Using the fungus *Entomophthora muscae (chon) Fresenius* to eliminate some larval roles of *Musca domestica*

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Abstract

Studied effect serial concentrations from spores filtrate of fungus *Entomophthora muscae* on some larvael roles of *musca domestica* in laboratory. Results were made clear that the insect roles are sensitive to fungus,and treated the food larva of *musca domestica* and sprinkle it by concentration 2.8×10^6 , 2.8×10^7, 2.8×10^8 (spore/ml) has led to get rates of destruction of cumulatire faculty certified on the concentration and time its magnitude 16.60 , 47.67, 53.30 % respectively , also recorded some phenotypic distortion infected dead larva represent by contraction and blackening body. The treatment of pupael by sprinkling the previous fungus concentration recorded rate of destruction of accumulative faculty its magnitude 13.33, 26.67, 33.33% respectively, also the rates emergence of adults ranged between 66.67 – 86.67 % in comparison with rates of emergence of adults in control treatment 96.67% The results are made clear that adults treatment by sprinkle with last concentration from fungus spore filtrate recorded rates of distraction its magnitude 46.61, 56.67, 70% respectively after one week from treatment .

Keywords: fungus, *Entomophthora muscae* , *musca domestica*, larvae, pupae.

Introduction:

The insect *Musca domestica* belong to family muscidae from order Diptera, and its hazardous on humans because of its spread in landfill and transfernce the diseases to human and animals (1,2) . More studies proved that the *musca domestica* the main transmission to large number of diseases causes such as bacteria and viruses (3,4) .Severl chemical pesticides were used to combat the insect *musca domestica* such as D.D.T. and carbamate pesticides (5). These pesticides fased resistance from insect by time (6).The fungus *Entomophthora muscae (chon) Fresenius* is one of some fungi that uses for biological combat against *Musca domestica*, it is characterized by its speed killing to flies and is observed glued on the walls and glasses by fungi filaments (7). The adult roles of *musca domestica* vulner able to fungus, it kills the fly after 4-6 days after exposure to conidia that come out of bodies of the fly infected (8). The fungus causes clear marks when infected the *musca domestica* such as swelling of the abdomen, legs spread, high wings above the chest, hose expansion and exist of white conidiophora on membrane between abdomen segment (9,10), where the fungus work on punch the cuticle and form the fungal hypha, Its reproduction happens through 28 hour, after that it invades the fly fully specially abdomen and fat bodies (11). The fly males attracted highly to infected females because of the large abdomen that serves as an instrument to attract male (12) .This study aims to use biological peticides for eliminate on *Musca domestica*.In all larval roles of it life, and stay away from chemical peticides that it have remaining effect in environment.

Materials and methods:

For seven day. After that the growth colony are cultured again on medium PDA for purification . The fungus are identify by appearance and microscopically identification on taxonomical keys (13) .

2.Insect breeding:

Numbers of adults *Musca domestica* collected from garbage places and put in breeding cages are designed to the form of a parallel rectangles the dimensions of (40×35×40) cm , wooden base and the four side coverd with cloth tulle while the upper surface covered with glasses. Adult are fed

by using cotton wet water and milk powder in petridish a rate of two petridish for each cage , then the eggs collected and transferred to glass pots contain on the artificial medium to breeding larvae contain from 60gm fertilizer animal, 10gm sugar barley and 5gm yeast put in another breeding cages and are followed up even the role thus the farm pured to three stages before holding the expirements .

3.Preparation of fungus filterates:

Add 5ml from distilled sterile water to fungus culture with age seven days the spores separation by glass rod have L letter shape, nominated content dish by funnel contain a piece of sterilized tulle, the filterate collected in glass flask and this filterate was the stock solution and to account the number of spore in filtrate used the Haemocytometer, transferred 1ml from stock solution 99ml sterile distilled water for mitigation and count then put 0.1 ml of it on Haemocytometer account number of sporesin five internal squares then use these equation (14)

Number of spores = rate number of spores × coefficient of mitigation × 25×10^4 (spore/ml)

Found that rate of spore equal to 2.8×10^9 (spore/ml) and to preparation concentration of fungus filtrate for study 2.8×10^6, 2.8×10^7, 2.8×10^8 (spore/ml) used this equation

V1 C1 = V2 C2

V1 =required size from water for used to treatment ,V2=required size from stock solution to add to V1

C1=the concentration required to find, C2=spores concentration in stock solution

4.Study the effect of fungus spore stuck in roles of *musca domestica*

Concentration made about 2.8×10^6, 2.8×10^7, 2.8×10^8 (spore/ml) from fungus spores stuck of *Entomophthora muscae* to study its effect in roles of *musca domestica* , first transferr10 larvae of *musca domestica* to cylinder plastic pot basic diameter 4.5 and high 3.5 cm , after that the larvaes sprinkle about 2ml from spores fungus stuck with concentration 2.8×20^6 (spore/ml) by hand sprinkler direct sprinkle off 10-15cm ,after treatment larvae's are transferred to another pot contain 10gm from nutrition medium benefit for larvae's growth that treated with 2ml from the same fungus spores stuck concentration while the control treatment the larvae's and medium was sprinke by 2ml of distill water. The pot covered with plastic hoods perforated for breathing larvae's, then make six repetitions for concentration and three for control. The experiment repeated to the other concentration, pots incubated in incubator in27°c and 12 hours lighting the dead and distorted larvae's were observed to account the rates of destructions and distortion , rates of pupae's distortions, rates of adults emergence (15) while for the pupaes of *musca domestica* to pupaes transferred from breeding colony in age 24 hours to plastic pot its capacity about 100ml padded by filter paper from type what man N01 .The pupaes sprinkle by 2ml from fungus spores stuck in concentrations 2.8×10^6 (spore/ml) then pot covered with plastic hoods perforated and then made six repeaters and then experiments repeated by use fungus spore stuck with two concentration 2.8×10^7 , 2.8×10^8 (spore/ml) while control treatment the pupae sprinkle by 2ml distill water , pots incubated in incubator in 27°c and 12hours lighting, the changes follow up from distortion , destructions and adults emergence (16).For adults the cage contain adults fly put in deep-freeze for two minute to reduce it movement, 10adults (male and female) were transferred to plastic container its capacity 120ml (high 7.5cm, diameter 4.5cm) padded with filter papper from type what man N01, sprinkle with 4 ml of fungus spore stuck in concentration 2.8×10^6 (spore/ml) the containers covered with cloth tulle link by bond of rubber put upper of it cotton saturated with sugar and water by rate 10% for adults feeding , the treatment repeated six times ,while control treatment the filter papers treated with sterilized distill water this work repeated three times and the whole experiment repeated to the other concentration after that the containers incubated in incubator in degree 27^0c and lighting 12 hours and recorded the rates of adults destruction every 24 hours (16)

Results :

Results shown in table (1) the treatment of larvae food and sprinkle it by fungus concentration 2.8×10^6, 2.8×10^7, 2.8×10^8 (spore/ml does not cause destruction after two days passed of treatment .The reason belongs to alienation of larvae and turned to the third larval role , thus the larvae gets rid from fungus spore that stick-on larvae , thus unable to enter inside the body , but some cumulative destruction were recorded by concentration 2.8×10^7, 2.8×10^8 (spore/ml) respectively, also the table (1) show the rates of destructions cumulative debugger (larvae and pupaes) which increased with the increase of used concentration about 16.60, 46.67, 53.30% when treatment in concentration 2.8×10^6, 2.8×10^7, 2.8×10^8 (spore/ml) respectively, also the results show rates of adults emergence inversely proportional to the used concentration about 83.33, 53.33, 46.67% respectively when treatment with upper concentration in compared with control treatment the adults emergence about 96.67%

Table (1) the effect of serial concentrations of fungus spore stuck E.musca on larvaes of Musca domestica .

Concentration (spore/ml)	Debugger distortion error after 24hours	%destruction of cumulative ±standerd error	%distortions ±standerd error	% Debugger distortion cumulative total (larvaes +pupes)± standard error	%adults emergence ±standard error
Control treatment	0.00±0.00	0.00±0.00	0.00±0.00	3.33±1.35	96.67 ±3.33
2.8×10^6	0.00±0.00	0.00±0.00	0.00±0.00	16.60 ± 3.16	83.33±3.33
2.8×10^7	0.00±0.00	16.67±16.6	13.33±6.00	46.67 ±26.67	53.33±26.67
2.8×10^8	0.00±0.00	33.33±16.67	6.67 ± 3.33	53.30 ± 14.54	46.67±12.01
LSD	NS 0.00	NS 47.69	NS 28.78	31.02	NS 58.81

N.S. meaning non- significant differences * meaning significant differences rates possibility ($p \leq 0.05$) on less significant difference.

The results of treatment of *musca domestica* pupaes in serial concentration of fungus spores stuck *E.musca* . Showed in table (2), the rate of destruction of cumulative directly proportional with the used concentration. The rate was 13.33, 26.67, 33.33% when treated with concentration $2.8*10^6$, $2.8*10^7$, $2.8*10^8$ (spore/ml) respectively. While the destruction rates in control treatment about 3.33% only. The table shows clear record rates of distraction in dead pupae represented by it elongation also some states of partial emergence for adults appeared.

Table (2) effect serial concentration of fungus spore stuck E. muscae on pupae of musca domestica

Concentration (spore/ml)	%distortion of accumulative ±standard error	% destruction ±standard error	%adult emergence ±standard error
Control treatment	3.33 ± 1.17	0.00±0.00	96.67 ±3.33
2.8×10^6	13.33±3.33	6.67 ± 3.33	86.67 ±3.33
2.8×10^7	26.67 ±8.82	3.33 ± 1.17	73.33± 8.82
2.8×10^8	33.33± 8.82	3.33 ± 1.17	66.67 ± 8.82
LSD	* 21.74	NS 9.41	* 21.74

N.S. meaning non - significant differences * meaning significant differences rates possibility ($p \leq 0.05$) on less significant difference .

Table (3) showed that results of treatment of *musca domestica* adults by direct sprinkle of serial concentration from fungus spores stuck of *E.muscae* , The deads rate after 24 hours from treatment was 33.33, 16.67, 26.67 when treated by concentration $2.8*10^6$, $2.8*10^7$, $2.8*10^8$ (spore/ml) respectively. While the distortion rate in control treatment only 3.33% only while after one week on treatment the distortion rate increasesd to 46.67, 56.67, 70.00% respectively. While the distortion rate in control treatment 10%.

Table (3) effect serial concentration of spores fungus stuck of E.muscae on adults of musca domestica .

Concentration (spore/ml)	%distortion after 24 hours ±standard error	%distortion after one week ± standard error
Control treatment	3.33 ± 1.17	10.00± 5.0
2.8×10^6	12.02±3.33	46.67 ± 14.52
2.8×10^7	16.67 ± 8.82	56.67 ± 14.52
2.8×10^8	26.67 ±8.82	70.00 ± 5.77
LSD	* 28.76	38.43

N.S. meaning non-significant differences * meaning significant differences rates possibility ($p\leq0.05$) on less significant difference.

Discussion:

Results in table (1) . The dead of these larvae belong to presence of fungus spores in food that cause repellent effect to feeding larvaes subsequently reduce from it consumption to food led to starve the larvae's and reduce its activity ,also the fungal growth causes secretion of enzyme and toxic compounds that analyzed body organs. These results agreed with (17), that larvae's of *Musca domestica* that treated with concentration 2.3×10^8 spore \ml with fungus spore stuck appeared idle and infection larvae shrunked and have heavy colour , also agreed with (18) that larvae's of insect *Agrotis ipsilom* sensitive to fungus spores stuck *M.anisopliae*. Also these results agreed with (19) that fungal infection and growth inside host body lead to secretion of toxic compound and fungal hypha and result tearing and damage body organs esspecilly digestive subsequently effected in amount of food consumption of larvaes.The results of treatment of *Musca domestica* pupaes in serial concentration of fungus spores stuck *E.musca* . appeared in table (2). The table shows clear record rates of distraction in dead pupae represented by it elongation also some states of partial emergence for adults appeared .The reason of dead and distruction to fungal infection that lead to drain internal tissue for pupae also led to prevent emergence of complete insect or dead insect inside pupaes envelope. Also the adult that completed the emergence from envelope were dead after few days comparative with control, the reason is that infected with fungus during out from pupaes envelope that with fungus spores , these results agreed with (20) that treatment pupae of *spordoptera litura* belong to pollution with in concentration 10^8 (spore/ml) with fungus spore stuck of *M.anisopliae* led to get rate of distortion about 85.8.Table (3) showed that the results of treatment of *Musca domestica* adults by direct sprinkle of serial concentration from fungus spores stuck of *E.muscae*, While the distortion rate in control treatment 10% the resean of adults dead belong to growth of fungus inside insect body that led to tearing organs and body addition to secretion enzyme and toxic from fungus that led to decompose and death the tissue and insect this results agreed with (21) that adults flies of Mediterranean *Ceratitis capitata* sensitive to fungus *M.anisopliae* by concentration 10^6 (spore/ml) caused 100% distortion.

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