International Journal of Agriculture and Environmental Research

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

COUNTERACTING THE EFFECT OF *OROBANCHE CRENATA* INFESTATION ON FABA BEAN (*Vicia faba* L.) BY SOIL MICROORGANISMS AND CHEMICAL FERTILIZERS

Yahia M.Y.A.[*1], Hassan M.M.[2], Elamien M.A.M.[1], Abdalla N.K.[3], Rugheim A.M.E.[1], Abusin R.M.A.[4], Abakeer R.A.[2], Ahmed M.M.[2], Osman A.G.[2], Abdelgani M.E.[2] and Babiker A.G.T.[2]

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

[2]Environment, Natural Resources and Desertification Research Institute, National Center for Research, Sudan

[3]Shendi Research Station, Agriculture Research Corporation, Shendi, Sudan

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

*Corresponding Author

ABSTRACT

A field experiment was conducted at two experimental sites at Khartoum, Sudan, season 2016-2017, to study the effects of *Trichoderma harzianum*, *Bacillus megatherium* var. *phosphaticum* (BMP), *Rhizobium leguminosarum* biovar *viceae* strain (TAL1399), Nitrogen and Phosphorus fertilizer (two levels each) on *Orobanche crenata* incidence and faba bean growth and yield. Nitrogen was applied as urea 95.2kg/hectare (N1) or 190.4kg/hectare (N2) and phosphorus as triple super phosphate 95.2kg/hectare (P1) or 190.4kg/hectare (P2), Treatments were laid out in a Randomized Complete Block Design (RCBD) with four replicates. Results showed that *O. crenata* displayed maximum emergence (76.50 plants/m^2) in the untreated control at 100 days after sowing, the combination of *T. harzianum* + BMP+TAL1399 + fertilizers both levels significantly ($P \leq 0.05$) reduced *O. crenata* emergence as compared to the control at both sites. Application of *T. harzianum* alone or in combination with BMP+TAL1399 + N2P2 significantly ($P \leq 0.05$) reduced *O. crenata* dry weight as compared to the control, irrespective to the site. *T. harzianum* alone or in combination with N1P1 or BMP+TAL1399+N2P2 and the combination with BMP+TAL1399+N2P2 significantly ($P \leq 0.05$) increased plant height as compared to the infested control. The combinations of *T. harzanium*+BMP+TAL1399+N1P1fertilizers significantly ($P \leq 0.05$) increased number of pods per plant, seeds per pod, 100 seed weight and yield (kg/ha), as compared to the control in both sites.

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

Keywords: BMP, Nitrogen, *Orobanche*, Phosphorus, *Rhizobium*, *Trichoderma*

1. INTRODUCTION

Parasitic plants are distributed from the arctic to the tropics. Parasitic weeds of the family Orobanchaceae (*Orobanche*) are considered to be among the most serious agricultural pests of economic importance in many parts of the world [1]. The most economically damaging root parasitic weeds are *Striga*, *Orobanche* and *Phelipanche*. *Orobanche* and *Phelipanche* are widespread in the Mediterranean areas, Asia, East Europe and North Africa attacking crops [2]. Parasitic weeds control methods, including herbicides, soil fumigation, solarisation, mechanical, physical and biological control can provide only unassuming or even zero control [3].

The microbe-plant interaction in the rhizosphere can be beneficial, neutral or deleterious for plant growth [4]. The potential of exploiting natural antagonists (biocontrol) to control parasitic weeds such as *Orobanche* spp. and *S. hermonthica* have received increasing attention [5]. A considerable number of plant pathogens have been studied for their possible use as bioherbicides [6].

Fertilization of faba bean with high rates of nitrogen reduced broomrape (*Orobanche crenata)* infestation and increased faba bean grain yield [7]. Nitrogen, albeit has suppressive effects on parasitism, it adversely affect nodulation and nitrogen fixation in faba beans [8]. Inoculation of legumes with efficient rhizobia at sowing is one of the most important and agronomically eco-friendly practices used for improvement of N fixation. Besides enhanced nodulation and nitrogen fixation, rhizobial seed inoculation can stimulate production of phytohormones, siderophores, and HCN (hydrogen cyanide) as well as microbial diversity and structure, potentially enhancing plant growth-promoting rhizobacteria [9]. Rhizobial inoculation (with *R. leguminosarum*) enhanced phosphate solubilization and P, N uptake and Fe content in lettuce and carrots [10].

This study was conducted to evaluate the effects of *Trichoderma harzianum*, bacterial strains, nitrogen and phosphorus fertilizers on *Orobanche crenata* in faba bean under field conditions.

2. MATERIALS AND METHODS

A field experiment was conducted in two sites [Experimental farm of Environment, Natural Resources and Desertification Research Institute (ENDRI), National Centre for Research (NCR), at Soba south of Khartoum (Soba Site) and at The College of Agricultural Studies farm, Sudan University of Sciences and Technology, Shambat, Khartoum North (Shamat Site)], season 2016-2017.

The treatments included microorganisms [*Trichoderma harzianum*, *Bacillus megatherium* var. *phosphaticum* (BMP), *Rhizobium leguminosarum* biovar *viceae* strain (TAL1399)] and fertilizers [nitrogen and phosphorus] on *O. crenata* incidence and faba bean (cv: BB7) growth and yield.

The fields of both sites were 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 *O. crenata* seeds (5 µg *O. crenata* seeds $hole^{-1}$).

Faba bean seeds treated (when applicable) with TAL1399 and/or BMP as described by Rugheim and Abdelgani [11]. Five grams of *T. harzianum* inoculum carried on rice grains were added to the hole when applicable. Nitrogen (95.2kg/hectare or 190.4kg/hectare) and phosphorus (95.2kg/hectare or 190.4kg/hectare) were applied at sowing when applicable. Untreated seeds were used as a control for comparison. Weeds other than *O. crenata* were manually removed biweekly, starting from two weeks after crop emergence. Treatments were laid out in a Randomized Complete Block Design (RCBD) with four replicates. Subsequent irrigations were made every 2 weeks thinning of plants was done after 15 days from the germination.

Emergent *O. crenata* plants (incidence) were counted at two week intervals starting from 6 weeks after crop emergence. Faba bean height was measured at 4 weeks intervals starting from sowing. Faba bean and *O. crenata* shoot dry weight were measured at harvest. Data collected for faba bean yield and yield components [number of pods/plant, number of seeds/pod, 100-seed weight (g) and grain yield ($kg.ha^{-1}$)] were determined at the end of experiment.

Data collected from the 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 $P \leq 0.05$. *O. crenata* emergence data were square root transformed to fulfill ANOVA requirements.

3. RESULTS

3.1 Effects of treatments on time of *O. crenata* emergence

Shambat site

Results showed that *T. harzianum* alone or in combination with BMP+TAL1399 and N_2P_2 alone significantly ($P \leq 0.05$) delayed *O. crenata* emergence by 26.42 and 24.2 days, respectively as compared to control (figure 1).

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

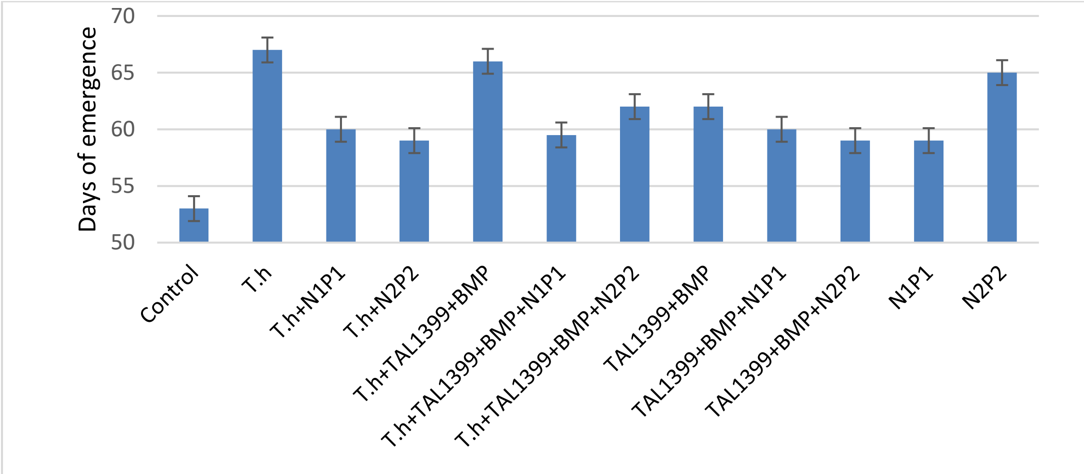

Fig. 1: Effects of treatments on time of *O. crenata* emergence at Shambat site

Soba site

The combination of *T. harzianum* +BMP+TAL1399 + N_1P_1 followed by *T. harzianum* alone significantly ($P \leq 0.05$) delayed *O. crenata* emergence, respectively as compared to control (Figure 2).

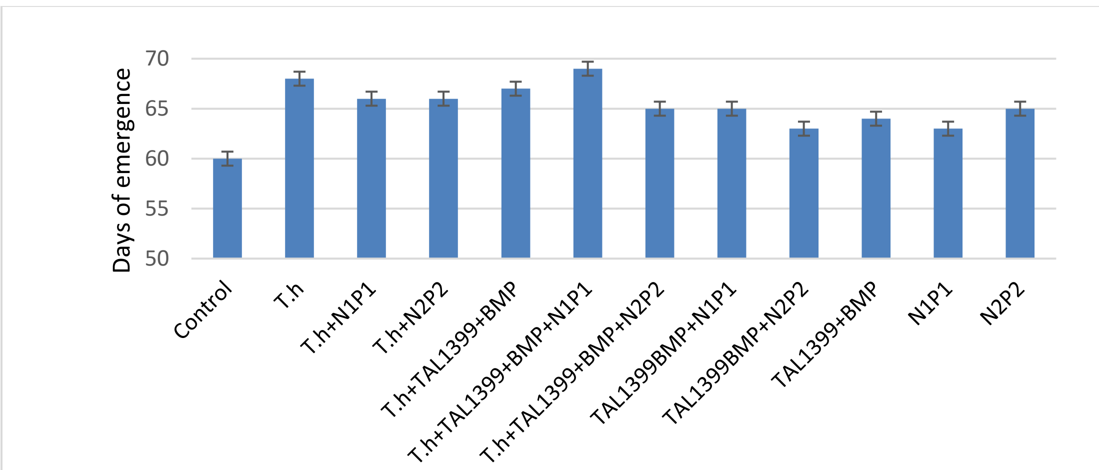

Fig. 2: Effects of treatments on time of *O. crenata* emergence at Soba site

3.2 Effects of treatments on *O. crenata* emergence

Shambat site

ISSN: 2455-6939
Volume:05, Issue:04 "July-August 2019"

At 70 days after sowing (DAS), results showed that application of N2P2 alone or in combination with *T. harzianum* and the combination of *T. harzianum* + BMP+TAL1399 + N1P1 significantly (P≤0.05) reduced the number of *O. crenata* emerged as compared to the infested control. At 85 DAS, the combination of *T. harzianum* + BMP+TAL1399 + N1P1 significantly (P≤0.05) reduced the number of *O. crenata* emerged as compared to the control. At 100 DAS, the combination of *T. harzianum* + BMP+TAL1399 + N2P2 significantly (P≤0.05) reduced the number of *O. crenata* emerged as compared to the control.

Table 1: Effects of treatments on *O. crenata* emergence at Shambat site

Fungi	Bacteria	Fertilizer	Number of *O. crenata* plants/m^2		
			70 DAS	85DAS	100 DAS
Without	Without	N0 P0	1.33^{d}(21.00)	1.83^{f}(68.75)	2.13^{g}(136.00)
		N1P1	0.64^{ab}(5.50)	1.68^{ef}(48.00)	1.82^{de}(68.75)
		N2P2	0.52^{a}(3.50)	1.47^{bcde}(29.50)	1.68^{bc}(48.50)
	BMP + TAL1399	N0 P0	0.80^{abc}(6.00)	1.31^{abc}(21.00)	1.97^{f}(94.25)
		N1P1	1.37^{d}(23.00)	1.52^{cde}(34.25)	1.84^{e}(70.00)
		N2P2	1.35^{d}(22.50)	1.61^{de}(46.25)	2.00^{f}(100.75)
T. harzianum	Without	N0 P0	0.76^{abc}(5.75)	1.30^{abc}(20.75)	1.69^{bcd}(49.00)
		N1P1	1.06^{cd}(10.75)	1.25^{ab}(18.00)	1.58^{ab}(38.50)
		N2P2	0.50^{a} (3.25)	1.35^{bc}(23.00)	1.74^{cde}(55.25)
	BMP + TAL1399	N0 P0	0.96^{bcd}(8.75)	1.44^{bcd}(28.75)	1.81^{cde}(65.50)
		N1P1	0.54^{a}(3.50)	1.12^{a}(13.50)	1.58^{ab}(39.50)
		N2P2	1.23^{d}(18.00)	1.25^{ab}(19.00)	1.53^{a}(34.75)
p≤ 0.05			**	**	**
SE±			0.05661	0.03347	0.02669
SD±			0.39221	0.23192	0.18490

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

Soba site

In Soba field experiment results showed that the application of *T. harzianum* + N2P2, *T. harzianum* + BMP+TAL1399 and *T. harzianum* + BMP+TAL1399 + N1P1 significantly (P≤0.05) reduced *O. crenata* emergence at 70, 85 and 100 DAS as compared to the control (Table 2).

Table 2: Effects of treatments on *O. crenata* emergence at Soba site

Fungi	Bacteria	Fertilizer	Number of *O. crenata* plants/m^2		
			70 DAS	85DAS	100 DAS
Without	Without	N0 P0	0.85^{bc} (10.75)	1.40^{bc} (26.00)	1.79^{c} (62.75)
		N1P1	0.19^{a} (1.75)	1.34^{bc} (25.00)	1.72^{bc} (51.25)
		N2P2	0.52^{abc} (6.50)	1.64^{c} (50.75)	1.77^{c} (66.25)
	BMP + TAL1399	N0 P0	1.02^{c} (15.00)	1.50^{c} (39.00)	2.15^{d} (143.25)
		N1P1	0.21^{a} (2.50)	1.12^{abc} (14.50)	1.82^{c} (66.75)
		N2P2	0.08^{a} (1.25)	1.04^{abc} (19.00)	1.55^{abc} (42.00)
T. harzianum	Without	N0 P0	0.35^{ab} (2.50)	1.28^{abc} (20.75)	1.58^{abc} (38.75)
		N1P1	0.23^{a} (2.75)	1.04^{abc} (14.00)	1.56^{abc} (37.75)
		N2P2	0.00^{a} (1.00)	0.53^{a} (6.50)	1.30^{a} (21.25)
	BMP + TAL1399	N0 P0	0.08^{a} (1.25)	0.56^{a} (7.25)	1.47^{ab} (33.00)
		N1P1	0.37^{ab} (3.50)	0.677^{ab} (12.00)	1.46^{ab} (30.75)
		N2P2	0.00^{a} (1.00)	0.52^{a} (4.50)	1.54^{abc} (36.75)
$p \leq 0.05$			**	**	**
SE±			0.06585	0.08066	0.03847
SD±			0.45624	0.55883	0.26650

Values without brackets () indicate logarithmic transformed data.

Values in brackets () indicate original data.

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

3.3 Effect of treatments on faba bean plant height

Shambat site

At 40 DAS, none of the treatments affected faba bean plant height (Table 3). At 55 and 70 DAS, application of *T. harzianum* +N2P2, *T. harzianum* + N1P1 and BMP+TAL1399 increased plant height insignificantly as compared to infested control. At 100 DAS, application of *T. harzianum* + N2P2, *T. harzianum* + BMP+TAL1399 + N1P1 and BMP+TAL1399+ N2P2 significantly ($P \leq 0.05$) increased plant height as compared to the control.

Soba site

At 40 DAS application of BMP+TAL1399 + N_2P_2 increased plant height albeit not significantly (Table 4). At 55 DAS, *T. harzianum* alone or in combination with N1P1 significantly ($P \leq 0.05$) increased plant height as compared to the control. At 70 and 100 DAS, *T. harzianum*, N2P2,

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

BMP+TAL1399 + N2P2 and *T. harzianum* + BMP+TAL1399 + N2P2 significantly (P≤0.05) increased plant height as compared to the infested control.

Table 3: Effects of treatments on faba bean plant height at Shambat site

Fungi	Bacteria	Fertilizer	Plant height (cm.)			
			40 DAS	55DAS	70DAS	100DAS
Without	Without	N0 P0	28.45	39.45^{ab}	57.71^{abc}	61.30^{c}
		N1P1	28.00	38.45^{ab}	58.62^{abc}	71.25^{ab}
		N2P2	28.00	38.30^{ab}	56.10^{bcd}	70.55^{abc}
	BMP + TAL1399	N0 P0	28.35	41.65^{a}	59.95^{abc}	71.45^{ab}
		N1P1	28.35	39.25^{ab}	51.85^{cd}	67.40^{abc}
		N2P2	26.70	38.133^{ab}	53.67^{bcd}	73.96^{ab}
T. harzianum	Without	N0 P0	27.35	42.00^{a}	59.30^{abc}	73.55^{ab}
		N1P1	30.15	41.00^{a}	62.05^{ab}	73.80^{ab}
		N2P2	28.55	41.60^{a}	65.10^{a}	76.75^{a}
	BMP + TAL1399	N0 P0	29.88	35.65^{b}	49.05^{d}	64.55^{bc}
		N1P1	27.50	38.85^{ab}	61.15^{ab}	75.65^{a}
		N2P2	26.50	41.45^{a}	57.0^{bcd}	74.10^{ab}
Un-infested control			30.25	41.38	61.95	76.95
p≤ 0.05			ns	*	**	**
SE±			.3147	.4589	.8704	.9812
SD±			2.1805	3.3091	6.2762	7.0756

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

Table 4: Effects of treatments on faba bean plant height at Soba site

Fungi	Bacteria	Fertilizer	Plant height (cm.)			
			40 DAS	55 DAS	70 DAS	100 DAS
Without	Without	N0 P0	26.50^{ab}	33.75^{d}	39.95^{b}	48.78^{c}
		N1P1	27.90^{ab}	42.15^{abc}	48.68^{ab}	52.05^{abc}
		N2P2	29.30^{ab}	44.18^{ab}	53.85^{a}	60.85^{a}
	BMP + TAL1399	N0 P0	27.25^{ab}	40.75^{abc}	41.95^{b}	51.45^{bc}
		N1P1	24.90^{b}	41.30^{abc}	45.28^{ab}	50.70^{bc}
		N2P2	32.55^{a}	42.90^{ab}	52.75^{a}	59.78^{ab}
T. harzianum	Without	N0 P0	31.60^{ab}	46.20^{a}	53.50^{a}	59.25^{ab}
		N1P1	26.90^{ab}	45.00^{ab}	51.00^{ab}	56.20^{abc}
		N2P2	25.50^{ab}	39.18^{bcd}	46.10^{ab}	51.00^{bc}
	BMP + TAL1399	N0 P0	24.95^{b}	36.00^{cd}	47.45^{ab}	54.60^{abc}
		N1P1	26.85^{ab}	43.70^{ab}	48.35^{ab}	51.50^{bc}

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

		N2P2	25.88[ab]	40.45[abc]	54.05[a]	58.50[ab]
Un-infested control			28.45	45.33	52.78	57.38
p≤ 0.05			*	**	**	**
SE±			.63380	.71365	1.0699	.98587
SD±			4.39106	4.94428	7.41249	6.83033

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

3.4 Effects of treatments on faba bean and *O. crenata* shoot dry weight

Shambat site

Application of *T. harzianum*, *T. harzianum* + BMP+TAL1399 + N2P2, BMP+TAL1399 + N1P1 and *T. harzianum* + N1P1 significantly (P≤0.05) reduced *O. crenata* dry weight as compared to the control (Table 5).

T. harzianum + N2P2, *T. harzianum* + BMP+TAL1399 + N1P1, *T. harzianum* + BMP+TAL1399 + N2P2 and BMP+TAL1399 + N1P1 significantly (P≤0.05) increased faba bean shoot dry weight as compared to the control.

Table 5: Effects of treatments on faba bean and *O. crenata* shoot dry weight at Shambat site

Fungi	Bacteria	Fertilizer	**shoot dry weight** (g)	
			O. crenata	Faba bean
Without	Without	N0 P0	23.7458[c]	65.0005[d]
		N1P1	17.4890[abc]	68.8693[cd]
		N2P2	16.1150[abc]	73.2303[bcd]
	BMP + TAL1399	N0 P0	22.5275[bc]	69.5558[cd]
		N1P1	12.7002[ab]	82.7415[abc]
		N2P2	13.7258[abc]	77.0485[abcd]
T. harzianum	Without	N0 P0	11.8918[a]	78.7058[abcd]
		N1P1	13.4225[ab]	69.8855[cd]
		N2P2	16.9523[abc]	87.9235[a]
	BMP + TAL1399	N0 P0	16.5040[abc]	75.2548[abcd]
		N1P1	14.4270[abc]	86.9813[ab]
		N2P2	12.5433[ab]	83.1528[abc]
p≤ 0.05			**	**
SE±			.93798	1.51157
SD±			6.49852	10.47250

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

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

Soba site

Results indicated that all treatments significantly (P≤0.05) reduced *O. crenata* dry weight as compared to the control (Table 6). The highest reduction was obtained by *T. harzianum*+ BMP+TAL1399 + N2P2 followed by *T. harzianum* + N2P2. *T. harzianum*+ BMP+TAL1399+ N1P1 gave the highest significant (P≤0.05) result on increasing faba bean shoot dry weight followed by all other *T. harzianum* combinations treatments as compared to the control.

3.5 Effects of treatments on faba bean yield components and yield

Shambat site

Results displayed that application of *T. harzanium*+ N2P2, N1P1 and *T. harzanium* + BMP+TAL1399 + N1P1 significantly (P≤0.05) increased the number of pods per faba bean plant as compared to the control (Table 7). Application of *T. harzanium* + BMP+TAL1399 + N1P1 and BMP+TAL1399 + N2P2 gave significant (P≤0.05) increment on number of seeds per pod as compared to the control. *T. harzanium* + N2P2 and BMP+TAL1399 + N2P2 significantly (P≤0.05) increased 100 seed weight as compared to the control. The combination of *T. harzanium* + BMP+TAL1399 + N1P1 followed by *T. harzanium* + N2P2 and N1P1 alone significantly (P≤0.05) increased faba bean yield (kg/hectare) as compared to the untreated control.

Table 6: Effects of treatments on faba bean and *O. crenata* shoot dry weight at Soba site

Fungi	Bacteria	Fertilizer	**shoot dry weight** (g)	
			O. crenata	Faba bean
Without	Without	N0 P0	27.9675^{d}	34.2975^{d}
		N1P1	18.1350^{c}	48.0675^{cd}
		N2P2	12.150^{b}	49.0000^{cd}
	BMP + TAL1399	N0 P0	6.6300^{ab}	60.1725^{bc}
		N1P1	6.8275^{ab}	51.0775^{bcd}
		N2P2	4.3225^{a}	54.5025^{bc}
T. harzianum	Without	N0 P0	4.5625^{a}	51.5200^{bcd}
		N1P1	3.4225^{a}	69.8855^{ab}
		N2P2	2.8425^{a}	67.7100^{abc}
	BMP + TAL1399	N0 P0	5.3375^{a}	64.2725^{abc}
		N1P1	3.0675^{a}	81.6175^{a}
		N2P2	2.6100^{a}	69.7350^{ab}
p≤ 0.05			**	**

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

SE±	1.18018	2.30847
SD±	8.1765	15.99357

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

Table 7: Effects of treatments on yield components at Shambat site

Fungi	Bacteria	Fertilizer	Yield component			
			No. of pods/plant	No. of seeds/pod	100 seed weight (g)	Yield (Kg/h)
Without	Without	N0 P0	9.00^{e}	2.53^{b}	28.19^{e}	188.21^{d}
		N1P1	28.00^{a}	2.88^{ab}	31.65^{d}	748.34^{ab}
		N2P2	15.50^{cd}	2.63^{b}	35.83^{cd}	500.06^{bcd}
	BMP + TAL1399	N0 P0	13.00^{d}	2.70^{b}	37.77^{bc}	387.99^{bcd}
		N1P1	14.50^{d}	2.700^{b}	29.575^{g}	339.44^{cd}
		N2P2	14.50^{d}	3.33^{a}	41.04^{a}	580.17^{abcd}
T. harzianum	Without	N0 P0	13.50^{d}	2.73^{b}	36.69^{bcd}	396.48^{bcd}
		N1P1	14.25^{d}	2.95^{ab}	33.64^{e}	414.64^{bcd}
		N2P2	28.75^{a}	3.00^{ab}	41.20^{a}	881.60^{a}
	BMP + TAL1399	N0 P0	23.50^{abc}	2.73^{b}	35.33^{de}	664.59^{abc}
		N1P1	26.25^{ab}	3.33^{a}	36.82^{bcd}	943.003^{a}
		N2P2	21.50^{bc}	2.65^{ab}	25.3^{h}	422.09^{bcd}
Un-infested control			17.75	2.93	28.187	429.10
p≤ 0.05			**	*	**	**
SE±			1.04782	.05392	.68870	38.21573
SD±			7.55593	.38881	4.96632	275.5773

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

Soba site

The combinations of *T. harzanium*+ BMP+TAL1399 + N1P1 and *T. harzanium* + BMP+TAL1399 + N2P2 significantly (P≤0.05) increased faba bean pods number per plant, seeds per pod, 100 seed weight and yield (kg/hectare) as compared to the control (Table 8).

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

Table 8: Effects of treatments on faba bean yield components at Soba site

Fungi	Bacteria	Fertilizer	Yield component			
			No. of pods/plant	No. of seeds/pod	100 seed weight (g)	Yield (Kg/h)
Without	Without	N0 P0	12.75^{e}	2.63^{b}	43.8250^{d}	483.97^{d}
		N1P1	16.00^{cde}	2.80^{ab}	45.51^{bcd}	565.46^{cd}
		N2P2	13.50^{de}	2.63^{b}	45.84^{bcd}	489.76^{d}
	BMP + TAL1399	N0 P0	18.50^{bcde}	2.70^{b}	45.82^{bcd}	692.06^{bcd}
		N1P1	16.00^{bcde}	2.70^{b}	49.61^{a}	714.40^{bcd}
		N2P2	19.00^{bcde}	3.33^{a}	48.63^{ab}	687.46^{bcd}
T. harzianum	Without	N0 P0	21.75^{abcd}	2.73^{b}	44.7675^{cd}	781.88^{bcd}
		N1P1	24.75^{ab}	2.95^{ab}	45.26b^{cd}	904.88^{b}
		N2P2	22.75^{abc}	3.00^{ab}	47.49^{abc}	899.90^{b}
	BMP + TAL1399	N0 P0	22.00^{abcd}	2.73^{b}	47.99^{abc}	862.11^{bc}
		N1P1	23.00^{abc}	3.33^{a}	49.44^{a}	1207.99^{a}
		N2P2	27.75^{a}	2.65^{b}	50.49^{a}	970.09^{ab}
Un-infested control			16	2.64	45.51	565.46
$p \leq 0.05$			**	*	**	**
SE±			.90869	**.05392**	**.41199**	**38.7137**
SD±			6.29558	**.38881**	**2.85438**	**26.8216**

Means followed by the same letter (s) are not significantly different according to DMRT at $P \leq 0.05$

DISCUSSION

Global increases in food production achieved in recent decades have required large increases in the use of synthetic pesticides to control pests, pathogens and weeds of crops [12], but the increasing use of synthetic pesticides is no longer sustainable. Considering the constraints to successfully controlling parasitic weeds, *Orobanche* so far, it is well recognized that no single method of control can provide effective and economically acceptable solution. Therefore, an integrated control approach is essential and useful to the farmers, in order to achieve sustainable crop production.

In the present study, integration between biological and chemical control for management of broomrapes' infection was investigated under field conditions. In terms of *O. crenata* emergence, application of *T. harzianum* alone or in combination with BMP+TAL1399 or BMP+TAL1399 + N1P1 and N1P1 alone significantly delayed the parasite emergence. At Shambat site, the *T. harzianum* delayed the *O. crenata* emergence up to 63.97% at 100 DAS. *T.*

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

harzianum +BMP+ TAL1399+N1P1 or N2P2 delayed emergence of *O. crenata* up to 70% at 100 DAS, *T. harzianum* + N1P1, N2P2 between 59-71 at 100 DAS. At Soba site, *T. harzianum* delayed the *O. crenata* emergence up to 38.25% at 100 DAS. *T. harzianum* +BMP+TAL1399+N1P1 or N2P2 delayed emergence of *O. crenata* up to 47.84% at 100 DAS *T. harzianum* + N1P1, N2P2 up to 66% at 100 DAS. *O. crenata* emergence was inconsistent and maximum emergence was 17.25 plants/m^2. At 100 DAS the parasite displayed maximum emergence (76.50 plants/m^2) in the untreated control. Furthermore, the combination of *T. harzianum* + BMP+TAL1399 plus fertilizers significantly reduced *O. crenata* emergence as compared to the control, irrespective to site. While the application of *T. harzianum* alone or in combination of BMP+TAL1399 + N2P2 significantly ($P \leq 0.05$) reduced *O. crenata* dry weight as compared to the control, irrespective to the site. Akiyama *et al*. [13] reported the link between poor soil fertility, strigolactones production and mycorrhizal infection. Nutrient deficiency is conducive to strigolactones biosynthesis, reduction of shoot branching, maximization of the symbiotic interactions with AMF and facilitation of nutrients uptake. Boari *et al*. [14] reported that *Trichoderma* spp. used for biocontrol of parasitic weeds, acting as a 'physiological'barrier, by preventing the germination of their seeds through the ability to biotransform the stimulatory signals. Furthermore, Neondo *et al*. [15] reported that most of the bacterial isolates that caused *S. hermonthica* seed decay had close genetic relationship with members of the genus *Bacillus* and with only a single bacterial isolate having close genetic affiliation to *Streptomyces* and *Rhizobium*.

Parasitic plants generally prevail on nutrient deficient soils, and many studies have reported a decrease in parasitic weeds infection upon application of N and P [16]. The germination of *Striga* and *Orobanche* seeds are associated with the secretion of germination stimulants by host plants. The secretion ultimately depends upon the nutrient status of the soil [17]. It has been demonstrated that under N and P deficiency, host plants secrete high amounts of germination stimulants. The suppressive effects of nitrogen on parasitic weeds infestation were attributed to delayed germination, reduced radical elongation, stimulants production and reduction of seeds response to the stimulants [18]. Mokhtar *et al*. [19] reported that soil treatment with three fungal agents (*T. harzianum* T1, T. *harzianum* T3and *T. viride* T2) each alone was efficient and cost-effective method in reducing infection minimizing the number of spikes of parasitic plant on host plants and increasing yields of faba bean. Borriss *et al*. [20] demonstrated that *B. amyloliquefaciens subsp. Plantarum* is able to colonize plant roots and produce plant growth hormone known as indole-3-acetic acid. Other studies have also established that *B. amyloliquefaciens* have the ability to produce numerous antimicrobial and bioactive metabolites such as surfactin, iturin and fengycin which have well-established in vitro activity,the production of these compounds highlights *B. amyloliquefaciens* and its close relative in the study (KY041696) as good candidates for the development of biocontrol agents [21].

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

The notable negative impact on faba bean growth attributes due to unrestricted *Orobanche* parasitism is in line with several reports [2, 22]. From the results of two sites, application of *T. harzianum* alone or in combination withN_1P_1 or BMP+TAL1399 + N_2P_2 and the combination of BMP+TAL1399+N_2P_2 significantly increased plant height as compared to the infested control. Biofertilizers play a significant role in reduction of chemical fertilizer use. Wu *et al*. [23] reported that the mixture of four microbes {AMF (*Glomus mossae* or *Glomus intraradices*), N-fixer (*Azotobacter chroococcum*), P-solubiliser (*Bacillus megatherium* var *phosphaticum*) and a K-solubilizer (*Bacillus mucilaginous*)} significantly increased the growth of *Z. mays*.

With respect to faba bean shoot dry weight, results showed that the combinations of *T. harzianum* + BMP+TAL1399+ N1P1 or *T. harzianum* + N2P2 significantly increased faba bean shoot dry weight, irrespective to site as compared to the control. Root colonization with microbes frequently results in enhancing growth and development, crop productivity or induction of resistance to abiotic and biotic factors.

In terms of faba bean yield components, results displayed that the combinations of *T. harzanium*+ BMP+TAL1399 +fertilizers significantly ($P \leq 0.05$) increase number of pods per plant, seeds per pod, 100 seed weight and yield (kg/hectare), irrespective to site as compared to the control. Haque *et al*. [24] reported that application of N fertilizer with *Trichoderma*-enriched bio-fertilizers increased tomato yield by 203% over the control. Furthermore, *Trichoderma*-enriched bio-fertilizer could save at least 50% N fertilizer uses for tomato and could reduce excessive uses of NPK for crop cultivation. The present study revealed that the combination of *T. harzianum* plus bacterial strains and chemical fertilizers was appeared to be significantly reducing *O. crenata* infestation and improve growth, yield and yield components of faba bean as compared to the control under field conditions.

CONCLUSIONS

- Appropriate cropping systems, microbial control (*Trichoderma* fungi) and fertilizers (NP) can be used to alleviate the *Orobanche crenata* harmful effect.
- It was clearly observed that the *Trichoderma* and bacterial strains enriched with fertilizers had a positive impact on growth and yield of faba bean.
- Further work will be carried out to develop a seed treatment with the mycoherbicide in order to reduce the amount of the inoculum required as well as labor during application.
- Additional research is needed to determine the functions and mode of action of the virulent enzymes of these microbes needed for development of novel biological control remedies of *Orobanche* spp.

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

REFERENCES

[1] Habimana, S.; Nduwumuremyi, A.; Chinama, R. Management of *Orobanche* in field crops- A review. *Journal of Soil Science and Plant Nutrition*, 2014, 14(1): 43-62.

[2] Parker, C. and Riches, C.R. Parasitic Weeds of the World: Biology and Control. CAB International, Wallingford, UK. 1993, pp332.

[3] Gressel, J. and Joel, D.M. Weedy Orobanchaceae: the problem, in Parasitic Orobanchaceae. Parasitic Mechanisms and Control Strategies, ed. by Joel DM, Gressel J and Musselman LJ. Springer-Verlag, Berlin/Heidelberg, Germany, 2013, pp. 309–312.

[4] Mastretta, C.; Barac, T.; Vangronsveld, J.; Newman, L.; Taghavi, S. and van der Lelie, D. Endophytic bacteria and their potential application to improve the hytoremediation of contaminated environments. *Biotechnology and Genetic Engineering Reviews*, 2006, 23: 175–207.

[5] Gafar, N.; Hassan, M.; Rugheim, A.; Osman, A.; Mohamed, I.; Abdelgani, M.; and Babiker, A. Evaluation of endophytic bacterial isolates on germination and haustorium initiation of *Striga hermonthica* (Del.). *International Journal of Farming and Allied Sciences*, 2015, 4(4): 302–308.

[6] Pilgeram, A. and Sands, D. Bioherbicides. In M. Hofrichter (Ed.), *Industrial Applications* (2nd ed.). Berlin Heidelberg: Springer Verlag. 2010, pp. 395–405

[7] Elabaied, M.E.; Hassan, M.M.; Rugheim, A.M.E.; Osman, A.G.; Ahmed, M.M.; Yahia, M.Y.A.; Abakeer, R.A..; Abusin, R.M.A.; Mohamed, M.A.; Abdelgani , M.E. and Babiker A.G.E. Role of bacteria, nitrogen, phosphorus and the herbicide imazethapyr on inhibiting *Orobanche crenata* Forsk infestation in *Vicia faba* L., *Journal of Agriculture and Veterinary Science*, 2017, 10(10): 13–22.

[8] Igbinnosa, I.; Cardwell, K.E. and Okonkwo, S.N.C. The effect of nitrogen on the growth and development of giant witchweed, *Striga hermonthica* Benth. Effect on cultured germinated seedlings in host absence. *European Journal of Plant Pathology*, 1996, 102: 77- 86.

[9] Trabelsi, D.; Ammar, H.B.; Mengoni, A. and Mhamdi, R. Appraisal of the crop-rotation effect of rhizobial inoculation on potato cropping systems in relation to soil bacterial communities. *Soil Biol. Biochem.*, 2012, 54: 1–6.

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

[10] Yadav, J. and Verma, J.P. Effect of seed inoculation with indigenous *Rhizobium* and plant growth promoting rhizobacteria on nutrients uptake and yields of chickpea (*Cicer arietinum* L.). *Eur. J. Soil Biol.*, 2014, 63: 70–77.

[11] Rugheim, A.M.E. and Abdelgani, M.E. Effects of microbial and chemical fertilization on yield and seed quality of faba bean (*Vicia faba*). *International Food Research Journal*, 2012, 19 (2): 417-422.

[12] Oerke, E.C. Centenary review crop losses to pests. *Journal of Agricultural Science*, 2006, 144: 31–43.

[13] Akiyama, K.; Matsuzaki, K. and Hayashi, H. Plant sesquiterpenes induce hyphal branching in arbuscular mycorrhizal fungi. *NATURE*, 2005, 435: 824-827.

[14] Boari, A.; Ciasca, B.; Pineda-Martos, R.; Lattanzio, V.M.; Yoneyama, K. and Vurrom M. Parasitic weed management by using strigolactone-degrading fungi. *Pest Manag. Sci.*, 2016, 72(11): 2043-2047.

[15] Neondo, J.O.; Alakonya, A.E. and Kasili, R.W. Screening for potential *Striga hermonthica* fungal and bacterial biocontrol agents from suppressive soils in Western Kenya. *BioControl*, 2017, 1-13.

[16] Adagba, M.A; Lagoke, S.T.O. and Imolehin, E.D. Nitrogen effect on the incidence of *Striga hermonthica* (Del.) Benth in upland rice. *Acta Agronomica Hungarica*, 2002, 50(2):145-150.

[17] Jamil, M.; Charnikhova, T. and Cardoso, C. Quantification of the relationship between strigolactones and *Striga hermonthica* infection in rice under varying levels of nitrogen and phosphorus. *Weed Research*m 2011, 51: 373–385.

[18] Hassan, M.M.; Abdel gain, M. E. and Babiker, A.G.T. Potential of bacterial strains and nitrogen in reduction of *Striga hermonthica* (Del.) Benth. infesting sorghum. *Advances in Environmental Biology*, 2009, 3(1): 1-9.

[19] Mokhtar, M.; Abdel, K. and Nehal, S.E.M. Prospects of mycoherbicides for control of broomrapes (*Orobanche* spp.). *Egypt. J. Plant Prot. Res.*, 2009, 49(1): 64-74.

[20] Borriss, R.; Chen, X.H.; Rueckert, C.; Blom, J.; Becker, A.; Baumgarth, B.; Fan, B.; Pukall, R.; Schumann, P.; Sproer, C.; Junge, H.; Vater, J.; Pühler, A. and Klenk, H.P. Relationship of *Bacillus amyloliquefaciens* clades associated with strains DSM 7T and

ISSN: 2455-6939

Volume:05, Issue:04 "July-August 2019"

Bacillus amyloliquefaciens subsp. plantarum subsp. nov.based on their discriminating complete genome sequences. *Int. J. Syst. Evol. Microbiol.*, 2011, 61:1786–1801.

[21] Liu, X.; Min, Y.; Wang, K.; Wan, Z. and Zhang, Z. Draft genome sequence of *Bacillus amyloliquefaciens* HB-26. *Standards in Genomic Sciences*, 2014, 9: 775–782.

[22] Yahia, M.A.; Hassan, M.M.; Elamien, M.A.M.; Abdalla, N.K.; Rugheim, A.M.E.; Elsalahi, R.H.; Abusin, R.M.; Abakeer, R.A.; Ahmed, M.M.; Osman, A.G.; Abdelgani, M.E. and Babiker, A.E. Laboratory and Field Studies of *Trichoderma harzianum*, Bacterial Strains and Imazethapyr on *Orobanche crenata* Forsk Infesting *Vicia faba*. *Asian Journal of Agriculture and Food Sciences*, 2018, 6 (6): 209-217.

[23] Wu, S.C.; Cao, Z.H.; Li, Z.G.; Cheung, K.C. and Wong, M.H. Effect of biofertiliser containing N-fixer, P and K solubilizers and AM fungi on maize growth: a greenhouse trial. Geoderma, 2005, 125: 155-166.

[24] Haque, Md.M.; Ilias, G.N.M. and Molla, A.H. Impact of *Trichoderma*-enriched Biofertilizer on the growth and yield of mustard (*Brassica rapa* L.) and tomato (*Solanum lycopersicon* Mill.). *The Agriculturists*, 2012, 10(2): 109-119.

