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Effectiveness of *Beauveria bassiana* (Bals.) Vuill. and *Metarhizium anisopliae* (Metsch.) (Deuteromycotina: Hyphomycetes) as Biological Control Agents of the Onion Thrips, *Thrips tabaci* Lind.

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ABSTRACT

This experiment was carried out at the experimental farm, Faculty of Agriculture, Tanta University, during 2014 and 2015 seasons, to study the population density of onion thrips, *Thrips tabaci* Lind.(Thysanoptera: Thripidae) under onion field conditions. In the middel of February the population density exceeded 10 alive thrips per plant which the time of control. Population peak reached during 2nd week of April 2014. Two entomopathogenic fungi *Beauveria bassiana* (Bals.) Vuill. and *Metarhizium anisopliae* (Metsch.) (Deuteromycotina: Hyphomycetes) isolates were treated on *T. tabaci* in labortatory conditions with concentrations of 10^4, 10^5, 10^6, 10^7 and 10^8 conidia/ml. Therefore the LC_{50} values were 0.9 x 10^5 and 2.9 x 10^6conidia/ml. for *B. bassiana* and *M. anisopliae* isolates, respectively. Accordingly the two isolates were sprayed in the middel of february in the onion field to control *T. tabaci* with concentrations of 0.9 x 10^6 and 2.9 x 10^7 conidia/ml for *B. bassiana* and *M. anisopliae* isolates, respectively and compare with Actellic 50% as a recommended pesticide with 500 ml/Fadan. The reduction in insect numbers were 74, 62.87 and 87.08 % for *B. Bassiana, M. anisopliae* isolates and Actellic 50%, respectively. This result revealed that the two isolates may be effective against *T. tabaci* in field conditions especially *B. bassiana* isolate.

Keywords: *Thrips tabaci*, *Beauveria bassiana*, *Metarhizium anisopliae*,Onion crop.

INTRODUCTION

In Egypt, The onion thrips, *Thrips tabaci* Lindeman it is a serious insect pest that influence onion product by direct nutrition on plant sap as long decreasing the quality and quantity. Heavey infestation leads to decrease the onion yield (Alston and Drost, 2008). The damage is caused by severe feeding of adults and nymphs on green plant tissue (Trdan *et al*., 2005). Damaged leaves areas become desiccated causing a silvery flecked appearance as well as distorted (Diaz-Montano *et al*., 2011). Great yield damages of up to 50% have been refered to *T. tabaci* on garlic in India (Butani and Verma, 1976; Mote, 1976). Control of thrips pests is intricate due to their little size and their hidden habits, as they feed hidden in incisions of flowers and leaf sheaths. Their rise reproductive capability leads quickly to big numbers invading individual plants (Lewis, 1999). In generality target crops, use of synthetic pesticides is the widely used choice for mangementing thrips. The secretive behavior and speedy reproduction of thrips entail recurrent applications of Pesticides. These treatments cause residue and insecticide resistance problems, are costly with regarded to risks to nontarget organisms (Maniania *et al*., 2003). Two species of entomopathogenic fungi in specific, *B. bassiana* and *M. anisopliae*, have near global allocations (Bidochka *et al.,* 1998) and have been used widely in biological control. The entomopathogenic fungus, *B. bassiana* is used in greenhouse and crops as a agent for the control of many agricultural pest arthropods, including whiteflies, thrips, aphids, weevils, mealybugs, and psyllids (Shah and Goettel, 1999). Maniania *et al*., (2003) found the entomopathogenic fungi *M. anisopliae* had potential pathogenic effect against *T. tabaci* in field trial. Therefore, the aim of this study was to evaluate the efficacy of certain entomopathogenic fungi, *B. bassiana* and *M. anisopliae* against onion thrips, *T. tabaci* in onion fields, under Egyptian conditions.

MATERIALS AND METHODS

1-Population denesity of onion thrips, *T. tabaci*

This experiment was carried out at the experimental farm, Faculty of Agriculture, Tanta University, during 2014 season, to study the population density of Onion thrips, *T. tabaci* under field conditions. the normal agricultural practices were adopted without any insecticidal applications. Sampling started on 8th January, and countinued till 23th April, 2014. Samples were randomly taken at 7 days intervales and each sample comprised 10 onion plants, each plant was kept inside paper bag and transported to the laboratory. Plants were subjected to investigate and number of adults and nymphs of the onion thrips were recorded by a binocular sterio in the same day.

2-Susceptibility of *T. tabaci* to entomopathogenic fungi

Experimental insects:

The onion plants with high infestion of *T. tabaci* were collected from field and maintained in paper pags for 2 days. Dead and injured insects were discarded and healthy ones were used in the experiments.

The entomopathogenic fungi:

The two entomopathogenic fungi, *B. bassiana* and *M. anisopliae* isolates from the Mycology Center, Faculty of Science, Assiut University (AUMC) were used (El-Sheikh, M. F.,2012) as shown in table 1. Fungi isolates were grown on rice in incubator at 26 ± 1 c (Castineiras *et al.,* 1996).

Table 1. Entomopathogenic fungi isolates used in the present study.

Isolate	AUMC number	Date	Host insect	locality
Beauveria bassiana	AUMC 4072	2006	*Sitona lividipes* Fab. adults under bark of Eucalyptus trees	Behera governorate
Metarhizium anisopliae	AUMC 5130	2008	*Hypera brunneipennis* Boh. larvae in clover field	Gharbia governorate

Bioassay

The Susceptibility of *T. tabaci* to the entomopathogenic fungi isolates, *B. bassiana* and *M. anisopliae* were bioassayed on second instar nymph of *Thrips tabaci*. For each fungi isolate, conidia suspensions contained 10^4, 10^5, 10^6, 10^7 and 10^8 conidia/ml and 0.1% Tween 80 were prepared and used to contaminate nymphs by spraying them in 5 ml of the suspension in Petri dish 15cm in diameter. For each concentration 80 nymphs were used. Treated nymphs were divided into 4 replicates, each of 20 nymphs were released using camel hair brush on 10 cm of fresh onion leaf which renewed every two days. Control insects were treated with water containing 0.1% Tween 80. All insects were kept at 25±2°C, The mortality data was recorded after 3, 5, and 7 days, and dead nymphs were counted, recorded and removed. The LC_{50} for each isolate was determined. Immobilized insects were removed and considered as dead if they did not move when prodded with camel hair brush.

3- Efficacy of entomopathogenic fungi againts *T. tabaci* under the field conditions

The study was carried out to evaluate the efficacy of two fungus isolates, being *B. bassiana* and *M. anisopliae* to control *T. tabaci* in a onion field. The experiment was conducted at the experimental farm of the Faculty of Agric., Tanta Univ. in 2015 season. The area was divided into 4 splits (2 fungus isolates, recommended pesticide (Actellic® EC (Pirimiphos Methyl 50%) 500 cm3 /fedan) and control). Onion was sown on 1st of December 2014 and plants received usual agricultural practices without chemical applications. The spraying was done on the 15th of February, 2015. For each application 5 replicates, each with three rows 15 m long were used. The sampled included five plants to investigate for alive adults and nymphs of *T. tabaci*. Plants were gently cut, placed in paper bags and transmitted to the laboratory to thrips counts. The *B. bassiana* isolate was carried out with a concentration of 0.8 x 10^6 conidia/ml and the *M. anisopliae* isolate was carried out with a concentration of 2.44 x 10^7 conidia/ml containing 0.1% Tween 80. Control plots were sprayed with tap water containing 0.1% Tween 80. The different treatments were randomly distributed in each split.

The number of survived *T. tabaci* nymphs and adults were counted after 3, 7 and 10 days (Sayeda, S. A. and M. M. El-Mogy 2011). Reduction rate among the populations of the target insects were calculated according to the formula of Henderson and Tilton, 1955.

Statistical analyses.

Data were subjected to the analysis of variance (ANOVA). Treatment means were compared by Tukey honestly significant different test. Computation were done using computer software Minitab 16 all data were corrected for control mortality using Abbott formula (Abbott, 1925).

RESULTS AND DISCUSSION

1. populations density of *T. tabaci* in onion fields.

Population density of onion thrips *T. tabaci* (adults and nymphs) on onion field in Tanta at Gharbia governorate in 2014 season are presented in Fig. (1). Thrips were found on onion plants from 15 Jan. to 23 Apr. Population density was relatively with low numbers in January and February then they tended to increase rapidly to reach maximum abundance in the 2nd of April. Total number of thrips during the whole period of onion growing season 2014 was 24694 insects. The economic threshold level of thrips was 10 thrips per plant (Kendell and Capinera, 1987) which sited between 12 to 19 Fab., therefore the control time of thrips in the field was determined in the middle of February.

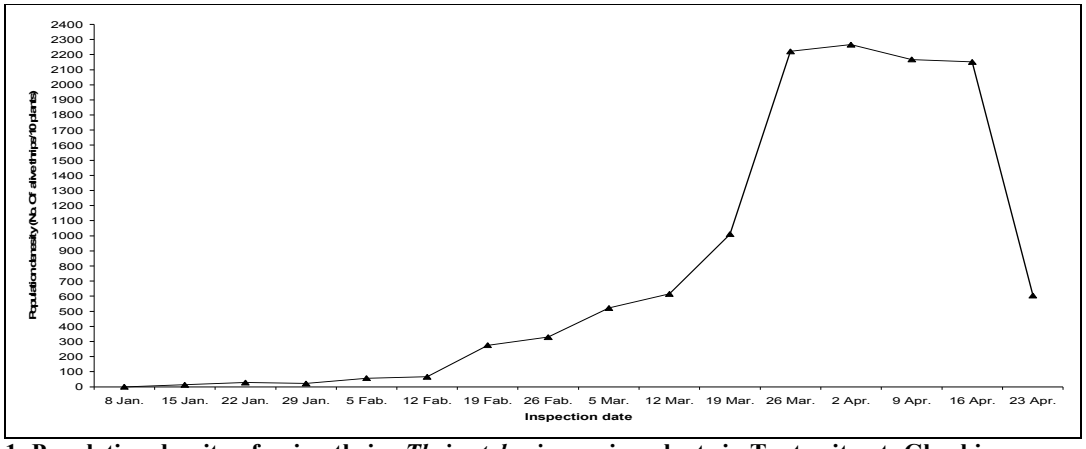

Fig. 1. Population density of onion thrips *Thrips tabaci* on onion plants in Tanta city at Gharbia governorate during season 2014.

In similar study, Hayder and sherif (1987) observed that *T. tabaci* population began to build up in onion fields by early February and reached maximum during April and May in Giza governorate. Also Massry 2002 studied the population fluctuations of thrips in El-Menoufia governorate and found two successive population peaks during the 3rd week of March, and the 1st week of May.

2- Susceptibility of *T. tabaci* to entomopathogenic fungi

As shown in Table (2) the Susceptibility of the *T. tabaci* to entomopathogenic fungi isolates, *B. bassiana* and *M. anisopliae* were bioassayed on second instar of *T. tabaci* nymphs. For each fungi isolate, conidia suspensions contained 10^4, 10^5, 10^6, 10^7 and 10^8 conidia/ml. All concentrations induced mortality for the

insects and mortality increased as the conidia concentration increased. All the tested concentrations began to kill nymphs throughout the period extending from the 3rd to the 7th days after treatment. In *B. bassiana* isolate corrected mortality ranged 35.90 – 93.59 % while in *M. anisopliae* isolate was ranged 8.97 – 84.62 %. and mortality was very low for the concentrations 10^4 and 10^5 conidia/ml.

Table 2. Mortality of the second instar nymph of *T. tabaci* treated with different conidia concentrations of *B. bassiana* and *M. anisopliae* under laboratory conditions.

The fungus isolates	Concentration Conidia/ml	Mean No. of dead nymphs (days after treatment)			Mortality	
		3	5	7	%	corrected
Beauveria bassiana	1 x 10^4	11	24	30	37.5	35.90
	1 x 10^5	15	30	37	46.25	44.87
	1 x 10^6	25	50	52	65	64.10
	1 x 10^7	47	65	73	91.25	91.03
	1 x 10^8	53	71	75	93.75	93.59
Metarhizium anisopliae	1 x 10^4	2	4	9	11.25	8.97
	1 x 10^5	3	7	11	13.75	11.54
	1 x 10^6	14	31	38	47.5	46.15
	1 x 10^7	14	40	47	58.75	57.69
	1 x 10^8	43	63	68	85	84.62

Data represented in Table (3) showed that the calculated LC_{50} values were 0.9 x 10^5 and 2.9 x 10^6 conidia/ml. for *B. bassiana* and *M. anisopliae* isolates, respectively and values of slopes. The probit regression lines presented in fig. 2. This results indicated that the *B. bassiana* isolate is more virulent than *M. anisopliae* isolate.

Table 3. Values of LC_{50} (conidia/ml) for the two isolates of the entomopathogenic fungi against 2nd instar of *T. tabaci* nymphs.

Fungal isolates	LC_{50} value conidia/ ml.	Lower	Upper	Slop
B. bassiana	0.9 x 10^5	0.3 x 10^5	2.1 x 10^6	0.4819
M. anisopliae	2.9 x 10^6	1.17 x 10^6	7.2 x 10^6	0.5551

These results are in line with those of Arthurs *et al*., (2012) who estimated commercial strains of entomopathogenic fungi for control of chilli thrips, *Scirtothrips dorsalis* Hood (Thysanoptera: Thripidae). In laboratory bioassays, LC_{50} values against second instars of *S. dorsalis* were 1.1 x 10^8 CFU/mL for *B. bassiana* GHA, with higher values 7.0 x 10^5 for *M. brunneum* F52 and 9.9 x 10^5 for *Isaria fumosorosea* Apopka 97. Second instars were relatively less susceptible to all isolates. Also, Sengonca *et al*. (2006) examined 41 isolates from 25 species and 11 genera of entomopathogenic fungi in Thailand against first instar of *F. occidentalis* on bean leaves. Among the 14 most virulent isolates, LC_{50} values of *Beauveria* spp. Ranged from 2.4 x 10^4 to 5.9 x 10^6 conidia/ml., *Metarhizium* spp. from 2.0 x 10^4 to 5.0 x 10^5 conidia/ml., and *Isaria* spp. from 3.9 x 10^4 to 5.5x 10^6 conidia/ml.

3- Efficacy of entomopathogenic fungi againts *T. tabaci* under the field conditions

Data in Table (4) presented reduction in the population density of *T. tabaci* nymphs and adults after treating onion plants with *B. bassiana* and *M. anisopliae* isolates with 0.9 x 10^6 and 2.9 x 10^7 conidia/ml. respectively, compareson with Actellic 50% as a recomended insectcide. Treating the onion plants with *B. bassiana* isolate resulted in a reduction in insect population ranged from 70.59 to 78.62% with a total reduction 74%, and in the case of *M. anisopliae* isolate the reduction in the insects populations ranged from 61.47 to 65.67% with a total reduction 62.87%, while Actellic 50% resulted a reduction ranged from 78.34 to 93.45% with a total reduction of 87.08%. Total reduction was hightest in Actellic 50% followed by *B. bassiana* and *M. anisopliae* isolates. This result revealed that the two isolates may be effective against *T. tabaci* in onion fields.

Results of the present study are in agreement with those of Sayeda and El-Mogy (2011) who reported that two rates of *B. bassiana* (Bio-Power) formulations were sprayed (5 and 10 ml/L.) three times for two seasons and compared with Malathion (reference insecticide). For one spray reductions were 68.7 -79.8% for two rates, respectively for first season while on second season were 25.4- 96.8% for two rates. Also Ekesi *et al*., 1998 executed field experiments at Mbita, western Kenya for two seasons to estimate the capability of the entomopathogenic fungus, *M. anisopliae*, as a biological control of the legume flower thrips, *Megulurothrips sjostedti* (Trybom), on cowpea. An ultra-low-volume (ULV) oil/aqueous formulation and a high-volume (HV) aqueous formulation of conidia were applied three times each at two concentrations of 1 x 10^{11} and 1 x 10^{13} conidia ha -I. Compared with the untreated control, both formulations significantly reduced thrips populations and plant damage in both seasons.

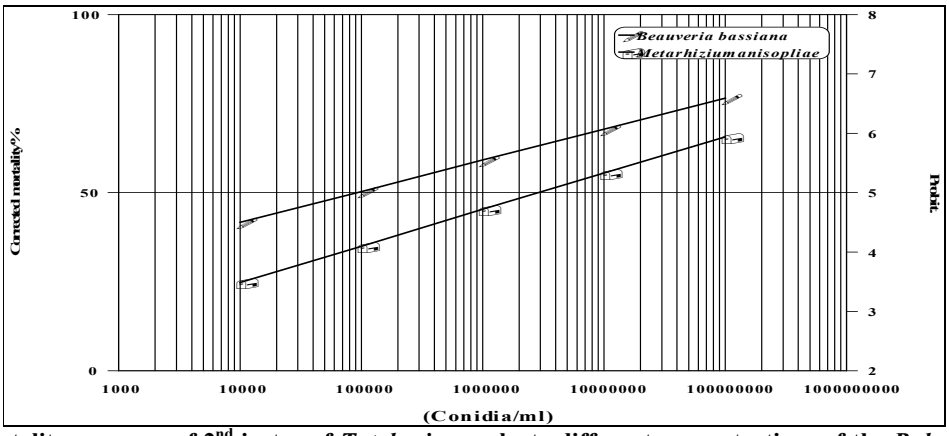

Fig 2 . Mortality response of 2nd instar of *T. tabaci* nymphs to different concentrations of the *B. bassiana* and *M. anisopliae* isolates

Table 4. The efficiency of *B. bassiana* and *M. anisopliae* isolates against *T. tabaci* in onion field.

Treatment	Mean of alive thrips	Reduction % of alive thrips±SE			Mean of alive thrips	Reduction %
	Before spray	Day post spray			Grand mean	
		3	7	10		
Beauveria bassiana	143±3.11	70.59±1.32 ab	73.15±2.13 b	78.26±3.31 a	61±1.55	74±1.88 b
Metarhizium anisopliae	87±2.54	61.47±2.22 b	65.67±3.22 b	61.47±6.42 b	53±1.02	62.87±2.81 c
Actellic 50%	121±2.92	78.34±3.66 a	89.42±2.14 a	93.45±1.41 a	25.66±2.34	87.08±0.782 a
F value		7.84	20.73	14.71		37.44
P value		0.007	0.000	0.001		0.000

Means within a column followed by the same letter are not significant using Duncan multiple range test. Waller and Duncan (1969)

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فعالية الفطريات الممرضة للحشرات (Bals.) *Beauveria bassiana* و (Metsch.) *Metarhizium anisopliae* (الفطريات الناقصة: الفطريات الهيفية) كعوامل مكافحة حيوية علي تربس البصل *Thrips tabaci* Lind.

محمد فاضل محمود الشيخ

قسم وقاية النبات - الحشرات الاقتصادية- كلية الزراعة بطنطا – جامعة طنطا

أجريت هذه الدراسة بالمزرعة البحثية بكلية الزراعة بطنطا بجامعة طنطا خلال موسمي ٢٠١٤ -٢٠١٥ لدراسة الكثافة العددية لتربس البصل *Thrips tabaci* تحت الظروف الطبيعية لحقول البصل. تجاوزت الكثافة العددية لحشرات التربس الحي ١٠ حشرات / نبات في منتصف فبراير والذي بالتالي يصبح وقت للمكافحة. وتصل ذروة تعداد الحشرة في الأسبوع الثاني من ابريل. تم استخدام العزلتين الفطريتان الممرضتان للحشرات وهما فطريي *Beauveria bassiana* و *Metarhizium anisopliae* علي تربس البصل تحت الظروف المعملية بتركيزات 10^{4} ، 10^{5} ، 10^{6}، 10^{7} ، 10^{8} جرثومة/مللتر. ومن ثم كانت قيم التركيز المميت لنصف العشيرة هو 0.9×10^{5} و 2.9×10^{6} جرثومة/مللتر لكل من العزلتين الفطريتان *B. bassiana* و *M. anisopliae* علي التوالي. وبناء عليه تم رش العزلتين الفطريتان في حقل البصل لمكافحة تربس البصل بتركيزات 0.9×10^{6} و 2.9×10^{7} جرثومة/مللتر لكل من العزلتين الفطريتان *B. bassiana* و *M. anisopliae* علي التوالي وقورنت بالاكتليك ٥٠% كمبيد موصي به بتركيز ٥٠٠ مل/ فدان. الخفض في عدد الحشرات كان ٧٤ , ٦٢.٨٧ و ٨٧.٠٨ % لكل من العزلتين الفطريتان *B. bassiana* و *M. anisopliae* و مبيد الاكتليك ٥٠% علي التوالي . تكشف هذه النتائج ان العزلتان الفطريتان ربما تكون فعالة في مكافحة تربس البصل تحت الظروف الحقلية وخاصة عزلة *B. Bassiana*.