability to generate volatile and non-volatile compounds against pathogen with great
218 anti-
microbial activity. 219

220 3.3 Effect of treatments on plant height (cm) of broccoli

221
222 The data presented in the Table (3) reveal that the plant height (cm) on broccoli significantly
223 increased plant height in T₃–(*T.harzianum*+ Neem leaf extract at 10% (36.66
224 cm) followed by T₆–(*T.harzianum* at 10% (33.70 cm), T₅–(*T.*
225 *harzianum*+Blacknightshade leaf extract @ 10% (31.10 cm), T₂–(*T.harzianum*
226 +Eucalyptus leaf extract at 10% (29.93 cm), T₄–(*T.harzianum*+Lantana leaf
227 extract at 10% (28.41 cm), T₁–(*T.harzianum*+Datura leaf extract at 10% (27.78
228 cm) as compared to the T₇–Mancozeb at 0.2% (treated check) (39.47 cm) as well as (T₀)
229 –control (untreated check) (23.01 cm). The probable reason for this result may be due to
230 **Neem** aqueous extracts showed a promotive effect on shoot lengths, branches and leaves
231 numbers. The bio-efficacy of **Neem** extract over pathogens can be attributed to the fact that
232 **Neem** has active compounds such as azadirachtin, nimbin, nimbidin, nimbinene and
233 azadirone which are antifungal, anti-bacterial and anti-insecticidal in nature **Nahak et al.**
234 **(2015)**. *Trichoderma* strains colonise plant roots, establishing chemical communication and
235 systemically altering the expression of numerous plant genes that alter plant physiology.
236 Application of *T.harzianum* significantly increased curd circumference length, the
237 diameter of the stem **Rosa et al. (2019)**.

238 3.4 Effect of treatments on number of leaves per plant of broccoli

239 The data presented in Table (4) reveal that number of leaves significantly increased in T₃–
240 (*T.harzianum*+Neem leaf extract at 10% (28.00) followed by T₆–(*T.*
241 *harzianum* at 10% (25.20), T₅–(*T.harzianum*+Blacknightshade leaf extract
242 at 10% (23.40), T₂–(*T.harzianum*+Eucalyptus leaf extract at 10% (21.13), T₄
243 –(*T.harzianum*+Datura leaf extract at 10% (20.66), T₁–(*T.harzianum*+Lantana
244 leaf extract at 10% (20.40) as compared to the T₇–Mancozeb at
245 0.2% (treated check) (31.53) as well as (T₀) control (untreated check) (16.00). The probable
246 reason for this result may be due to **Neem** aqueous extracts showed a promotive effect on
247 shoot lengths, branches and leaves numbers. The bio-efficacy of **Neem** extract over
248 pathogens can be attributed to the fact that **Neem** has active compounds such as
249 azadirachtin, nimbin, nimbidin, nimbinene and azadirone which are antifungal, antibacterial
250 and anti-insecticidal in nature **Nahak et al. (2015)**. *Trichoderma* strains colonise plant roots,
251 establishing chemical communication and systemically altering the expression of numerous
252 plant genes that alter plant physiology. Application of *T.harzianum* significantly
253 increased curd circumference length, the diameter of the stem, and the leaf greenness index
254 value **Rosa et al. (2019)**.

255 3.5 Effect of treatments on head weight (kg) of broccoli

256 The data presented in Table (5) and Figure (2) reveal that the head weight (kg) of broccoli
257 significantly increased in T₃–*T.harzianum*+Neem leaf extract at 10% (0.38 kg)
258 followed by T₆–*T.harzianum* at 10% (0.35 kg), T₅–*T.harzianum*+
259 Blacknightshade leaf extract at 10% (0.304 kg), T₄–*T.harzianum*+Lantana leaf
260 extract at 10% (0.301 kg), T₂–(*T.harzianum*+Eucalyptus at 10% (0.21), T₁–
261 *T.harzianum*+Datura leaf extract at 10% (0.20 kg) as compared to the T₇–
262 Mancozeb at 0.2% (treated check) (0.40 kg) as well as (T₀)–control (untreated check)
263 (0.16 kg). The probable reason for such findings may be that the *Trichoderma* and plant
264 interaction produce these secondary metabolites such as auxin-like compounds or auxin-

265 inducing substances that resulted in improved growth, head diameter and yield **Tanwaret**
 266 **al. (2013)**. Neem extract is composed of antimicrobial ingredients such as alkaloids,
 267 glycosides, flavonoids, and saponins, which are common antibiotics found in plants. At
 268 concentrations of 0.1 and 0.4 g mL⁻¹, **Neem** extract inhibited the growth of soil
 269 microorganisms **Sarawaneeyaruk et al. (2015)**.

270 3.6 Effect of treatments on yield (t/ha) of broccoli

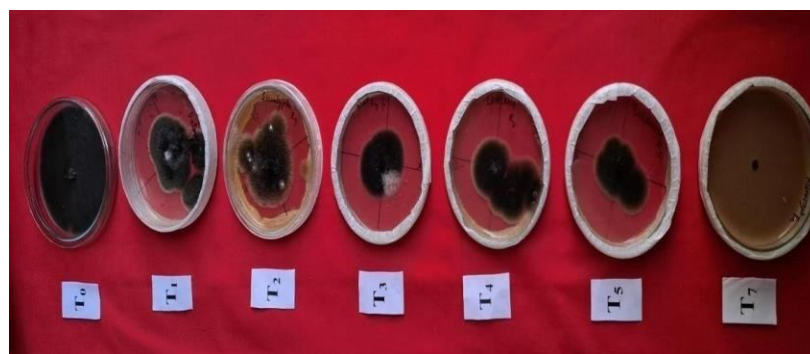
271 The data presented in **Table (6)** and **Figure (3)** revealed that yield of broccoli is significantly
 272 increased head weight T3– *T. harzianum* + Neem leaf extract at 10% (9.68
 273 t ha⁻¹) followed by T6– *T. harzianum* at 10% (7.92 t/ha), T5– (*T.*
 274 *harzianum* + Black night shade leaf extract at 10% (6.81 t ha⁻¹), T4– (*T. harzianum*
 275 + Lantana leaf extract at 10% (6.50 t ha⁻¹), T4– (*T. harzianum* + Eucalyptus leaf
 276 extract at 10% (6.19 t ha⁻¹), T1– (*T. harzianum* + Datura leaf extract @ 10% (5.68
 277 t ha⁻¹) as compared to the T7– Mancozeb at 0.2% (treated check) (11.55 t ha⁻¹) as well as
 278 (T0)– control (untreated check) (3.85 t/ha). The probable reason for this result may be due
 279 to the production of secondary metabolites such as antibiotics, isocyanide, acids and cell wall
 280 degrading enzymes which are implicated in the growth inhibition of many
 281 phytopathogenic fungi **Supriya et al. (2022)**. *T. viride* has shown significant
 282 results on yield attributes, the yield has been significantly increased which may be attributed
 283 to the mycoparasitic nature and ability to generate volatile and non-volatile compounds
 284 against pathogen with great anti-microbial activity **(Ravella et al., 2023)**.

285 **Table 1. Effect of selected plant extracts on radial growth (mm) of *A.***

286 *brassicacae*

| Sr.No. | Treatments | Concentration | Mean colony diameter (mm) | Percent inhibition |
|-----------------|--------------------------------|---------------|---------------------------|--------------------|
| T0 | Control (untreated check) | - | 90.00 | 0.00 |
| T1 | Datura leaf extract | 10% | 26.98 ^a | 70.02 |
| T2 | Eucalyptus leaf extract | 10% | 21.87 | 75.69 |
| T3 | Neem leaf extract | 10% | 18.20 | 79.77 |
| T4 | Lantana leaf extract | 10% | 26.00 ^a | 71.10 |
| T5 | Black night shade leaf extract | 10% | 23.83 | 73.51 |
| T6 | Mancozeb | 0.2% | 2.50 | 97.21 |
| S.Em.(+) | | | 0.51 | 0.57 |
| C.D(5%) | | | 1.59 | 1.76 |

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Figure1:-Efficacyofplantextractonpercentmycelial inhibitionof *A.brassicae*bypoisonedfood technique.

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Table2.Percentdiseaseintensity(%)at45,60and75DAT asaaffectedbytreatments

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| Sr.No. | Treatments | 45DAT | 60DAT | 75DAT |
|-----------------|-------------------------------------------------------------------|-------|-------|-------|
| T ₀ | Control(untreatedcheck) | 25.98 | 33.83 | 40.64 |
| T ₁ | <i>T.harzianum</i> 10%(S.T.)+Daturalea fextract10%(F.S.) | 23.40 | 29.88 | 37.28 |
| T ₂ | <i>T.harzianum</i> 10% (S.T.)+Eucalyptusleaf extract10%(F.S.) | 22.50 | 29.55 | 36.83 |
| T ₃ | <i>T.harzianum</i> 10%(S.T.)+Neem leafextract10%(F.S.) | 16.59 | 19.85 | 23.28 |
| T ₄ | <i>T.harzianum</i> 10%(S.T.)+Lantanale afextract10%(F.S.) | 21.34 | 27.51 | 32.56 |
| T ₅ | <i>T.harzianum</i> 10%(S.T.)+Blacknightshadeleaf extract10%(F.S.) | 20.28 | 25.64 | 30.26 |
| T ₆ | <i>T.harzianum</i> 10%(S.T.)+ <i>T.harzianum</i> 10%(F.S.) | 18.26 | 22.36 | 26.31 |
| T ₇ | Mancozeb(treatedcheck)0.2%(S.T.+F.S.) | 11.66 | 15.90 | 18.96 |
| S.Em.(+) | | 0.36 | 0.30 | 0.34 |
| C.D(5%) | | 1.09 | 0.92 | 1.03 |

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Table3.Effectoftreatmentsonplantheight(cm)ofbroccoliat30,60and90DAT

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| Sr.No. | Treatments | 30DAT | 60DAT | 90DAT |
|----------------|--------------------------------------------------------------|-------|-------|-------|
| T ₀ | Control(untreatedcheck) | 6.92 | 15.40 | 23.01 |
| T ₁ | <i>T.harzianum</i> 10%(S.T.)+Daturaleafextract10% (F.S.) | 7.46 | 17.06 | 27.78 |
| T ₂ | <i>T.harzianum</i> 10%(S.T.)+Eucalyptusleafextract1 0%(F.S.) | 8.96 | 19.16 | 29.93 |
| T ₃ | <i>T.harzianum</i> 10%(S.T.)+Neem leafextract10%(F.S.) | 9.78 | 23.84 | 36.66 |
| T ₄ | <i>T.harzianum</i> 10%(S.T.)+Lantanaleafextract10% (F.S.) | 8.21 | 18.28 | 28.41 |

| | | | | |
|-----------------|-------------------------------------------------------------------|-------|-------|-------|
| T ₅ | <i>T.harzianum</i> 10%(S.T.)+Blacknight shadeleafextract10%(F.S.) | 9.64 | 20.12 | 31.10 |
| T ₆ | <i>T.harzianum</i> 10%(S.T.)+ <i>T.harzianum</i> 10%(F.S.) | 10.43 | 21.37 | 33.70 |
| T ₇ | Mancozeb(treatedcheck)0.2%(S.T.+F.S.) | 12.72 | 26.43 | 39.47 |
| S.Em.(+) | | 0.17 | 0.40 | 0.45 |
| C.D(5%) | | 0.53 | 1.23 | 1.31 |

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Table4.Effectofselectedtreatmentsonnumberofleavesperplantofbroccoli at 30,60and90DAT

| Sr.No. | Treatments | 30DAT | 60DAT | 90DAT |
|-----------------|-------------------------------------------------------------------|--------------|--------------|--------------|
| T ₀ | Control(untreatedcheck) | 7.73 | 10.46 | 16.00 |
| T ₁ | <i>T.harzianum</i> 10%(S.T.)+Daturaleaf extract10%(F.S.) | 8.16 | 12.40 | 20.66 |
| T ₂ | <i>T.harzianum</i> 10%(S.T.)+Eucalyptusleafextract10 % (F.S.) | 8.93 | 13.40 | 21.13 |
| T ₃ | <i>T.harzianum</i> 10%(S.T.)+Neem leafextract10%(F.S.) | 9.36 | 17.26 | 28.00 |
| T ₄ | <i>T.harzianum</i> 10%(S.T.)+Lantanale afextract10%(F.S.) | 8.46 | 12.93 | 20.40 |
| T ₅ | <i>T.harzianum</i> 10%(S.T.)+Blacknightshadeleaf extract10%(F.S.) | 8.86 | 14.16 | 23.40 |
| T ₆ | <i>T.harzianum</i> 10%(S.T.)+ <i>T.harzianum</i> 10%(F.S.) | 9.76 | 15.73 | 25.20 |
| T ₇ | Mancozeb(treatedcheck)0.2%(S.T.+F.S.) | 10.13 | 20.13 | 31.53 |
| S.Em.(+) | | 0.11 | 0.21 | 0.21 |
| C.D(5%) | | 0.33 | 0.66 | 0.64 |

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Table5.Effectoftreatmentsonheadweight(kg)ofbroccoli

| Sr.No. | Treatments | Headweight/kg |
|-----------------|-------------------------------------------------------------------|----------------------|
| T ₀ | Control(untreatedcheck) | 0.16 |
| T ₁ | <i>T.harzianum</i> 10%(S.T.)+Daturaleafextract 10%(F.S.) | 0.20 |
| T ₂ | <i>T.harzianum</i> 10%(S.T.)+Eucalyptusleafextract10%(F.S.) | 0.21 |
| T ₃ | <i>T. harzianum</i> 10%(S.T.)+Neemleafextract 10%(F.S.) | 0.38 |
| T ₄ | <i>T.harzianum</i> 10%(S.T.)+Lantanaleafextract 10%(F.S.) | 0.301 |
| T ₅ | <i>T.harzianum</i> 10%(S.T.)+Blacknightshadeleafextract10%(F. S.) | 0.304 |
| T ₆ | <i>T.harzianum</i> 10%(S.T.)+ <i>T.harzianum</i> 10%(F.S.) | 0.35 |
| T ₇ | Mancozeb(treatedcheck)0.2%(S.T.+F.S.) | 0.40 |
| S.Em.(+) | | 0.05 |
| C.D(5%) | | 0.15 |

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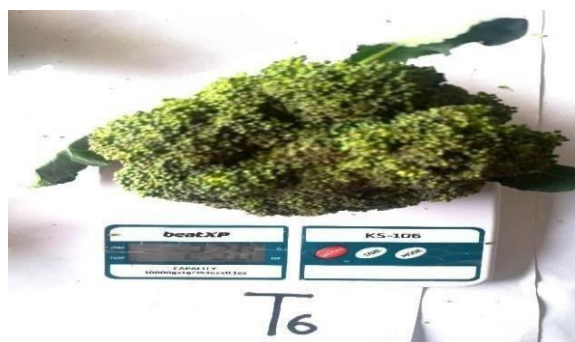


Figure2:-Weighingofbroccoli

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Table6.Effectoftreatmentsontotalyield(t/ha)ofbroccoli

| Sr.No. | Treatments | Yield(t/ha) |
|-----------------|------------------------------------------------------------------|-------------|
| T ₀ | Control(untreatedcheck) | 3.85 |
| T ₁ | <i>T.harzianum</i> 10%(S.T.)+Daturaleafextract 10%(F.S.) | 5.68 |
| T ₂ | <i>T.harzianum</i> 10%(S.T.)+Eucalyptusleafextract10%(F.S.) | 6.19 |
| T ₃ | <i>T.harzianum</i> 10%(S.T.)+Neemleafextract10%(F.S.) | 9.73 |
| T ₄ | <i>T.harzianum</i> 10%(S.T.)+Lantanaleafextract 10%(F.S.) | 6.50 |
| T ₅ | <i>T.harzianum</i> 10%(S.T.)+Blacknightshadeleafextract10%(F.S.) | 6.81 |
| T ₆ | <i>T.harzianum</i> 10%(S.T.)+ <i>T.harzianum</i> 10%(F.S.) | 7.35 |
| T ₇ | Mancozeb(treatedcheck)0.2%(S.T.+F.S.) | 11.55 |
| S.Em.(+) | | 0.12 |
| C.D(5%) | | 0.37 |

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Figure3:-Yieldofbroccoli

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4. CONCLUSION

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It can be concluded that *T. harzianum* and botanicals Neem leaf extract is significant in controlling *A. brassicae* in broccoli. This study contributes valuable insights into sustainable disease management practices by the use of bio-control agent and plant extracts that can benefit both farmers and the agricultural sector. It is important to note that this investigation has been conducted in a specific cropping season within the agro-climatic conditions of Prayagraj. Further trials in diverse locations and seasons are recommended to expand upon these promising results.

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Disclaimer (Artificial intelligence)

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Option 1:

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Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

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Option 2:

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Author(s) hereby declare that generative AI technologies such as Large Language Models, etc have been used during writing or editing of manuscripts. This explanation will include the name, version, model, and source of the generative AI technology and as well as all input prompts provided to the generative AI technology

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Details of the AI usage are given below:

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