Effect of native *Trichoderma viride* and *Pseudomonas fluorescens* on the development of *Cuscuta campestris* on chickpea, *Cicer arietinum*

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**Abstract:** *Cuscuta campestris* Yuncker is a serious parasite on several leguminous crops including chickpea in India. Chickpea is an important pulse crop in India and severe incidence of *Cuscuta* may result in yield loss of about 85.7%. Management of *Cuscuta* is very difficult because of their intricate relationship with the host, wide host range and lack of resistant genes in the host. Thus induced systemic resistance (ISR) by plant growth promoting microbes (microbial elicitors) may be an effective alternative method for the management of *Cuscuta*. In the current study, to induce systemic resistance, native isolates of *Trichoderma viride* Pers. and *Pseudomonas fluorescens* Flügge were used as seed treatments and foliar spray on chickpea and then infested with *C. campestris*. Salicylic acid and thiobenzamidazole (synthetic elicitors) were used as standard inducing agents for comparison. Results indicated that fresh seeds of *C. campestris* germinated rapidly even without scarification and that the germination was not influenced by the proximity of the seeds to the host. Seed treatment followed by foliar sprays with the bioagents and synthetic elicitors induced at 20 and 40 days after sowing (DAS) induced increased production of defense enzymes in chickpea (*Cicer arietinum L.*) and thus delayed the development (1.8-5 days) and flowering (2.4-4.2 days) of *C. campestris*. Treatment with both the elicitors also resulted in the enhanced activities of scavengers of enzymes related reactive oxygen species (ROS). Thus the above work would help in the integration of the application of bioagents for effective management of *Cuscuta* in chickpea.

**Keywords:** Chickpea; *Cuscuta campestris*, Defense enzymes, ISR, *Pseudomonas fluorescens*, *Trichoderma viride*

**INTRODUCTION**

*Cuscuta* spp. (commonly called as Dodder) are rootless, chlorophyllous, heterotrophic, obligate angiosperms twining on dicotyledonous crops. They belong to the family Cuscutaceae (earlier known as Convolvulaceae), containing about 170 different species distributed throughout the world (Holm *et al.*, 1997). *Cuscuta* are broadly nonspecific; attacks a wide range of plant species including many cultivated plants and dicotyledonous weeds, but rarely the monocotyledonous plants (Wright *et al.*, 2011). *Cuscuta* enjoys a very intimate relationship with the host plants throughout its life cycle, except for a very short, post germination independent period of about 8-10 days, in which even a two way transfer of genes between host and the parasite has been reported (Mower *et al.*, 2004). Among the 12 different species of *Cuscuta* reported to occur in India, *C. campestris* and *C. reflexa* are most common and cause significant economic losses on crops like niger, lucerne, berseem and chickpea (Gaur, 1999). Incidence of *Cuscuta* spp. is reported mainly in the states of Andhra Pradesh, Chhattisgarh, Gujarat, Odisha, West Bengal and Madhya Pradesh on oilseed crops like niger, linseed, pulses viz., blackgram, greengram, lentil, chickpea (prominently in rice-fallows) and fodder crops including lucerne, berseem (Mishra, 2009). Chickpea is an important pulse crop, cultivated in about 8.56 million ha with an annual production of 7.35 million tones and India is the largest producer, accounting for nearly 64% of the global production (Gaur *et al.*, 2010). Vyas and Joshi (1975) first reported the incidence of *Cuscuta* sp on chickpea in the state of Uttar Pradesh, in India. Mishra (2009) reported that *C. campestris* is the dominant species attacking chickpea in India. Yield loss of about 85.7 % has been reported in chickpea as a result of *Cuscuta* infestation (Moorthy *et al.*, 2003) and 54.7 to 98.7 % by 1-10 *C. campestris* twines /m² (Mishra, 2009). The choice of chemicals for control of *Cuscuta* is very limited. Pre-plant incorporation and post emergence application of imazethapyr at 75 g/ha produced better control of the *Cuscuta* on various crops (Mishra *et al.*, 2007). Inherent genetic resistance in the host against *Cuscuta* is not very common (Lanini and Kogan, 2005) and crop rotation is not a feasible technique often because of its wide host range. Thus induced systemic
resistance (ISR) by microbes is thought of as an integrated strategy in the management of *Cuscuta*. Plant growth promoting microbes induce resistance in plants by activation of host anti-stress genes to produce more defense proteins and phytoalexins against plant pathogens (Van loon et al. 1998; Kannan and Karthik, 2009; Sriram et al., 2009), alter the composition of host root exudates and their volatile signaling molecules (Harsh et al., 2006), thereby interfering with the recognition of the host by the parasite. *Cuscuta* resembles the plant pathogenic fungi in the use of haustoria as the main invading organ to infect and establish in the host (Meyer, 2006) and thus would fit well in the scheme of management by ISR. Keeping this in view, the present investigation was conducted to study the effect of native *Trichoderma viride* and *Pseudomonas fluorescens* on the development of *Cuscuta campestris* on chickpea (*Cicer arietinum*). This study would help in the integrated management of *Cuscuta* by means of application of microbes at appropriate stages of cultivation. Further since the awareness about the ill effects of more usage of pesticides is increasing, this safe and natural method of management using friendly microbes would be of significant importance in the overall strategy for the management of this dreaded weed.

**MATERIALS AND METHODS**

**Location:** Experiments were conducted in controlled conditions in the containment facility at the Directorate of Weed Science Research (DWSR), Jabalpur (23°13’59.00"N, 79°58’02.25"E, elevation 390.45m) during 2009 to 2012. *C. campestris* seeds were collected from the farmer’s fields in Mandla district of Madhya Pradesh (23°31’51.54"N, 80°27’55.49"E, elevation 456.60 m).

**Antagonistic microbes:** Fungal and bacterial bioagents were isolated from native soils of chickpea using appropriate selective media viz., *Trichoderma* selective medium (Elad et al., 1981) for *T. viride* and King’s B for *P. fluorescens*. To prevent attenuation the bioagents were periodically inoculated in the pots with chickpea infested by *C. campestris* and again reisolated.

**Effect of antagonists and synthetic elicitors on Cuscuta C. campestris on chickpea:** Plastic tubs of size 30 cm³ were filled with pot mixture containing sterilized soil, sand and decomposed (Farm Yard Manure) FYM (1:1:1). Locally popular variety of chickpea, JG-16 was sown and seedlings were thinned to maintain 2 healthy seedlings per pot. Seeds of *C. campestris* was sown by thoroughly mixing about 20 seeds with the top soil of the pot and with a rose can, watered gently using tap water (EC = 2 ds/m, pH = 7.1).

Antagonists were multiplied in their respective broths, 6 days (*P. fluorescens*) and 10 days (*T. viride*) by incubating at 30 °C in a shaking incubator, after which the broth solution along with the microbial mat was collected, homogenized in a blender and applied as foliar spray or used for seed treatment. Synthetic elicitors 0.5M viz., salicylic acid and thiobenzamidazole (Bion 50% obtained from M/S Syngenta India Ltd.) were similarly used for comparison. Chickpea treated with distilled water was maintained as control.

**Antagonists- chickpea-Cuscuta interactions:** To study Induced Systemic Resistance (ISR) in chickpea, the potted plants were treated with the bioagents and infested with *Cuscuta*. The treatments with five replications per treatment are given as follows:

- **T₁:** Seed treatment with *P. fluorescens*
- **T₂:** Seed treatment with *T. viride*
- **T₃:** Seed treatment with salicylic acid (0.05M)
- **T₄:** Foliar spray with *P. fluorescens* at 20 DAS and 40 days
- **T₅:** Foliar spray with *T. viride* at 20 DAS and 40 days
- **T₆:** Foliar spray with salicylic acid (0.05M) at 20 DAS and 40 DAS
- **T₇:** Negative control (Chickpea+*Cuscuta*)
- **T₈:** Control (only chickpea)

Activity of five key defense enzymes viz., Chitinase (CH), Catalase (CT), Poly Phenol Oxidase (PPO), Peroxidase (PO) and Phenylalanine Ammonia Lyase (PAL) were estimated from the stem tissues of young plants collected from the above treatments periodically viz., immediately after the spraying (0 day), and further upto 50 days at an interval of 10 days from application, when the enzyme activity became static or declines. Colorimetric assay of enzyme CH was carried out according to Bolter and Mauch (1988). PAL activity was estimated as described by Dickerson et al. (1984). The enzyme PO was analysed as given by Hammerschmidt et al. (1982), CT according to Aebi (1983) and PPO according to Meyer et al. (2000). To study the activity of the antioxidant enzymes like superoxide dismutase (SOD), Glutathione Reductase (GR) and Glutathione Peroxidase (GPX) both in chickpea and *C. campestris*, the samples were drawn from the above experiments and analyzed. The SOD activity was estimated using xanthine-xanthine oxidase system as suggested by Beyer and Fridovich (1987). The enzyme GPX was assayed as per the method suggested by Inoue et al. (1999).

**Statistical analysis:** All the experiments were conducted in Randomized Block Design (RBD) for two consecutive years and since there were no significant interactions between observations, the data were combined over the years and subjected to analysis of variance (ANOVA). Regression analysis was used where appropriate: Otherwise means were separated using least significant difference (LSD) at 5% level of significance. Before the analysis,
normality of data and the equality of variances were checked using Kolmogrov-Smirnov test and some variables were transformed using suitable transformation. ANOVA was performed on data using general linear models procedure using PROCANOVA procedure with the SAS 9.2 statistical software (SAS Institute Inc., USA). Significant differences between different treatments were observed using Tukey’s Honest Significant Difference. Linear model was best fitted to the flowering in *Cuscuta* at different distance from host plant chickpea. The model is given as $Y= a+bx$, where, $a$ and $b$ are the regression coefficients of the model and $y$ and $x$ represents the flowering in *Cuscuta* and distance of *Cuscuta* from the chickpea, respectively.

**RESULTS AND DISCUSSION**

Germination, host search and development of *C. campestris*: From the above study, it was observed that *C. campestris* germinated within a period of 3-4 days after sowing without acid scarification, when the fresh seeds were used. Germination of *C. campestris* was not influenced by the distance of its seed to the host seedling (Table 1) and there was no significant difference in the percentage germination of *C. campestris* when sown at different distances from the host plant. However treatment of chickpea with the bioagents or the synthetic elicitors influenced the germination of the *C. campestris* seeds and also affects the number of days taken by *C. campestris* to establish in the host and initiate flowering (Table 2, Fig. 1). Bion, when applied as seed treatments caused maximum delay in establishment of *C. campestris* by 16.4 days (30.50% over control) when sown at 12 cm away from the host, followed by salicylic acid 10.6 days (28.3% over control) when sown at 6 cm away. Among the bioagents, *P. fluorescens* was able to delay the process of establishment by 10.42 days (26.92% over control). However, when compared for the days taken to first flowering by *C. campestris*, which indicates the development and physiological maturity of the parasite, *P. fluorescens* was found to cause maximum delay of 25.20 days (29.36% over control).

| Treatments | *Cuscuta* infecting chickpea (DAS) | Flowering in *Cuscuta* (DAS) |
|------------|----------------------------------|-----------------------------|
|            | 3 cms   | 6 cms  | 9 cms  | 12 cms | 3 cms  | 6 cms | 9 cms | 12 cms |
| Seed treatment followed by foliar spray with *T. viride* at 20 DAS and 45 days | 6.80ab | 9.00bc | 12.40a | 13.20c | 20.40c | 22.00ab | 26.60c | 29.40b |
| Seed treatment followed by foliar spray with *P. fluorescens* at 20 and 45 DAS | 7.00ab | 10.40ab | 13.40a | 14.40bc | 25.20a | 23.00a | 28.80a | 31.00ab |
| Foliar spray with salicylic acid (0.05M) at 20 DAS and 45 DAS | 7.40a | 10.60a | 12.80a | 14.80b | 23.20b | 22.80a | 27.20bc | 29.80ab |
| Foliar spray with Bion (0.05M) at 20 DAS and 45 DAS | 8.00a | 10.80a | 13.40a | 16.40a | 24.00ab | 23.40a | 28.40ab | 31.20a |
| Chickpea + *C. campestris* (control) | 5.80b | 7.60c | 9.80b | 11.40d | 17.80d | 20.40b | 22.20d | 27.00c |
| LSD @0.05 | 1.20 | 1.40 | 1.38 | 1.40 | 1.39 | 1.85 | 1.33 | 1.78 |

**Table 1.** Effect of seed treatment and folia spray of bioagents and synthetic elicitors on germination and host search of *Cuscuta* in chickpea.

| Treatments | Coefficient estimates | $R^2$ |
|------------|-----------------------|-------|
|            | $a$(SE) | $b$(SE) |     |
| Seed treatment followed by foliar spray with *T. viride* at 20 DAS and 45 days | 16.70 (1.06) | 1.05 (0.13) | 0.97 |
| Seed treatment followed by foliar spray with *P. fluorescens* at 20 and 45 DAS | 20.10(0.54) | 0.92 (0.07) | 0.99 |
| Foliar spray with salicylic acid (0.05M) at 20 DAS and 45 DAS | 19.5(1.36) | 0.83(0.17) | 0.92 |
| Foliar spray with Bion (0.05M) at 20 DAS and 45 DAS | 19.80(1.41) | 0.93(0.172) | 0.94 |
| Chickpea + *C. campestris* (control) | 14.5(1.20) | 0.98(0.15) | 0.96 |

**Table 2.** Linear model fitting of data on flowering in *C. campestris*.
Table 3. Studies on the activity of five defence enzymes upon treatment with the bioagents and the synthetic elicitors.

| Enzyme                          | Days of observation after treatment |
|---------------------------------|-------------------------------------|
|                                 | Phenylalanine ammonia lyase (PAL)   |
|                                 | Chitinase (CHI)                      |
|                                 | Days 0 10 20 30 40 50              |
|                                 | Days 0 10 20 30 40 50              |
|                                 | LSD at 0.05                         |
|                                 |                                     |
|                                 | T1 2045.00f 2107.25f 2410.75f 2213.50f | 2188.00f 2478.25e 2249.00b 2305.75b |
|                                 | 2395.25b 2353.50b 2346.75b          |
|                                 | T2 2173.75e 2274.25e 2449.25e 2417.50e | 2407.25e 2477.25e 2211.00c 2249.00c |
|                                 | 2357.00c 2305.75b 2304.50c 2398.75c |
|                                 | T3 2242.50d 2312.50d 2515.50d 2455.00d | 2455.75d 2579.50d 2016.25d 2059.00d |
|                                 | 2230.50d 2102.50d 2091.50d 2261.25d |
|                                 | T4 2951.25b 3017.00b 3228.00b 3114.75b | 3011.25b 3247.50b 1848.25f 1902.75f |
|                                 | 2005.00f 1957.50f 1951.50f 2055.00f |
|                                 | T5 2648.75c 2710.00e 2943.75c 2850.75c | 2811.50c 2943.50c 1971.75e 2005.00e |
|                                 | 2102.50e 2051.00e 2044.25e 2155.00e |
|                                 | T6 3104.50a 3211.25a 3520.50a 3425.50a | 3404.00a 3542.75a 2351.75a 2395.25a |
|                                 | 2493.50a 2405.75a 2396.50a 2507.50a |
|                                 | T7 2046.00f 2008.25g 2214.25g 2125.50g | 2112.50g 2242.25f 1589.00g 1623.00g |
|                                 | 1747.00g 1690.75g 1652.00g 1777.50g |
|                                 | T8 1012.50g 1105.50h 1308.50h 1203.50h | 1206.25h 1334.75g 1051.50h 1093.00h |
|                                 | 1187.25h 1157.00h 1151.00h 1238.75h |
|                                 | LSD at 0.05 11.09 11.02 11.72 14.62 | 10.70 10.82 5.35 4.77 21.64 6.3 4.97 8.63 |
|                                 |                                     |
|                                 | Peroxidase (PO)                     |
|                                 | Poly phenol oxidase (POO)           |
|                                 | Catalase (CAT)                      |
|                                 | Days 0 10 20 30 40 50              |
|                                 | Days 0 10 20 30 40 50              |
|                                 | LSD at 0.05                         |
|                                 |                                     |
|                                 | T1 1.79d 2.10de 4.33bc 3.64b 2.80b 4.03c | 2.30c 3.24b 4.84a |
|                                 | 4.10a 4.14a 5.32b 0.39d 0.54c 0.74d |
|                                 | 0.64d 0.59d 0.82d                   |
|                                 | T2 1.34e 2.14de 3.69d 3.11d 2.84b 3.78d | 2.44b 3.16c 4.43b |
|                                 | 3.80b 3.71c 5.07e 0.21f 0.35d 0.54ef |
|                                 | 0.42e 0.39e 0.61f                   |
|                                 | T3 1.92e 2.27cd 4.15c 3.30c 3.10b 4.36b | 2.15d 2.76d 3.80c |
|                                 | 3.17c 3.11d 4.16e 0.65b 0.80b 1.28b |
|                                 | 1.28b 0.94b 0.84b 2.18f             |
|                                 | T4 2.16b 3.00a 4.17c 4.11a 4.02a 4.95a | 1.72f 2.44e 3.17e |
|                                 | 2.71d 3.12d 3.63f 0.59c 0.72b 0.88c |
|                                 | 0.80c 0.76c 0.93c                   |
|                                 | T5 1.75d 1.96e 4.15c 2.97d 3.89a 4.91a | 1.94e 2.30f 3.39d |
|                                 | 2.76d 2.71e 4.43d 0.34e 0.45cd 0.66de |
|                                 | 0.56d 0.57d 0.75de                  |
|                                 | T6 2.39a 2.54b 4.38b 3.33c 3.10b 4.53b | 2.71a 3.33a 4.84a |
|                                 | 4.11a 4.00b 5.53a 0.91a 1.65a 3.35a |
|                                 | 2.23a 2.15a 4.18a                   |
|                                 | T7 1.18f 2.39bc 4.71a 3.11d 3.09b 4.89a | 1.66g 2.15g 3.80c |
|                                 | 2.76d 2.59f 4.14e 0.23f 0.35d 0.53f |
|                                 | 0.39e 0.42e 0.64ef                  |
|                                 | T8 0.59g 0.68f 1.95e 1.04e 1.03c 2.05e | 0.83h 1.11h 1.95f |
|                                 | 1.74e 1.72g 2.29g 0.07g 0.11e 0.22g |
|                                 | 0.16f 0.14f 0.27g                   |
|                                 | LSD at 0.05 0.10 0.21 0.18 0.17 0.31 | 0.21 0.05 0.11 0.12 0.11 0.07 0.10 |
when sown at 3cm away from the host. This was followed by the treatment with bion (24 days and 25.83% over control). Linear model was best fitted to the flowering in *Cuscuta* at different distance from host plant chickpea. Results shows that initially, maximum delay in flowering occurs in treatment T2 (at 20.10 days) followed by treatment T4 (19.80 days) with slope 0.92 and 0.93 respectively. As the distance of the *Cuscuta* from the host plant increases, delays in flowering in *Cuscuta* also increases linearly. Understanding the process of their parasitization and development would lead to develop efficient strategies for their management (Westwood *et al.*, 2012). Contrast to the earlier reports about physical and physiological dormancy of *C. campestris* and about a high percentage of newly matured seeds of *C. campestris* not imbibing water to germinate readily (Hutchison and Ashton, 1980) and the need for acid scarification (Jayasuriya *et al.*, 2008), our studies have proved that fresh seeds, before drying in the plants germinates immediately without any need for scarification. This indicates that when sprinkler irrigation is given just before the harvest of the crop,
the matured seeds will be germinated and killed during subsequent harvest of the crop. Further irrigation prior to sowing the main crop, to optimum wetness would also result in the suicidal germination of the seeds of _C. campestris_. Further manual cleaning of the twines before they mature would result in the depletion of the parasitic weed seed bank in the soil.

Upon germination, green to yellow fine threads of _C. campestris_ grew randomly for a day or and on reaching chickpea, the twines coils around the aerial parts, mainly the stem and leaves, produce haustoria to penetrate the host tissue and vascular system to draw the nutrients and water. Delayed flowering as an effect of bioagents and synthetic elicitor seed treatment could be due to the release of volatiles by the host to deter/suppress the development of _C. campestris_. It is well established that _T. viride_ and _P. fluorescens_ application results in the overall development of systemic resistance in the host plants (ISR) (Van Loon et al., 1998).

**Systemic resistance induced by antagonists in chickpea:** Observation on the effect of the treatments of elicitors on _C. campestris_ and chickpea indicated that seed treatment followed by foliar sprays at 20 and 40 DAS was found to have positive effect on the growth and health of the plants.

Estimation of defense enzymes at an interval of 10 days for 50 days indicated the initial increase, reaching a peak and the decline of enzymes activity in the plants (Table 3). This trend shows that the induction is purely temporary and the induction potential of the microbes and the elicitors decreases after a certain period of time (Kannan and Jose, 2009). Repeated application of the bioagents or the elicitors could maintain an enhanced activity of the enzymes which is evident from the fact that the seeds treated plants, followed by foliar spray of the elicitors (treatments T4 to T6) had overall more activity of the enzyme when compared with the plants with only seed treatment (T1 to T3). Further the bioagents vary in their ability to induce different enzymes viz., _P. fluorescens_ was very effective in inducing all the enzymes except CH, while _T. viride_ was found to induce more of CH. However salicylic acid was most effective in inducing the enzymes than the microbes. These enzymes are key components of local and induced systemic resistance (Jankiewicz and Kołtonowicz, 2012). Though initially salicylic acid was better than microbes in inducing the defense enzymes, under natural conditions over a longer period of time the antagonistic microbes would build up their populations and induce the plants to produce more of the enzymes, which will not be the case with salicylic acid. Though BTH, a functional analogue of SA, has been reported as a successful resistance activator of plants (Oostendorp et al., 2001) in the current study it was found to suppress the initial growth of chickpea even at a very minimal dose.

The scatter plot matrix (Fig. 2) shows the relationship among five enzymes taken two at a time. Matrices reveal information like clusters and any outlier treatment among many treatments present in the data. In this plot, adjacent plots share common axis. It shows the eclipses which cover the maximum data points in it for different treatments. Those treatment values falling outside the eclipse shows significant difference with other treatment values. It also shows that in most of the comparisons, _T_6 outperforms all the treatments and _T_8 (control) have outliers and does not
perform well.

Time vs treatment interactions were studied for different enzymes and treatments using proc GLM procedure in SAS to know the significance of treatments on each point of time. Results indicated that the enzyme PAL had the highest activity in treatment T6 and the activity differed significantly for other treatments also. However, with respect to the enzyme PO, the treatments T4 to T7 showed significant variations at different points of time, but never showed a constant trend. Again the treatment T8 was found to be the best one for the enzymes CH and CT during the entire period of observation. All the enzymes except PPO showed significant interaction between treatment and time. PPO did show some significant changes between treatments at the early period of observations, but at later stages, the differences were non-significant. The activities of the antioxidant enzymes were estimated both in chickpea and C. campestris to analyze the effect of the treatments with the elicitors. It was observed GPX, GR and SOD were found to be maximum induced (102.36, 36.02 and 29.39 units mg⁻¹ protein min⁻¹, respectively) by the application of bion as compared to control (Fig. 3). It was also observed that the antioxidant enzymes were more active in C. campestris (24.08, 5.36 and 10.79 units mg⁻¹ protein min⁻¹, respectively) upon application of salicylic acid and the activation was significantly high when compared to treatment with the elicitors. In the case of GPX, P. fluorescens induced more activity of the enzyme (71.18 units mg⁻¹ protein min⁻¹) followed by salicylic acid (40.35 units mg⁻¹ protein min⁻¹), while these two treatments were at par in the case of GR and SOD. The biochemical activation and accumulation of defense enzymes, mainly the reactive oxygen scavengers, help in recovery of plants from the damage caused by the invasion of the parasite (Scandalios, 2005; Nyochemeng et al., 2007).

Conclusion

The above study showed that the fresh seeds of C. campestris germinate rapidly in a period of 5 to 6 days. This observation would help in suggesting that irrigation immediately before or after harvest of the crop, would induce the germination of Cuscuta seeds in soil and after germination, in the absence of the host would die, akin to the suicidal germination strategy followed for Orobanche and Striga with the use of germination stimulants. Application of T. viride and P. fluorescens elicited systemic resistance in chickpea, resulting in the increased production of defense enzymes, have better growth and suppresses growth of C. campestris. These microbes have already been established for their effective role in suppression of soil borne plant diseases and nematodes in chickpea. Thus the current study helps in emphasizing the application of these two microbes for better production of chickpea. Thus the overall results obtained in the current study gives a positive trend for the management of this dreaded weed in chickpea using the bioagents, which can be easily integrated with the existing management practices at minimal cost.

REFERENCES

Aebi, H. (1983). Catalase. In: Bergmeies H (ed) Methods of enzyme analysis. Chemie Verlag, pp 273–277. Beyer, W.F. and Fridovich, I. (1987). Assaying for superoxide dismutase activity: some large consequences of minor changes in conditions. Annals of Biochemistry, 161: 559–566. Boller, T. and Mauch, F. (1988). Colorimetric assay for chitinase. Methods in Enzymology, 161: 430 – 435. Dickerson, D.P., Pascholati, S.F., Hangerman, A.E., Butler, L.G. and Nicholson, R.L. (1984). Phenylalanine ammonia-lyase and hydroxyl cinnaminate: CoA ligase in maize mesocotyls inoculated with Helminthosporium maydis or Helminthosporium carcomum. Physiology and Molecular Plant Pathology, 25: 111-123. Elad, Y., Chet, I. and Henis, Y. (1981). A selective medium for improving quantitative isolation of Trichoderma spp. from soil. Phytoparasitica, 9: 59-67. Gaur, R.D. (1999). Flora of the District Garhwal. North West Himalaya, Transmedia, Srinagar Garhwal, pp: 443-444. Gaur, P.M., Tripathi, S., Gowda, C.L.L., Ranga Rao, G.V., Sharma, H.C., Pandey, S. and Sharma, M. (2010). Chickpea Seed Production Manual. Patancheru 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics. pp 228. Hammerschmidt, R., Nockles, E.M. and Kuc, J. (1982). Association of enhanced peroxidase activity with induced systemic resistance of cucumber to Colletotrichum lagenarium. Physiologia et Molecular Plant Pathology, 20: 73-82. Harsh, P., Bais, Tiffany L., Weir, Laura G., Perry, Simon Gilroy and Jorge M., Vivanco. (2006). The Role of Root Exudates in Rhizosphere Interactions with Plants and Other Organisms. Annual Review of Plant Biology, 57: 233–66. Holm, L., Doll, J., Holm, E., Panch, J. and Herberger, J. (1997). World Weeds: Natural Histories and Distribution. John Wiley & Sons, New York, pp 1129. Hutchison, J.M. and Ashton, F.M. (1980). Germination of field dodder (Cuscuta campestris). Weed Science, 28: 330-333. Inoue, Y., Matsuda, T., Sugiyama, K., Izawa, S. and Kimura, A. (1999). Genetic analysis of glutathione peroxidase in oxidative stress response of Saccharomyces cerevisiae. Journal of Biological Chemistry, 274: 27002–27009. Jankiewicz, U. and Kołtonowicz, M. (2012). The involvement of Pseudomonas bacteria in induced systemic resistance in plants. Applied Biochemistry and Microbiology, 48: 244-249. Jayasuriya, K.M.G.G., Baskin, J.M. and Baskin, C.C. (2008). Cycling of sensitivity to physical dormancy- break in seeds of Ipomoea lacunose (Convolvulaceae) and ecological significance. Annals of botany, 101: 341-352. Kannan, C. and Jose, C.T. (2009). Activation of defense enzymes in arecanut (Areca catechu L.) seedlings upon inoculation with biocontrol agents. Journal of Plantation Crops, 37 (2): 134-137. Kannan, C. and Karthik, M. (2009). Systemic induction of
defense enzymes by rhizosphere microbes in cocoa seedlings. Journal of Biological Control, 23(4): 427–431.

Lanini, W.T. and Kogan, M. (2005). Biology and management of Cuscuta in crops. Ciencia e investigación agrarian, 32: 165-179.

Meyer, A.M. (2006). Pathogenesis by fungi and by the parasitic plants: Similarities and differences. Phytoparasitica, 34: 3-16.

Meyer, U.M., Spotts, R.A. and Dewey, F.M. (2000). Detection and quantification of Botrytis cinera by ELISA in pear stems cold storage. Plant Disease, 84: 1099-1103.

Mishra, J.S., Moorthy, B.T.S., Bhan, M. and Yaduraju, N.T. (2007). Relative tolerance of rainy season crops to field dodder (Cuscuta campestris) and its management in niger (Guizotia abyssinica). Crop Protection, 26: 625-629.

Mishra, J.S. (2009). Biology and Management of Cuscuta species. Indian J Weed Sci., 41: 1-11.

Moorthy, B.T.S., Mishra, J.S. and Dubey, R.P. (2003). Certain investigations on parasitic weed Cuscuta in field crops. Indian Journal of Weed Science, 35: 214-216.

Mower, J.P., Stefanovic, S., Young, G.J. and Palmer, J.D. (2004). Gene transfer from parasitic Cuscuta to host plants. Nature, 432:165-166.

Nyochembeng, L.M., Beyl, C.A. and Pacumbaba, R.P. (2007). Peroxidase activity, isozymes pattern and electrolyte leakage in roots of Cocoyum infected with Pythium myaiotylum. Journal of Phytopathology, 155:454-461.

Oostendorp, M., Kunz, W., Dietrich, B. and Staub, T. (2001). Induced disease resistance in plants by chemicals. European Journal of Plant Pathology, 107: 19-28.

Scandalios, J.G. (2005). Oxidative stress: molecular perception and transduction of signals triggering antioxidant gene defenses. Brazilian Journal of Medical and Biological Research, 38(7): 995-1014.

Sriram, S., Manasa, S.B. and Savitha, M.J. (2009). Potential use of elicitors from Trichoderma in induced systemic resistance for the management of Phytophthora capsici in red pepper. Journal of Biological Control, 23(4): 449-456.

Van Looon, L.C., Bakker, P.A.H.M. and Pieterse, C.M.J. (1998). Systemic resistance induced by rhizosphere bacteria. Annual Review of Phytopathology, 36: 453-483.

Vyas, S.C. and Joshi, L.K. (1975). A new record of parasitic dodder on chickpea (Cicer arietinum L.). Current Science, 44: 701-702.

Westwood, J.H., dePamphilis, C.D., Das, M., Fernández-Aparicio, M., Honaas, L.A., Timko, M.P., Wafula, E.K., Wickett, N.J. and Yoder, J.I. (2012). The Parasitic Plant Genome Project: New Tools for Understanding the Biology of Orobanche and Striga. Weed Science, 60: 295–306.

Wright, M.A.R., Welsh, M. and Costea, M. (2011). Diversity and evolution of the gynoecium in Cuscuta (Convolvulaceae) in relation to their reproductive biology: two styles are better than one. Plant System and Environment, 296: 51–76.