ISOLATION, IDENTIFICATION AND CONTROL OF PATHOGENS INFECTING THE MULBERRY SILKWORM, Bombyx mori L.

By

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Department of Plant Protection Faculty of Agriculture Ain Shams University

Approval Sheet

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ABSTRACT

Eman Ali Abd-Elrazek Abd-Elfatah: Isolation, Identification and Control of Pathogens Infecting the Mulberry Silkworm, *Bombyx mori* L. Unpublished Ph.D. Thesis, Department of Plant Protection, Faculty of Agriculture, Ain Shams University, 2022.

Mulberry silkworm, *Bombyx mori* is infected by many diseases, these diseases cause huge losses in the resulting crop of cocoons, and thus silk production. The present study was conducted in order to isolate and define fungal and bacterial pathogens both phenotypic and genotypic. Pathogenicity test and the influence of some natural and chemical compounds to control these pathogens were also conducted.

The classification of different fungal isolates was adopted according to their cultural and morphological properties. These isolates were identified as *Aspergillus fumigatus* (SW1) and *Aspergillus flavus* (SW2).

The pathogenicity test proved that artificially inoculated *A. fumigatus* and *A. flavus* to healthy silkworms showed typical symptoms of Aspergillosis. Young larvae infected with *A. fumigatus* and *A. flavus* separately died within few days, the body was hardened and completely covered with yellow fungus spores. When older larvae were infected with the fungus, black spots appeared on different areas of the body. The survived infected larvae spun their cocoons and died inside. Infected pupae produced deformed moths.

It was found that the fungal infection in the summer season was more dangerous to the larvae compared to the spring season. Also, the young larval instars were more sensitive to infection with fungi than the older larval instars, where the death rate reached 100%.

Treating larvae with fungicide (Actamyl), salicylic acid, propolis powder and henna leaves powder after being inoculated with *A. fumigatus* and *A. flavus* separately, reduced the larval mortality percentages and

increased larval survival compared to the control larvae. The lowest percentage of mortality (8.3%) was recorded when 15% of henna was used, 12 hrs. after inoculation with *A. flavus*.

Three different bacterial isolates (SW3, SW4 and SW5) were obtained from the silkworm larvae infected with the bacterial flacherie disease. The isolates were identified based on cultural, morphological, and biochemical characteristics and phylogenetic analyses performed using the 16S rRNA gene sequence. The isolates were identified as *Escherichia coli* (SW3), *Staphylococcus sciuri* (SW4) and *Serratia rubidaea* (SW5).

The pathogenicity test proved that artificially inoculation of third instar larvae with *E. coli*, *Staph. sciuri* and *S. rubidaea* separately, showed typical symptoms of bacterial flacherie disease. Larvae lost their appetite, had slow movement, and severe vomiting, then larvae died within few hours.

Antibiotics (Ibiamox and Garamycin) and Kombucha extract *In vitro* using agar well diffusion assay proved effective against *E. coli*, *Staph. sciuri* and *S. rubidaea*, Olive oil was effective against *Staph. sciuri* only.

Applying Ibiamox and Kombucha extract to control bacterial diseases *In vivo* reduced the percentage of larval mortality. The lowest percentage of mortality recorded when larvae were fed on antibiotic-treated mulberry leaves 24 hrs. after inoculation with *S. rubidaea* (4.3%), followed by *E. coli* and *Staph. sciuri* (8.7%). The percentage of mortality decreased to 8.7% when larvae were fed with mulberry leaves treated with Kombucha 24 hrs. after inoculation with *E. coli*, and reached 21% in larvae inoculated with *Staph. sciuri* and *S. rubidaea*.

Keywords: Mulberry, Silkworm, Bombyx mori, Aspergillosis, Aspergillus fumigatus, Aspergillus flavus, Escherichia coli, Staphylococcus sciuri and Serratia rubidaea.

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