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NON CHEMICAL METHODS TO CONTROL *OROBANCHE CERENATA* ON FABA BEAN PLANTS

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ABSTRACT

Pot experiments were designed to study the effect of three antagonistic fungi (*Trichoderma harizianum, T. koningii* and *Trichorus spirales*) and three organic amendments, German chamomile leaves (*Matricaria chamomilla*), garlic cloves (*Allium sativum*) and bakain leaves (*Millia azederach*) to control *Orbanche crenata* on faba bean plants. *T. harizianum* gave the best results in controlling *O. crenata.* It decreased the number and dry weight of *Orobanche* spikes. *T.kaningii* and *T. spirales* did not affect the growth of *O. crenata* on faba bean plants. Among the tested plants amendments garlic cloves gave the best result to control *O. crenata*. According to these results the authoress suggest that *T. harizianum* and garlic cloves could be used to minimize *O. crenata* plants on faba bean plants.

INTRODUCTION

Broomrapes are a holoparasite on faba bean plants causing serious losses in certain economic plants especially faba bean and tomato yield. The expensive chemical and physical measures are very effective for controlling *Orobanche* spp. The use of natural antagonistes of the parasite had been previously studied by (Al-Menoufi, 1986; Muehlchen *et al.,* 1990; Raju *et al.,* 1995 and Taslkh' Yan and Grigoryan, 1978).

Biological control should play a major role in reducing the incidence of prevailing weed species within an integrated pest management systems. There are several weed species causing serious losses in certain economic crops. One of these weeds is *Orobanche* sp. in vegetable, legumes and sun flowers in northern Africa and Middle east region (Abdel-Kader, *et al.,* 1996; Labrada, 1996 and Tsveta and Svetlana 1998).

Al-Menoufi, (1986) reported that *Fusarium oxysporium* Schlecht, *F. solani* (Mort) sacc., *Alternaria* sp. and *Sclerotinia* sp., which were previously isolated from rotted *O.crenata* fruits, significantly decreased the germination percentage of *O.crenata* seeds, as well as, the number of broomrape plants parasitizing on the host plants in the next season. The tested fungi were a virulent to certain economic winter crops.

Mazaheri *et al,I* (1991) isolate *F. oxysporum* from rotted shoots of *O.cernua* growing in tobacco field trials. The yield was increased by 85% as a result of the reduction of broomrape incidence. The fungus had no pathogenic effect on tobacco plants. In Egypt Abdel-Kader *et al,* (1998) found that *Alternaria, Fusarium* and *Trichoderma* decreased the number of emerged *O.crenata* without any harmfull effect on faba bean plants. These fungi were isolated as soil borne fungi from faba bean fields.

Organic amendments were used to control some fungal diseases and root knot nematode (Dubey and Dwivedi, 1991; Egunjobi and Afolami, 1976 and Muehlchen *et al,* 1990). The use of amendment to control

Orobanche spp. has not sufficiently studied. Ghosheh *et al,* (1999) used olive jift (a solid product from olive oil processing) to control *Orobanche* sp. in faba bean, pea and tomato plants. They found that jift in soil reduced broomrape germination and infection on all three hosts regardless of inoculation densities.

This work was done to study the antagonistic effect of *Trichoderma harizianum, T. koningii* and *Trichorus spirales* fungi and the organic amendments of German camomil leaves (*Matricaria chamomilla* L.), garlic cloves (*Allium sativum*) and bakain leaves (*Millia azederach*) to control *Orobanche crenata* on faba bean plants.

MATERIALS AND METHODS

A) Biological control

Three fungi were tested as biocontrol agents namely : *T.harizianum, T. koningii* (obtained from Plant Pathology Department, Faculty of Agriculture, Alexandria University) and *Trichorus spirales* (isolated from potato roots). Growing cultures were used. To prepare the inocula autoclaved bottles (500 ml) containing barley sand medium (75g barley and 25g clean sand and enough water to cover the barley sand mixture) were inoculated with discs (5 mm in diameter) of the tested fungi obtained from the marginal of the 7 days cultures growing on PDA at 28^{o}C. Bottles were incubated at 28^{o}C for two weeks. Sterilized plastic pots (30 cm in diameter) were filled with 2 kg autoclaved loamy sand. Cultures growing in the bottles were thoroughly mixed with the soil at the rate of 4% soil (w:w). Such infested pots were divided into two groups. One of these groups was inoculated with *O. crenata* seeds at the rate of 0.1 gm/pot, which were thoroughly mixed in the soil. The second set of pots was left without *Orobanche* seeds to check the effect of the tested fungi on faba bean plants. The infested soil were moisted every other day for one week to ensure even growth and distribution of the inoculated fungi. Soil mixed with sterilized barley sand medium at the same rate and infested, or not with *O. crenata* seeds were served as control.

Faba bean seeds (Giza 3 cv.) were surface sterilized with 1.0% Na-hypochlorite for ten min. Then thoroughly washed several times with sterilized tap water and sown at the rate of 5 seeds/pot. A set of 4 pots were used for each treatment.

After 90 days dry weight of faba bean plants, pods and *Orobanche* plants were determined. Number of faba bean pods and *Orobanche* plants growing on faba bean roots were also considered.

B) Organic amendments.

Dried German camomile leaves, garlic cloves and bakain leaves were used to study their effect on controlling broomrape (*O. crenata*). Sterilized plastic pots (30 cm in diameter) filled with 2 kg autoclaved loamy sand soil (1:1, w:w). Pots were inoculated with *O. crenata* seeds at the rate of 0.1 gm/pots, seeds were thoroughly mixed in the soil to insure the distribution of *Orobanche* seeds. Camomil plants, garlic cloves and bakain leaves were

air dried for 15 days then chopped and applied to soil at the rate of 40 gm/pot from each of the tested plants. Faba bean seeds (Giza 3 cv.) were sterilized and sown at the rate of 5 seeds/pot. A set 4 pots was used for each treatment. After 90 days dry weight of faba bean plants and their pods and *Orobanche* plants were determined. Number of faba bean pods and *Orobanche* plants growing on faba bean roots were also considered. All data were subjected to statistical analyses according to Snedecor and Cochran (1967).

RESULTS

Pot experiments were designed to study the antagonistic effect of *T. harizianum, T. koningii* and *Trichurus spiralis* as biocontrol agents to *O.crenata* on faba bean plants.

Dry weight of faba plants and their pods and *Orobanche* spikes, as well as, number of faba bean pods and *Orobanche* spikes were used as parameters to evaluate the efficiency of the tested fungi to control *O*. *crenata* on faba bean plants.

Data in Table (1) show that the tested fungi significantly decrease the number of *O.crenata* spikes. The number of *O.crenata* plants growing in pots infested with *T. harizianum* (4.66 plants/pot) was significantly less than those growing in pots infested with *T.koningii* or *T.spirales* and that growing in pots free from the tested fungi (10.0, 10.0 and 25 plants/pot respectively). All tested fungi did not affect the growth of faba bean plants.

Table (1) : Effect of three antagonistic fungi *T.harizianum, T.koningii* and *T.spirales* on the control of *O.crenata* on faba bean plants.

Treatments	Faba bean			*Orobanche*	
	Dry weight of plants (g)	Dry weight of pods (g)	Pods/ Number	Dry weight of spikes (g)	*Orobanche* number
T.harizianum	34.98[a]	5.68[a]	7.00[a]	0.00 (0.71)[e]	0.00 (0.71)[d]
T.harizianum + O.crenata	25.69[b]	3.09[b]	3.66[b]	3.87 (2.08)[d]	4.66 (2.26)[c]
T.koningii	26.68[b]	3.69[b]	3.33[b]	0.00 (0.71)[e]	0.00 (0.71)[d]
T.koningii + O.crenata	19.71[c]	1.76[c]	2.00[c]	5.38 (2.41)[c]	10.0 (3.22)[b]
T.spirales	27.46[b]	3.69[b]	4.00[b]	0.00 (0.71)[e]	0.00 (0.71)[d]
T.spirales + O.crenata	20.76[c]	1.61[c]	1.66[c]	7.35 (2.79)[b]	10.0 (3.22)[b]
O.crenata (control)	15.63[c]	0.50[d]	1.00[c]	7.49 (2.93)[a]	25.0 (4.93)[a]
Untreated (control)	31.76[a]	4.96[a]	4.30[b]	0.00 (0.71)[e]	0.00 (0.71)[d]
L.S.D	5.40	0.97	1.50	0.17	0.38

*** Values are the average of 4 replicates.**
**** Number between brackets are the angular transformation of data.**
***** Values in the same columns with the same letter are not significantly different.**

Table (2) : Effect of three plants as soil amendments, German camomile leaves, garlic bulbes and bakain leaves to control *O.crenata* on faba bean plants.

Treatments	Faba bean			*Orobanche*	
	Dry weight of plants (g)	Dry weight of pods (g)	Pods/ Number	Dry weight of spikes (g)	No. of *Orobanche* Spikes
German camomil	28.63^{ab}	3.40^{b}	4.33^{ab}	0.00 (0.71)^{c}	0.00 (0.71)^{e}
German camomil + *O.crenata*	21.28^{c}	1.53^{c}	1.96^{c}	3.97 (2.10)^{b}	7.33 (2.87)^{d}
Garlic bulbes	32.04^{a}	4.36^{a}	5.66^{a}	0.00 (0.71)^{c}	0.00 (0.71)^{e}
Garlic bulbes + *O.crenata*	27.46^{b}	3.23^{b}	3.66^{b}	3.49 (1.98)^{b}	3.66 (2.26)^{c}
Bakian leaves	30.45^{a}	2.86^{b}	3.66^{b}	0.00 (0.71)^{c}	0.00 (0.71)^{e}
Bakian leaves + *O.crenata*	23.77^{bc}	1.56^{c}	1.96^{c}	3.78 (2.04)^{b}	9.33 (3.12)^{b}
O.crenata (control)	16.63^{d}	0.50^{d}	1.00^{d}	8.13 (2.93)^{a}	21.33 (4.66)^{a}
Untreated (control)	29.63^{a}	5.01^{a}	4.30^{b}	0.00 (0.71)^{c}	0.00 (0.71)^{e}
L.S.D	4.30	0.93	0.94	0.35	

*** Values are the average of 4 replicates.**
**** Number between brackets are the angular transformation of data.**
***** Values in the same columns with the same letter are not significantly different.**

Dry weight of the growing *O.crenata* plants was also reduced as a result of fungal treatments. The dry weight of *O.crenata* plants growing in pots infested with *T.harizianum* (3.87g) was significantly less than these growing in pots infested with *T.koningii* or *T.spirales* (5.38 and 7.35 g respectively). These are significantly less than dry weight of plants growing in pots free from the tested fungi (7.49 g).

Concerning faba bean plants and their pods, data in Table (1) show that the growth of faba bean plants was improved in the presence of any of the tested fungi irrespective to the presence of *O.crenata*. Pods showed the same trend. Number of pods from pots infested only with *T.harizianum* and their dry weight were significantly higher than these obtained from any other treatment. Differences due to *T.koningii* and *T.spirales* were not significant in all parameters except the dry weight of *Orobanche* spikes. The dry weight of spikes growing in pots infested with *Trichorus spirales* was significantly higher than these of spikes growing in pots infested with *T.koningii* and *T. harizianum* 17.35, 5.38, 3.87 g respectively.

B- Organic amendments :

Pot experiment was designed to study the effect of three plants as organic amendments (German camomile leaves, garlic bulbes and bakian leaves) on the growth of *O.crenata* parasitizing faba bean plants.

Data presented in Table (2) revealed that all tested plants significantly decreased the number of *O. crenata* spikes and their dry weight. Numbers of *O.crenata* spikes developed in pots treated with garlic bulbes was significantly less than these developed in pots treated with German

camomile and bakian leaves (3.66, 7.25 and 9.33 respectively). Number of *Orobanche* spikes developed in infested control (21.33) was significantly higher than these developing in any other all tested treatments. However the dry weight of the developed *O.crenata* spikes was significantly decreased as a result of soil treatment with the tested plants.

Data in Table (2) also revealed that the dry weight of faba bean plants and number of pods and their dry weight were significantly increased when faba bean plants grow in pots treated with garlic cloves comparing with these obtained from pots treated with the other tested plants.

DISCUSSION

Biological control is an important suggested way to control broomrape. All previous work had been done by using fungi isolated from different parts of *Orobanche* plants. The isolated fungi were tested *in vitro* and *in vivo* for their ability to affect *Orobanche* plants and their hosts (Al-Menoufi, 1986; Abd El-Kader *et al.,* 1996; Bedi and Donchev 1991 and Talsakh Yan and Gigorgan 1978).

Results obtained in the present work showed that *T.horizianum* significantly decreased the number and dry weight of *O.crenata*, while *T.koningii* and *T.spirales* had no retarding effect on the growth of *O.crenata* comparing with the control treatment. These results are in agreement with those reported by Abd El-Kader *etal.,* (1998). They reported that *Fusarium, Alternaria* and *Trichoderma* fungi isolated from Egyptian soil infested with *O. crenata* gave promising effect in controlling *Orobanche* sp. Bedi and Sauerborn (1998) found that an isolate of *Fusarium oxysporium* f. sp. *Orthoceras* was very effective in controlling *O.cumana* on sunflower. Their results showed that chlamydospores were the most suitable structure of *F.oxysporium* f. sp. *orthoceras* for the development of mycoherbicide.

The effect of organic amendments were used to control some fungal diseases and root knot nematode. The use of amendments to control *Orobanche* spp. has been intensively studied (Egunjobi and Afolami 1976 and Muehlchen *et al,* 1990).

Ghosheh *et al.,* (1999) they used olive jift to control *Orobanche* sp. in faba bean, pea and tomato plants. They found that jift in soil reduced broomrape germination and infection on the three hosts regardless of inoculation densities. Fresh and dry weight of all crops were not adversely affected by mixing jift with soil. They suggest a possible use of jift as an in expensive organic material for brommrape control.

The results in the present work showed that the three tested plant materials which used as organic amendments, German camomile leaves, garlic bulbes and bakian leaves reduced the number and dry weight of *O. crenata* developed on faba bean plants. The dry weight of faba bean plants was not adversely affected by mixing organic amendments with soil. The best results was that when garlic bulbes was used as organic amendments. Dubey and Dwivedi (1991) found that extracts of leaves and bulbes of onion, garlic and fruit and bark of *Allium arabic* inhibited the mycelial growth of

Macrophomina phaseolina in *in vivo* test and the extract of garlic bulbes was more effective than the others. They also reported that extracts of these plants gave promising results in controlling such pathogen infect soybean plants.

Results of the present work indicated that *T.harizianum* could be effective in controlling *O.crenata* on faba bean plants. Accordingly the authoress could be suggested that the use of such fungus as bioherbicide might reduce the infection with this noxious parasitic weed. For wide application in different locations, more studies are needed. Also the use of amendments to control parasitic weed needs more studies.

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المقاومة الغير كيميائية للهالوك المتطفل على نباتات الفول

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أجريت تجربة أصص استخدم فيها ثلاثة فطريات لدراسة تأثيرها الحيوى على مقاومة الهالوك المتطفل على نباتات الفول وكانت هذه الفطريات هي (تريكودرما هاريزيانم، تريكودرما كوننجيى وتريكورس سبيرالس) كذلك استخدم أجزاء من ثلاثة نباتات كمواد عضوية تضاف للتربة وهى أوراق الشيح البابونج، بصيلات الثوم واوراق نبات الزنزلخت بنيت النتائج أن الفطر تريكودرما هاريزيانم أعطى أفضل النتائج في مقاومة الهالوك. حيث قلل أعداد والوزن الجاف لنباتات الهالوك المتطفل على نباتات الفول وبالتالى ككل من وزنها الجاف. بينما لم يكن للفطران الآخران (تريكودرما كوننجيى وتريكورس سبيرالس) أي تأثير في مقاومة الهالوك.

بالنسبة للنباتات المستخدمة فقد أعطت فصوص الثوم افضل النتائج في مقاومة الهالوك. بينما كان تأثير أوراق كل من الشيح البابونج والزنزلخت في تثبيط نمو الهالوك اقل من تأثير الثوم.

تبعا لهذه النتائج يمكن الاقتراح بأنه يمكن إستخدام فطر تريكودرما هاريزيانم ونباتات الثوم في تقليل إصابة الفول بالهالوك.