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"Studies on some entomopathogenic fungi isolates as bio-control agent against certain piercing sucking pests"

A THESIS

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SUMMARY AND CONCLUSION

Schizaphis graminum (Rondani) and *Tetranychus urticae* Koch are considered as dangerous serious pests all over the world; because they have a wide host range, and they transmit plant pathogenic viruses. As a result of wide spread application of chemical control and their harmful impact of natural enemies and environment, there was a necessitate for using a modern types of insecticides which have different modes of action that are effective, much safer to human and ecosystem and could be useful as alternatives for the integrated management approach. Entomopathogenic fungi are considered most important elements of microbial control agents.

The present study aimed to studying the following points:-

1. Introduce some natural microorganism, like entomo-pathogenic fungi as alternative trends of pesticides.
2. Identify the isolated entomopathogenic fungi at least at the generic level.
3. Determine the pathogenicity of a native isolated entomopathogenic against adult stage of *S. graminum* and *T. urticae*.
4. Evaluate insecticidal activity of isolates metabolites against target pests.
5. Study mode of action of entomopathogenic fungi.

6. Compared virulence of isolated entomopathogenic fungi and commercial bio-insecticide to conventional insecticides control under field condition.

1. Isolation and identification of fungi:

Studies were carried out at some locations and crops to search occurrence of entomopathogenic fungi in Egyptian soils by using *Galleria* bite method. The obtained results revealed the presence of four entomopathogenic fungi were *B. bassiana* and *M. anisopliae* isolates (B1, B2, M1 and M2) and non entomopathogenic isolates.

2. Virulence of fungi isolates:

All isolates (B1, B2, M1 and M2) have pathogenicity against adult stages of *S. graminum* and *T. urticae*. Mortality% reached its maximum after 7 days.

The LC_{50} value of B1 was 3.11×10^{6} spores/ml while M1, M2 and B2 revealed greater LC_{50} values (6.09×10^{6}, 2.32×10^{7} and 1.15×10^{8}) spores/ml, respectively against aphid. The data showed that isolate B1 caused high mortality in shortest time, LT_{50} value was 3.04 days. While for the other isolates M2, B2 and M1 the LT_{50} values were 3.37, 3.70 and 3.95 days, respectively. No significant between isolates against *S. graminum.*

While against *T. urticae*. B2 was most effective, LC_{50} for B1, B2, M1 and M2 were (4.9×10^{8}, 2.86×10^{7} 2.75×10^{9} and 4.58×10^{8}) spores/ml, respectively. The obtained results showed that isolate B2 caused high mortality in shortest time, LT_{50} value was 3.57 days. While, for the other isolates M2, B1 and M1 the LT_{50} values were 9.24, 10.16 and 23.17 days. Significant levels were between isolates.

3. Evaluation insecticidal activity of metabolites

The results shown that four tested metabolic crudes were toxic to *S. graminum* and mortality percentage increases by the increasing concentration of crude extract and time. The highest effective toxin was of (M1) followed by (B1), (M2) and then (B2).The mortality at the sixth day of concentration 100% crude/ml were 92..04, 73, 63.3 and 64.83 % for M1, B1, B2 and M2, respectively. LC_{50} value of M1 was 23.48 crude/ml while B1, M2 and B2 revealed greater LC_{50} (80.45, 105.39 and 133.5) crude/ml, respectively.

Also, four tested metabolic crudes were toxic to *T. urticae* and mortality percentage increases by the increasing concentration and time of crude extract. Also, the highest effective toxin was of M1, followed by B2, M2 then B1.The mortality at seventh day and for concentration 100% crude/ml were (85.16, 76.12, 73.81 and 61.79) % for M1, B2, M2 and B1,

respectively. The LC_{50} value of M1 was 45.67crude/ml while M2, B2 and B1 revealed greater LC_{50} value, 50.92, 62.47 and 101.65 crude/ml, respectively.

Generally, the highest values of mortality were in the M1 treatment. Indicated that were significant levels of effect between different fungal crude M1, M2, B1and B2 and between concentration within days and treatments on two pests.

4. Scanning electron microscope

Scanning electron microscopy are convenient tools allowed us to observe adhesion and penetration structures *S. graminum* infected with *B. bassiana* and *M. anisopliae.* The results of SEM described the development cycle of fungus on *S. graminum*. Conidia were adhered and germination started on surface forming special structural appressoria and germ tube witin 24hours post-infection. The phase of host colonization occurred between 72 and 120 hours, and most of the aphid died between 72 and 120 hours after inoculation. Both *B. bassiana* and *M. anisopliae* are able to colonize and infect the *S. graminum*, finally death of infected aphid.

5. Experiment of Field

The present study tested direct spray effect of treatments (Malathion, *B. bassiana, M. anisopliae,* Biossiana and Bio-Meta) against cereal aphidas compared with untreated control on wheat

crop. The higher effect was obtained by the treatment Malathion 57%. The two tested fungi *B. bassiana* (B1) and *M. anisopliae* (M1) were effective in controlling cereal aphid at concentration of 1×10^8 spores/ml reduced aphid population from third day until 10 days of treatment. Commercial bio-insecticide; Biossiana and Bio-Meta also, reduced aphid population. It is clear that all tested materials were highly significantly effective as compared with control.

Therefore, it could be concluded that using such entomopathogenic fungi (*B. bassiana* and *M. anisopliae*) and its botanical extracts (secondary metabolites) fungi had strong insecticidal activities against cereal aphid and two spotted spider mite. The insecticidal activities were time and dose-dependent and may be useful in a manner for reducing the hazards and harmful effect of that pests Also, the study attempts to applied Entomopathogenic fungi in open field condition. Therefore, the study attempts to elucidate if it is possible to rationalize the use of such tested materials via integrated pest management (IPM) programs to control cereal and mite.