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Effect of *Trichoderma harzianum* in the Protection of Wheat Grains from Infestation with *Rhyzopertha dominica* (Fab) During the Store

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ABSTRACT: Currently, the economic losses generated by the attack of pests on the storages constitute a severe problem. Conventional grain protection methods based on the application of chemical pesticides to control these undesirable insects have also formed to represent an annoyance since the excessive usage of these compounds is linked to contamination problems and serious harm to consumers' health. This research aims to possibility of creating a means to protect the grain stores from infestation with *Rhyzopertha dominica* through testing for the efficiency of *Trichoderma harzianum* with three concentrations ($7x10^4$, $7x10^6$, $7x10^8$ and spores /ml) in the protection of grains in jute sacks from infection by this pest after three months of storage period and the effect of *T. harzianumon* . The results showed that the high efficiency of *T. harzianumon* in protecting the grain stores from infestation by these insect sacks was measured when treated in the bags with the highest concentration of 7x108 spores/ml of fungi, and the total weight loss was 2.0269. while the control treatment gave 6.4012-gram weight losses. also shows the total weight loss was 4.054 when the grains were treated with the highest concentration of 7x108 spores/ml of fungus. in contrast to 8.0629-gram weight decreases with the control treatment. and treated the bags and grains with 7x108 spores/ml of fungi, the total percentage of weight losses was 1.7033 after three months from storage. while the control treatment gave 11.475 grams of weight loss.

Keywords: *Rhyzopertha dominica*, Biological control Entomopathogenic fungi,

1. INTRODUCTION

The most important element of the human diet in the world is the grains, their production is seasonal and their consumption is endless. Thus, most of the products should be stored more safely, if the storage conditions are not appropriate the grains could be harmed by pests [1]. *Rhyzopertha dominica* (Coleoptera, Bostrichidae) considerable is one of the harmful pests of stored cereal grain mainly wheat, commonly named a lesser grain borer, which presents in warmer areas of the world. The impacts of pest *R. dominica* on some cereal during storage have been noted [2]. Stored product pests cause massive losses in agricultural items during storage. It is estimated that the losses may reach up to 9% in developed countries while up to 20% in developing countryside's [3]. The point that stored grains are utilized as human nourishment makes chemical management of stored-grain pests extremely risky Also, chemical pesticides damage the environment as well [4]. Entomopathogenic fungi are a class of microorganisms with several applications in biological control also utilized to create industrial enzymes and antibiotics as well as manage plant pests and numerous other applications [5]. *Trichoderma* spp. includes roughly 100 species distributed throughout the world that have been widely used as hostile fungal agents against many pests by enzymes and secondary metabolites, these systems are typical biological control [6]. *T. harzianum* is a widely accepted biological control agent against several pests. Entomopathogenic fungi are considered an environmentally safe control method compared with chemical pesticides. However, little research

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has been reported on the insecticidal action of these fungi against pests of storage [7]. This study aims to evaluate the biological control potential of *Trichoderma harzianum* against *Rhyzopertha dominica* in the store during 3 months of storage.

2. MATERIALS & METHODS

2.1. Collection *R. dominica*

The culture of *Rhyzopertha dominica* was collected from the public cereal storages and reared on wheat grains. Insects were carried in a container of plastic covered (10 x 25 Cm) with a clear muslin cloth. All insects were reared under conditions of 28 ±2°C and 75 ± 5% R.H.

2.2. Preparation of extract:

The fungus was obtained from the Department of Plant Protection, College of Agriculture, University of Duhok. and grown on a Petri dish (9 x 1.5cm) containing (PDA) Potato Dextrose Agar media. The media was autoclaved at 120°C for 20 minutes under sterilized conditions. The fungi were inoculated at 25- 30°C in the dark for 15 - days. Conidia spores were obtained with sterile distilled water containing Tween-80. Conidia spores were washed twice with sterile distilled water and centrifuged at 5000 revolutions/minute for 5 min. A haemocytometer was utilized to calculate the concentration of conidia spores, Sequent alleviation was then created to obtain the required concentration [8]. Three concentrations of spore's suspension (7x104, 7x106, 7x108 and spores /ml) from the entomopathogenic fungi, *T. harzianum* were prepared using the Neuberger counting chamber. All bioassay treatments were carried out in the incubator under a controlled temperature of 25±2 ºC.

2.3. Bioassay:

Jute bags were treated with fungal concentrations (7x104, 7x106, 7x108 and spores /ml) full with full of (50 grams) of wheat grains, grade (Ababa 99), obtained from the College of Agriculture - Department of Crops. After the grains were purified, cleaned of impurities, and placed in the freezer at -20°C for 48 hours to ensure that they were internally free of any insect infestation, As for the second treatment, the same steps were followed except that the grains were treated instead of the bags with concentrations of the fungal suspension, While the third treatment treated grains and packing bags with concentrations of fungal suspension, the bags were placed each concentration was placed in plastic containers with a diameter of (11) cm and a height of (25) cm, open on one side and covered with organza cloth to prevent the escape of the adults after they are released. 10 pairs (males and females) of newly emerged adults, 24 hours old, were released for each container containing one type of bag, the weight of the jute bags with grains was recorded after treatment [9]. And the weight of the bags filled with grains continued to be recorded monthly starting from the first month of storage.

2.4. Statistical Analysis:

The statistical program SAS (Statistical System Analysis) was used to analyze the data to study the effect of various parameters on the studied traits. The significant differences between the studied ratios or rates were compared using the least significant difference test at $P \leq 0.05$.

3. RESULTS AND DISCUSSION

The results revealed the effect of suspension fungal concentrations in the protection of grains bags in jute from infection by these pests during storage after three months, in Table 1 when treated the bags with the highest concentration of 7x108 spores/ml of fungi were, the total weight loss was 2.0269. while the control treatment gave 6.4012-gram weight losses. Table 2 shows the total weight loss was 4.054 when the grains were treated with the highest concentration of 7x108 spores/ml of fungus. In contrast, the 8.0629-gram weight decreases with the control treatment. Table 3 treated the bags and grains with 7x108 spores/ml of fungi, the total percentage of weight losses was 1.7033 after three months from storage. while the control treatment gave 11.475 grams of weight loss. This agrees with the study by [10]. when sprayed on feed (wheat grains) the fungi T. album causes mortality rates to increase to 94% within 7 days in adults with lesser grain borer. Likewise, the Trichoderma spore's application in stored grains can reduce their consumption by the adults of A. obtectus to 10%, due to its repellent activity against one of the primary storage pests involving stored common beans [11]. Also agree with [12]. studied the fungus extract of *T. harzianum* and showed high-efficiency mortality of *Tribolium castaneum* and *Diuraphis noxia* (76% and 73%). Many studies revealed the efficiency of strains of Trichoderma spp. utilized in insect biological control have good results against many pests such as *Lucanus cervus* order Coleopteran [13]. author order Lepidoptera *Leucinodes orbonalis* [14]. and order Diptera Delia radicum [15]. Entomopathogens fungi can infect and mortality arthropods, by breaking through the cuticle directly and requiring adhesions and lytic enzymes such as chitinases to penetrate chitin, lipases and proteases, after that, the fungi overwhelm the immune system of insects and colonize their bodies, forming and applying new conidia from their deathly host [16]. In previous research, [17]. found that Beauveria bassiana elevated effectiveness as grain protection provided control of stored-grain pests (Rhyzopertha dominica and Cryptolestes Ferrugineus) during 90 days of storage period. In a recent study, [18]. provide efficacy of the

entomopathogenic fungus Beauveria bassiana and Vuillemin, in protection of stored grains when it was infested of stored-grain pests Cryptolestes ferrugineus , Tribolium castaneum, Trogoderma granarium, and Rhyzopertha dominica and also studied combinations with DEBBM for 180 days.

Table 1. - The effect of a suspension of the fungus *T. harzianum* with concentrations of (7x104, 7x106, 7x108 and spores /ml) spores/ml on the loss percentage of wheat grains packed in treatment jute bags

Concentrations	Weight Month 1	Weight Month 2	Weight Month 3	Total
Control	2.592 a	2.1824 a	3.2878 a	8.0629 a
7x104	1.4925 b	1.6465 b	1.7034 b	4.8424 b
7x106	1.625 b	1.6942 b	1.2945 b	4.6137 b
7x108	1.6342 b	1.3942 b	1.0256 b	4.054 b
LSD	*0.794	*0.598	*0.984	1.621*
		* (P<0.05).		

Table 2. - The effect of a suspension of the fungus T. harzianum with concentrations of (7x104, 7x106, 7x108 and spores /ml) spores/ml on the loss percentage of treatment wheat grains packed in jute bags

Concentrations	Weight Month 1	Weight Month 2	Weight Month 3	Total
Control	1.8187 a	1.9841 a	2.5984 a	6.4012 a
7x104	1.2142 b	1.4826 b	1.6461 b	4.3429 b
7x106	0.9091 b	1.5642 b	0.7982 b	3.2715 b
7x108	0.7985 b	0.6802 b	0.5482 c	2.0269 c
LSD	*0.775	*0.833	*1.042	*1.788
		* (P<0.05).		

Table 3. - The effect of a suspension of the fungus *T. harzianum* with concentrations of (7x104, 7x106, 7x108 and spores /ml) spores/ml on the loss percentage of treatment wheat grains packed in treatment jute bags

Concentrations	Weight Month 1	Weight Month 2	Weight Month 3	Total
Control	1.3801 a	4.9522 a	5.1431 a	11.475 a
7x104	0.8441 b	0.8142 b	0.7012 b	2.3595 b
7x106	0.9892 b	0.7211 b	0.4293 b	2.1396 b
7x108	0.7920 b	0.5201 b	0.3912 b	1.7033 b
LSD	*0.621	*0.792	*1.012	*1.987
		* (P<0.05).		

4. CONCLUSIONS

In conclusion, the Trichoderma harzianum was effective in the Protection of Wheat Grains from Infestation against Rhyzopertha dominica during the Store, by treating the bags and grains compared with untreated control. This technique can effortlessly be adopted by farmers as a standard way of grain protection without using any chemical pesticides or commercial formulations. that can negatively affect the health of the consumer Also, entomopathogenic fungi would not degrade grain quality, Nonetheless, additional analyses are needed to determine the exact mode of action of the insecticidal components, and field-level experimentations are required to decide the effect in stores.

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