Laboratory and Field Studies of *Trichoderma harzianum*, Bacterial Strains and Imazethapyr on *Orobanche crenata* Forsk Infesting *Vicia faba*

Mahdi A Yahia[1], Mohammed M Hassan[2,*], Muntasir A M Elamien[1], Nasreldin K Abdalla[3], Ahmed M E Rugheim[1], Randa H Elsalahi[2], Rashida R M Abusin[3], Rania A Abakeer[2], Magdoline M Ahmed[2], Awad G Osman[2], Migdam E Abdelgani[2], AddelGabar E Babiker[2]

[1] Faculty of Agriculture, Omdurman Islamic University
Omdurman, Sudan

[2] Environment, Natural Resources and Desertification Research Institute, the National Center for Research, Khartoum, Sudan.
[3] Shendi Research Station, Agriculture Research Corporation
Shendi, Sudan

[4] Pests and Plant Health, College of Agriculture, Bahri University
Khartoum, Sudan.

[*]*Corresponding author's email: mohkadis [AT] yahoo.com*

ABSTRACT— ***A series of laboratory and field experiments were undertaken at the laboratories and experimental farm of Environment, Natural Resources and Desertification Research Institute, NCR and Shendi Research Station experimental farm, ARC, Sudan at season 2015-2016, to evaluate efficacy of Trichoderma harzianum, Bacillus megatherium var. Phosphaticum (BMP), Rhizobium leguminosarum (TAL1399) and the herbicide imazethapyr (pursuit) against Orobanche crenata infesting faba bean. Treatments were laid out in a Complete Randomized Design (CRD) in laboratory experiment and in a Randomized Complete Block Design (RCBD) in the field experiments with four replicates. Results of laboratory experiments showed that T. harzianum and Imazethapyr each alone or in combination significantly reduced O. crenata germination. Field results revealed that, application of T. harzianum, Imazethapyr and Imazethapyr + BMP+TAL1399 significantly delayed the days of O. crenata emergence in Shendi and Soba sites. T. harzianum alone or in combination with bacteria or imazethapyr significantly reduced number of O. crenata emergence and increased faba bean plant height as compared to the corresponding control in Shendi and Soba sites. T. harzanium alone or in combination with T. harzanium + BMP+TAL1399 significantly increased faba bean biomass, pod/plant and grain yield and 100 seed weight insignificantly as compared to the infested control in Shendi and Soba sites.***

Keywrods---- Bacteria, Broomrape, Faba bean, Pursuit, *T. harzianum*

1. INTRODUCTION

Annual grain and forage legumes such as faba bean (*Vicia faba* L.), vetches (*Vicia* spp.), lentil (*Lens culinaris* Medik.), pea (*Pisum sativum* L.), grass pea (*Lathyrus sativus* L.) and chickling vetch (*Lathyrus cicera* L.) are important crops grown worldwide as a source of protein both for human food and animal feed. However, their cultivation is strongly hampered in Mediterranean and Middle East farming systems by the occurrence of broomrape causing important yield losses [1, 2, 3]. In Sudan faba bean is the most important cool season food legume. It is the major source of protein for a major sector of the population, particularly in urban areas. Faba bean yields about 70 thousand metric tons of grains annually and thus constitute about 70% of the country's needs [4].

Productivity and yield stability of cool season legumes are constrained by many biotic and abiotic factors. Among the biotic factors, parasitism by broomrapes (*Orobanche* spp) is by far the most important in West Asia, North Africa and Southern Europe [5]. In faba bean, losses from 5 to 95% have been reported depending on the infestation level and the planting date [6].

A number of strategies of root-parasitic weed control have been developed including cultural practices and biological and chemical control [2, 7, 8]. The integration of several control measures seems to be the most desirable strategy. The only way to cope with the weedy root parasites is through an integrated approach, employing a variety of measures in a concerted manner, starting with containment and sanitation, direct and indirect measures to prevent damage caused by the parasites, and finally eradicating the parasite seed bank in soil [9].

Only moderate to low levels of incomplete resistance of complex inheritance against *O. crenata* have been identified in legumes [1, 10, 11] making selection more difficult and slowing down the breeding process. Resistance to broomrape appears to have multiple components and to be based on a chain of escape and resistance mechanisms that either act alone or in combination and at different stages of the infection process [12].

Inoculation of the soil or the host plants with beneficial soil microorganisms which reduce the parasites success at early developmental stages is advantageous as it may hamper the growth of the parasite and curtails its deleterious effects on hosts [13]. Early developmental stages of *Striga*, *Orobanche*, and *Phelipanche* are especially fragile, modulated by phytohormones and are very likely targets for control methods [14]. Laboratory and field experiments were conducted to evaluate the potentials of several treatments as suppressors of early developmental stages of *O.crenata*. The treatments include i) the herbicide imazethapyr (pursuit), bacterial strains ii) *T. harzianum*, each alone and their combinations.

2. MATERIALS AND METHODS

The laboratory experiment was conducted at the Bio-pesticides and Bio-fertilizers Department, Environment, Natural Resources and Desertification Research Institute (ENDRI), National Centre for Research (NCR), Khartoum, Sudan. The field experiments were conducted at two sites Shendi Research Station, Agricultural Research Corporation (ARC) at River Nile State and at ENDRI experimental farm in Soba in the south of Khartoum State, at season 2015-2016, to study the effects of *Trichoderma harzianum* (T.h), *Bacillus megatherium* var. *phosphaticum* (BMP), *Rhizobium leguminosarum* strain (TAL1399) and the herbicide imazethapyr on *O. crenata* germination, incidence, faba bean growth and yield.

The strigolactone analogue synthetic stimulant (GR24) was provided by professor Zwanenberg, University of Nimijhen, the Netherlands. A stock (10 ppm) of GR24 was prepared by dissolving 1 mg in 1 ml acetone and completing to volume (100 ml) with sterile distilled water. The solution was kept refrigerated at 4ºC for further use.

The herbicide imazethapyr, *O. crenata* and faba bean seeds (cv; BB7) were obtained from the (ARC), Shendi Research Station. BMP and TAL1399 were obtained from the Biofertilization Department, Environment, Natural Resources and Desertification Research Institute (ENDRI), the National Centre for Research, Khartoum, Sudan. The *T. harzianum* was obtained from the microbial collection of the Faculty of Agriculture, Omdurman Islamic University.

O. crenata seeds were cleaned by placement in a measuring cylinder (1000 ml) containing tap water with a few drops of liquid soap. Floating materials containing derbies and immature light seeds were discarded, then seeds were surface disinfected by soaking in 70% ethanol for 3 minutes, with continuous agitation and rinsed three times in distilled water subsequently, the seeds were immersed in 1% sodium hypochlorite for 2 minutes and rinsed three times in sterilized distilled water. The seeds were plotted dry on Whatman filter paper under a Laminar flow hood, then were kept in sterilized glass vial at 10ºC for further studies. Glass fiber filter paper (GF/C) discs (8 mm diameter) were cut, wetted thoroughly with water and placed in an oven at 100ºC for I h to be dried, the discs, placed in 9 cm diameter Petri dishes lined with glass fiber filter paper (GF/C), were moistened with 5 ml distilled water, About 25-50 surface disinfected *O. crenata* seeds were sprinkled on each of the glass fiber discs in each Petri dish, the dishes, sealed with parafilm were placed in black polythene bags and incubated at 18ºC±2, for 11 days.

2.1 Laboratory experiment

Effects of T. harzianum, BMP, TAL1399 and imazethapyr on O. crenata germination

The discs containing *O. crenata* seeds conditioned in distilled water dapped on normal filter paper to remove excess water and then transferred to sterile Petri dishes. Each disc was treated with distilled water, diluted media, *T. harzianum*, BMP, TAL1399 or imazethapyr. The dishes sealed with parafilm, were placed in black polythene bags and incubated at 18ºC±2 for 3 days. Then treated with distilled water or GR24 at 30 ml of GR24 (10 ppm) concentration, the treated seeds in Petri-dishes were sealed with parafilm and wrapped in aluminum foil, then incubated in the dark at 18ºC±2 for 7 days prior to measurement of germination and radicle extension, using a stereomicroscope.

2.2 Field experiment

Effects of T. harzianum, BMP, TAL1399 and imazethapyr on O. crenata incidence, faba bean growth and yield

The field was disc ploughed, harrowed, leveled, ridged and divided into sub-plots (2.5 x 3m). All sub-plots, excluding those used as non-infested control, were artificially infested with *Orobanche* seeds by adding 1g of clean *Orobanche* seeds to1kg of soil, previously sieved through a 2 mm mesh metal screen, followed by thorough mixing, then applied to the soil (5 mg *Orobanche* seeds hole^{-1}) before sowing faba bean (cv: BB7).

Faba bean seeds were coated with BMP and TAL 1399 carried on charcoal when applicable. Five gram of *T. harzianum* inoculum carried on rice was added in each hole at sowing where applicable. The imazethapyr at rate of 47.6g/ha was applied at sowing where applicable.

Weeds other than *O. crenata* were removed 2 times at biweekly intervals, starting 2 weeks after crop emergence, using a hand hoe.

Treatments were laid out in a Randomized Complete Block Design (RCBD) with four replicates. Data collected for faba bean growth attributes, included: i) Plant height. ii) Plant biomass. iii) Number of pods. iv) 100-seed weight. v) Grain yield (kg ha^{-1}). Data for *O. crenata* included: i) Day of *O. crenata* emergence. ii) Number of *O. crenata* shoots per m^2.

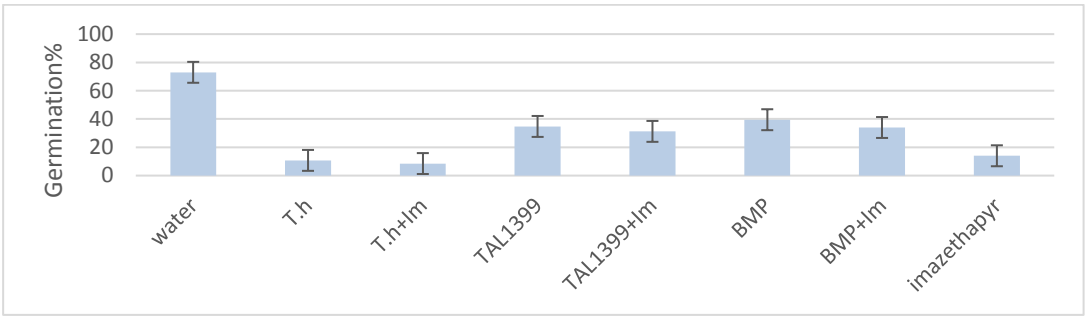
2.3 Statistical analysis

Data collected from laboratory and field experiments were subjected to statistical analysis (Analysis of Variance (ANOVA)), using SPSS 22 statistical package and means were separated for significance using the Duncan Multiple Range Test (DMRT) at $P \leq 0.05$.

3. RESULTS

3.1 Laboratory experiment

Results in figure (1) showed that application of GR24 at 10ppm to the seeds conditioned in water induced germination by 73%. Application of *T. harzianum* + imazethapyr gave the highest significant (P≤0.05) inhibition on seed germination of broomrape during conditioning by 88.36% followed by ***T. harzianum*** 85.27% and imazethapyr 80.82% as compared to the control.



T.h = *T. harzanium*; Im = imazethapyr

Fig. 1. Effects of Treatments on *O. crenata* Germination

3.2 Field experiment

3.2.1 Effect of treatments on plant height

Shendi site

The growth of faba bean represented by plant height increased with all the treatments compared to the infected control at the 65, 80 and 95 days after sowing (DAS) (Table 1). At 65 DAS, imazethapyr alone and imazethapyr + BMP + TAL1399 gave the highest plant height (92cm). At 80 and 95 DAS, the combination of BMP+TAL1399 + imazethapyr significantly (P≤0.05) increased plant height as compared to the infested control.

Soba site

From the results of Soba site, application of *T. harzianum* alone, in combination with BMP+TAL1399 and with imazethapyr significantly (P≤0.05) increased plant height at 80 DAS as compared to the control (Table 2). At 95 DAS, *T. harzianum* followed by the combination of *T. harzianum* + BMP+TAL1399 + imazethapyr and *T. harzianum* + imazethapyr significantly (P≤0.05) increased plant height as compared to the control.

Table (1) Effects of Treatments on Faba bean Height in Shendi Site

Fungi	Bacteria	Herbicide	Plant height (cm)			Mean
			65DAS	80 DAS	95DAS	
Without	Without	Without	79.93	83.40^{b}	85.00^{b}	**82.78**
		imazethapyr	92.15	95.27^{ab}	95.93^{ab}	**94.45**
	BMP + TAL1399	Without	80.53	88.20^{ab}	88.80^{ab}	**85.84**
		imazethapyr	92.13	101.33^{a}	104.67^{a}	**99.38**
T. harzianum	Without	Without	89.13	94.60^{ab}	98.60^{ab}	**94.11**
		imazethapyr	83.80	93.33^{ab}	99.40^{ab}	**92.18**
	BMP + TAL1399	Without	81.06	90.33^{ab}	94.53^{ab}	**88.64**
		imazethapyr	85.53	90.67^{ab}	92.53^{ab}	**89.58**
Un-infested control			91.00	96.27^{ab}	97.53^{ab}	**94.93**
P≤ 0.05			ns	*	*	
SE±			1.441	1.3723	1.4166	
SD±			8.013	8.320	8.4998	

Means followed by the same letter(s) are not significantly different according to DMRT at P≤0.05.

Table (2) Effects of Treatments on Faba bean Height in Soba Site

Fungi	Bacteria	Herbicide	Plant height (cm)			Mean
			65DAS	80 DAS	95DAS	
Without	Without	Without	44.40	49.35^{b}	53.87^{b}	**49.21**
		imazethapyr	50.71	53.52^{ab}	65.52^{ab}	**56.58**
	BMP + TAL1399	Without	52.75	57.10^{ab}	66.45^{ab}	**58.77**
		imazethapyr	52.85	57.45^{ab}	67.48^{ab}	**59.26**
T. harzianum	Without	Without	55.06	64.41^{a}	74.61^{a}	**64.69**
		imazethapyr	57.15	60.52^{a}	71.58^{a}	**63.08**
	BMP + TAL1399	Without	57.70	63.52^{a}	68.62^{ab}	**63.28**
		imazethapyr	55.20	58.30^{ab}	73.45^{a}	**62.32**
Un-infested control			53.30	58.00^{ab}	68.56^{ab}	**59.95**
P≤ 0.05			ns	*	*	
SE±			1.6534	1.2339	1.2627	
SD±			8.6838	8.5487	8.7569	

Means followed by the same letter(s) are not significantly different according to DMRT at P≤0.05.

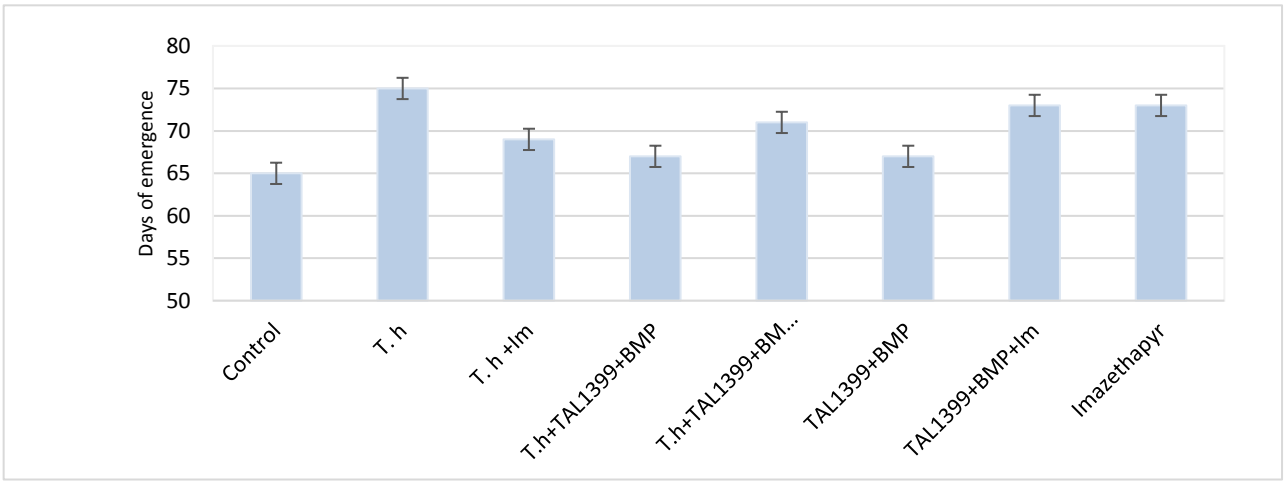
3.2.2 Effect of treatments on days of O. crenata emergence

Shendi site

From the results presented in Fig. 2, application of *T. harzanium*, imazethapyr each alone and imazethapyr + BMP+TAL1399 significantly (P≤0.05) delayed the time of *O. crenata* emergence as compared to control (Fig. 2).

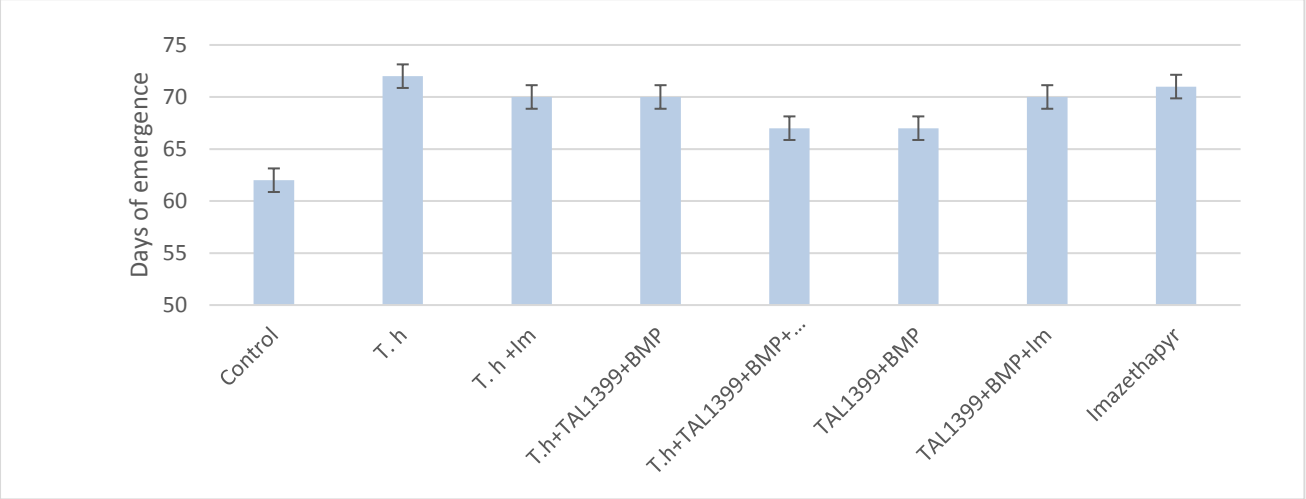
Soba site

Results of Soba site showed that *T. harzanium* and imazethapyr each alone significantly (P≤0.05) delayed the time of *O. crenata* emergence as compared to control (Fig. 3).



T.h = *T. harzanium*; Im = imazethapyr

Fig.2 Effects of Treatments on *O. crenata* Days of Emergence in Shendi Site



T.h = *T. harzanium*; Im = imazethapyr

Fig. 3 Effects of Treatments on *O. crenata* Days of Emergence in Soba Site

3.2.3 Effect of treatments on number of O. crenata emergence

Shendi site

Results of Shendi field experiment showed that *T. harzanium* applied alone significantly (P≤0.05) delayed the number of *O. crenata* emergence at 75, 90 and 105 DAS compared to the control treatment (Table 3). While the application of *T. harzanium* + BMP+TAL1399 significantly (P≤0.05) inhibited *O. crenata* emergence at 105 DAS as compared to the control.

Soba site

From the results of Soba Site, the application of *T. harzanium* + BMP+TAL1399 + imazethapyr significantly (P≤0.05) delayed the number of *O. crenata* emergence at 75, 90 and 105 DAS as compared to the control (Table 4). *T. harzanium* + imazethapyr combination TAL1399 significantly (P≤0.05) inhibited *O. crenata* emergence at 75 and 90 DAS as compared to the control.

Table (3) Effects of Treatments on Number of *O. crenata* in Shendi Site

Fungi	Bacteria	Herbicide	No of O. *crenata* /m^2			Mean
			75 DAS	90 DAS	105 DAS	
Without	Without	Without	1.36^{e} (23.00)	1.53^{e} (36.25)	1.88^{e} (76.50)	**45.25**
		imazethapyr	0.66^{b} (5.25)	1.16^{bcd} (17.50)	1.47^{abcd} (30.75)	**17.83**
	BMP + TAL199	Without	1.1^{d} (14.50)	1.35^{de} (23.75)	1.57^{bcd} (38.50)	**25.58**
		imazethapyr	0.93^{cd} (10.00)	1.14^{bcd} (15.00)	1.65^{de} (45.25)	**23.42**
T. harzianum	Without	Without	0.35^{a} (2.25)	0.74^{a} (5.75)	1.20^{a} (18.25)	**8.75**
		imazethapyr	0.71^{bc} (6.00)	1.01^{abcd} (11.50)	1.31^{ab} (20.25)	**12.58**
	BMP + TAL199	Without	0.54^{ab} (4.25)	0.83^{ab} (7.75)	1.22^{a} (17.25)	**9.75**
		imazethapyr	0.68^{bc} (5.00)	0.98^{abc} (9.75)	1.38^{abc} (24.00)	**12.92**
p≤ 0.05			**	**	**	
SE±			0.05201	0.06423	0.03716	
SD±			0.34273	0.29270	0.24646	

Values without brackets () indicate logarithmic transformed data. Means followed by the same letter (s) are not significantly different according to DMRT at P≤ 0.05

Table (4) Effects of Treatments on Number of *O. crenata* in Soba Site

Fungi	Bacteria	Herbicide	Number of O. *crenata* /m^2			Mean
			75 DAS	90 DAS	105 DAS	
Without	Without	Without	1.65^{c} (45.25)	1.82^{c} (67.67)	1.98^{c} (100.75)	**71.22**
		imazethapyr	0.78^{ab} (15.00)	1.16^{ab} (25.25)	1.72^{bc} (58.5)	**32.92**
	BMP + TAL199	Without	1.38^{b} (28.00)	1.65^{b} (46.75)	1.85^{bc} (71.5)	**48.75**
		imazethapyr	0.44^{a} (4.75)	1.18^{ab} (16.5)	1.36^{ab} (25.75)	**15.67**
T. harzianum	Without	Without	0.45^{a} (5.5)	1.13^{ab} (15.75)	1.38^{ab} (25.75)	**15.67**
		imazethapyr	0.37^{a} (3.25)	0.82^{a} (10.00)	1.39^{ab} (29.5)	**14.25**
	BMP + TAL199	Without	0.79^{ab} (9.25)	1.20^{ab} (17.75)	1.47^{bc} (30.5)	**19.17**
		imazethapyr	0.36^{a} (3.25)	0.79^{a} (12.00)	0.97^{a} (24.75)	**13.33**
p≤ 0.05			**	**	**	
SE±			0.08466	0.06529	0.05440	
SD±			0.57596	0.42116	0.36730	

Values without brackets () indicate logarithmic transformed data. Means followed by the same letter (s) are not significantly different according to DMRT at P≤ 0.05

3.2.4 Effect of treatments on faba bean growth and yield

Shendi site

The results revealed that in Shendi experiment, application of imazethapyr + BMP+TAL1399 and *T. harzanium* + BMP+TAL1399 significantly (P≤0.05) increased faba bean biomass in comparison to the control. The combination of *T. harzanium* + imazethapyr followed by *T. harzanium* significantly (P≤0.05) increased the number of pods per plant and the grain yield Kg/feddan (feddan=4200m^2) as compared to infested control (Table 5). Application of the combinations *T. harzanium* + BMP+TAL1399 and imazethapyr + BMP+TAL1399 increased the weight of 100 seed insignificantly as compared to the infested control.

Soba site

Results of Soba field experiment, showed that *T. harzanium* + BMP+TAL1399 significantly (P≤0.05) increased faba bean biomass as compared to the control. The combination of *T. harzanium* + BMP+TAL1399 followed by *T. harzanium* + BMP+TAL1399 + imazethapyr and *T. harzanium* alone significantly (P≤0.05) increased the number of pods per plant and the grain yield Kg/feddan (feddan=4200m^2) as compared to infested control (Table 6). Application of *T. harzanium* significantly (P≤0.05) increased the weight of 100 seed as compared to infested control.

Table (5) Effects of Treatments on Faba bean Growth and Yield in Shendi Site

Fungi	Bacteria	Herbicide	Biomass (g)	Pods / plant	100-seed weight (g)	Yield (Kg/fed.)
Without	Without	Without	268.33^{b}	26.33^{b}	40.43	513.67^{g}
		imazethapyr	368.33^{ab}	28.33 ^{b}	40.46	760.06^{cdef}
	BMP + TAL199	Without	291.67^{b}	25.66^{b}	37.63	573.78^{fg}
		imazethapyr	598.33^{a}	35.66^{ab}	41.13	890.43^{bcd}
T. harzianum	Without	Without	470.00^{ab}	49.33^{a}	39.63	1080.44^{ab}
		imazethapyr	423.33^{ab}	35.33^{ab}	36.70	820.24^{cde}
	BMP + TAL199	Without	571.67^{a}	52.00^{a}	41.23	1150.84^{a}
		imazethapyr	418.33^{ab}	43.00^{ab}	39.57	913.27^{bc}
Un-infested control			491.67^{ab}	48.00^{a}	41.33	1100.45^{ab}
p≤ 0.05			*	*	Ns	**
SE±			28.9244	2.2133	.59154	37.572
SD±			150.2955	12.021	3.601	224.37

Means followed by the same letter (s) are not significantly different according to DMRT at P≤ 0.0.

Table (6) Effects of Treatments on Faba bean Growth and Yield in Soba Site

Fungi	Bacteria	Herbicide	Biomass (g)	Pod / plant	100-seed weight (g)	Yield (Kg/fed.)
Without	Without	Without	58.66^{b}	11.00^{f}	41.02^{c}	314.07^{e}
		imazethapyr	77.66^{ab}	22.00^{bcdef}	46.19^{bc}	528.27^{cd}
	BMP + TAL199	Without	80.66^{ab}	21.00^{bcdef}	44.56^{c}	496.43^{d}
		imazethapyr	88.66^{ab}	15.90^{def}	47.99^{abc}	374.82^{e}
T. harzianum	Without	Without	83.33^{ab}	25.90^{bcde}	55.75^{a}	624.45^{ab}
		imazethapyr	90.33^{ab}	18.75^{cdef}	48.46^{abc}	481.44^{d}
	BMP + TAL199	Without	103.66^{a}	29.50^{ab}	50.22^{abc}	669.16^{a}
		imazethapyr	89.33^{ab}	26.87^{abc}	48.65^{abc}	597.15^{abc}
Un-infested control			95.66^{ab}	21.00^{bcdef}	491.67	583.33^{bc}
p≤ 0.05			*	***	**	**
SE±			3.9725	1.3445	0.7959	17.8183
SD±			20.1219	8.9323	5.0386	123.449

Means followed by the same letter (s) are not significantly different according to DMRT at P≤ 0.0.

4. DISCUSSION

Root-parasitic weeds of the genus *Orobanche* are noxious invasive weeds. They are economically important as they reduce crop yield and quality and present a serious threat to food security in many areas across the world (Parker and Riches, 1993). The parasites remove water, minerals and photosynthate from the host and thus reduce the latter ability to grow and compete for nutrients, light, water and space [2].

The laboratory results displayed that, germination of *O. crenata* was significantly lower in response to *T. harzanium* and the imazethapyr alone or in their combinations. It is believed that germination of *Striga* spp is affected by a joint action of germination stimulants and the ethylene produced by the seeds [16]. Germination stimulants elicit ethylene biosynthesis through induction of 1-aminocyclopropane-1-carboxylate (ACC) synthase, the limiting enzyme in ethylene production [16]. Babiker *et al*. [16] showed that the stimulants also increase activity of 1-aminocyclopropane-1- carboxylate oxidase which converts ACC into ethylene.

Moreover, rhizosphere bacteria produce compounds that can motivate protection against pathogens [17]. In fact most plants use similar defence responses to parasitic weeds infection in response to microorganisms [18], strengthens the argument that defence mechanisms against *Striga* were activated in sorghum by PGPR.

With respect to field experiments, faba bean inoculated with *T. harzanium* alone or in combination with bacteria or the herbicide, irrespective to site, significantly reduced *O. crenata* emergence. This may be attributed to a decrease in stimulant production [19] and/or to direct toxicity to the parasite at early developmental stages [15]. The observed reduction in parasitism of sorghum plants by *O. crenata* and mitigation of its suppressive effects on plant growth is consistent with the findings of Lendzemo *et al*. [20] and may be attributed, as reported in similar situations, to increased plant fitness, systemic induced resistance [21]. Previously research findings have determined the link between poor soil fertility, strigolactones production and mycorrhizal infection [22]. Nutrient deficiency is conducive to strigolactones biosynthesis, reduction of shoot branching, maximization of the symbiotic interactions with AMF and facilitation of nutrients uptake. Strigolactones have been shown to function as endogenous phytohormones that curtail shoot branching [23]. The application of herbicide imazethapyr leads considerable to significant reductions in *O. crenara* infestation. The herbicide when applied subsequent to *T. harzanium* was more suppressive to the parasite than when each was applied alone in both sites.

In the Shendi site, emergence of *O. crenata* delayed up to 77%. At 105 DAS, *Orobanche* emergence was inconsistent and maximum emergence was 17.25plant/m^2. At 105 DAS the parasite displayed maximum emergence (76.50plants/m^2) in the untreated control. Despite the encountered low emergence of the parasite, the imazethapyr at 47.6 g a.i. ha^{-1} showed excellent suppression of the parasite by 59.80%. These findings are consistent with those obtained by Elhag [24] who reported that chlorsulfuron at 1.78-2.98 g a.e. ha-1, alone, reduced *Striga* emergence by 61.9-79.4% and 63.5-72.4% early and late in the season, respectively and the corresponding reductions in biomass at harvest were 54.87-73.64%. *Glomus* sp., alone, reduced *Striga* emergence by 71.79-100% and the reduction in biomass at harvest was 74%. The combinations of chlorsulfuron and *Glomus* sp. reduced *Striga* emergence by 67.9-100% and 76-83% early and late in the season, respectively and the reductions in *Striga* biomass at harvest were 66.8-79.8%. These reductions could be attributed to the direct toxicity of the herbicide. The decline in germination of *O. crenata* seeds on prolonged conditioning and the synchrony of faba bean infestation with nodulation which normally occurs within 40 days after crop emergence, it is intriguing and needs to be studied in further details [15]. The observed reduction in parasitism of sorghum plants by *S. hermonthica* and mitigation of its suppressive effects on plant growth is consistent with the findings of [20] and may be attributed, as reported in similar situations, to increased plant fitness, systemic induced resistance [21].

The results revealed that unrestricted *O. crenata* negatively impacted faba bean growth. The observed reductions were (12.3-27.26%) in height, (85.73-114%), in yield. and (63.07-83.23%, in plant biomass. The notable negative impact on faba bean growth attributes due to unrestricted *O. crenata* parasitism is in line with several reports [15] and could be

attributed to perturbation of the hormonal balance of the host plant. *T. harzanium* alone or in combination with bacteria or the herbicide mitigated, to a large extent, the suppressive effects of the parasite on faba bean growth attributes. Among the treatments, *T. harzanium* plus bacteria showed the best performance. The results showed that *T. harzanium* have the potential to reduce *O. crenata* parasitism and damage to faba bean. Furthermore, *T. harzanium* or in combination with BMP+TAL1399 increased plant height by 23.14%, yield by 98.82-124.04%, plant biomass 75.15-113.04% as compared to the corresponding control. These significant increments in faba bean growth attributes are consistent with the concurrent increase in faba bean total biomass.

Faba bean inoculated with *T. harzanium* alone or in combination with further treatments, via bacteria plus the herbicide showed significantly higher growth attributes in comparison to un-inoculated control. The increase in faba bean growth parameters is consistent with the observed delay and decrease in *O. crenata* emergence in response to treatments. Un-inoculated faba bean, *O. crenata* emergence was early and high on the infested untreated control. Delayed *Striga* emergence is reported to reduce its debilitating effects on host plants [25]. Further, mycorrhization has been associated with priming of innate immune plant system and enhancement of resistance to invasion of pathogenic organisms including parasitic Orobanchaceae [26]. Rajapakse and Miller, [27] reported the increase in secondary thickening in higher order roots of mycorrhizal plants, in addition to the association of secondary thickening in plants roots and mechanical resistance to *Striga* [15].

The results revealed that unrestricted *O. crenata* negatively affected faba bean total dry matter accumulation. The reductions in shoot and root biomass are consistent with those previously reported by Frost *et al.* [25] and could be attributed to a multitude of factors related to dry matter production and partitioning including siphoning of nutrients, water and photosynthate by the parasite, reduction in photosynthesis resulting from reduction of chlorophyll contents, perturbations of the hormonal balance of the infected plants and concomitant reductions in stomatal conductance and gaseous exchange [28].These results displayed that the combination between *T. harzanium* and bacterial strains as a biofertilizers could play a vital role in Sudanese agriculture specially in the nitrogen and phosphorus deficient central clay plains.

T. harzanium alone and combination of *T. harzanium* + BMP+TAL1399 increased faba bean biomass, pod/plant and grain yield significantly and 100 seed weight insignificantly as compared to the infested control in both sites. Yield losses due to *Orobanche spp.* vary between 5 to 100% depending on host susceptibility, level of infestation and environmental conditions [29]. Guzm´an-Guzm´an *et al*. [30] reported that the mechanisms employed by *Trichoderma* include secretion of effector molecules and secondary metabolites that mediate the beneficial interaction of *Trichoderma* with plants, providing tolerance to biotic and abiotic stresses. In addition, the genus *Trichoderma* are well-known plant symbionts that exert a positive effect on plant growth, development, crop yield, and elicitation of plant defense responses through the modulation of plant hormonal mechanisms and production of several secondary metabolites [31]. These characteristics make *Trichoderma* an excellent subject for exploration for its positive potential as an important part of integrated *Orobanche* management.

It's worth mentioning that the effect of different treatments in parameters of *O. crenata* and faba bean growth showed same pattern at both sites.

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