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VI. SUMMARY

In the present study, the infectivity of the two entomopathegnic nematodes, *Steinernema riobrave* and *Heterorhabditis* sp., together with the entomopathegnic fungus, *Beauveria bassiana* on the cotton leafworm, *Spodoptera littoralis* was studied. Moreover, the effect of these pathogens on certain biochemical and physiological aspects of the host was also studied. *Heterorhabditis* sp. appeared to be more pathogenic than *S. riobrave* to the *S. littoralis* larvae, especially at the lower concentrations. Mortality rate of larvae treated topically with the conidia of *B. bassiana* increased with increasing the concentrations.

When *S. riobrave* and *Heterorhabditis* sp. were mixed together and applied to the host larvae either simultaneously or sequentially, synergism (or potentiation) was obtained, in terms of larval mortality. This is also true when either *S. riobrave* or *Heterorhabditis* sp. was mixed with the fungus, *B. bassiana*. The highest potentiation was obtained when *S. riobrave* and *Heterorhabditis* sp. were applied sequentially against *S. littoralis* larvae, applying the former nematode firstly for 24 hr. and then applying the latter nematode for also 24 hr, where the larval mortality was 100%.

The infective juvenile production was also affected due to the binary mixture between the nematodes and the fungus. The highest production (5750 infective juveniles) was obtained with the sequential application between *B. bassiana* and *S. riobrave*. The fungus,in this case was applied firstly, and then followed by the nematode. In contrast, non of the infective juvenile was produced when *S. riobrave* was applied

sequentially with *B. bassiana*, applying the nematode firstly, followed by the fungus.

Host plant affected also the Pathogenicity of both nematodes and fungus, and progeny production of the nematode. The lowest mortality was observed in larvae fed on cabbage leaves and infected by the two nematode species. This also accompanied by the lowest progeny production. Whereas, larvae infected by the nematodes and fed on castor leaves produced the highest progeny. In general, *Heterorhabditis* sp. was more pathogenic than *S. riobrave* to *S. littoralis* larvae fed on the five tested host plants (cabbage, castor, mellow, Jew's mallow and cowpea leaves). The reverse was true with the progeny production, where infective juvenile production from cadavers infected with *S. riobrave* was higher than that produced from cadavers infected with *Heterorhabditis* sp. fed on the five host plants. As the fungus, *B. bassiana*,

the highest pathogenicity was obtained for larvae fed on cowpea leaves, whereas larvae fed on castor leaves were highly tolerant to the infection by the fungus.

Food consumption and utilization in *S. littoralis* larvae were also affected due to infection by the two nematode species and the fungus tested, particularly in the 6^{th} instar. Although food consumption increased in infected larvae as compared to non-infected ones, the relative growth rate of food utilization decreased (in terms of the efficiency of conversion of ingested, ECI and digested food, ECD, into biomass).

The decrease in ECI and ECD and meanwhile the increase in food uptake from the gut, in terms of approximate digestibility, indicate that the interference of the three pathogens tested with the process of nutritional metabolism may be occurred after digestion.

The principale nutrients (total protein, carbohydrate and lipid) of the host larvae were highly decreased post-infection with the nematodes *S. riobrave* and *Heterorhabditis* sp. and the fungus, *B. bassiana.*

The activity of some larval enzymes was also affected due to infection by these pathogens. Thus, the activity of carbohydrate hydrolyzing enzymes (amylase, invertase and trehalose) changed depending on the species of the pathogen and the enzyme. Amylase activity decreased with the infection by *Heterorhabditis* sp. and *B. bassiana*, and the reverse was obtained with the infection by *S. riobrave* where such activity increased. Invertase activity increased with the infection by two nematode species, and the reverse was true with then infection by the fungus. However, trehalose activity was highly increased following infection by both the nematodes and fungus tested. Activities of acid and alkaline phosphatases increased due to infection by *S. riobrave*, *Heterorhabditis* sp. and *B. bassiana.* The only exception was a non-significent decrease in the alkaline phosphatase activities of larvae infected with *B. bassiana.* Whereas, the activity of transaminases (GOT and GPT) was highly decreased with the infection by the three pathogens tested.

SDS-PAGE analysis revealed the disappearance and appearance of some protein bands in *S. littoralis* larvae that were infected with *S. riobrave, Heterorhabditis* sp. and *B. bassiana.* The most obvious feature

was the disappearance of the slow moving protein bands (171.42, 154.47, 146.92 and 127.38 KDa) in all infected samples.

The correlation between the primary metabolites of host plants and the susceptibility of the host larvae to the nematodes and fungus, together with the infective juvenile production were discussed. In addition to, the role of acid phosphatase in defence mechanism of infected ,larvae together with the nematodes nutritional requirements.