Department of Entomology
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Improving Production and Potency of Bio-insecticides based on Entomopathogenic Nematodes

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BY

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Approval Sheet

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ABSTRACT

In the present work, two native isolates of entomopathogenic nematodes (EPNs), (*Steinernema carpocapsae* BA2, *Heterorhabditis bacteriophora* BA1) were studied in comparison with several worldwide species for their suitability for improving *in vivo* and *in vitro* solid mass production techniques. They were also studied for foliar applications against the cotton leaf worm *Spodoptera littoralis* and the black cut worm *Agrotis ipsilon* infesting corn plants with suitable adjuvants in laboratory, pot experiments and field standards.

For mass production of *Galleria mellonella*, the host for *in vivo* production of EPNs, four economical artificial diets were suggested for mass rearing of *G. mellonella* and compared to the natural food (bee-wax). The cost of diets, numbers, weights, food consumption and lipid content of *G. mellonella* larvae (last instars) reared on each diet were examined. *G. mellonella* larvae that were reared on different diets were tested, in turn, for their suitability of mass production of two Egyptian nematode isolates, *S. carpocapsae* BA2 and *H. bacteriophora* BA1. Two diets out of four were recommended for the best economic and effective mass production for *G. mellonella* as host larvae for EPNs.

Two Egyptian isolates of EPNs were studied for their suitability for improving *in vitro* production on both agar plates and in Bedding flasks in comparison to other worldwide EPNs (*Steinernema riobrave* and *Heterorhabditis marilatus*). In general, all strains were successfully propagated on the tested solid media. Quantitative evaluation has been carried out through nematode yielded number. This was the first trial to propagate *S. carpocapsae* BA2 on different artificial media. Results also showed that both bacterium and nematode inoculum size are important for optimizing the final yield in the production of *S. carpocapsae* BA2. Four virulent worldwide nematode species viz, *S. riobravae, Steinernema scaptarasci*, *Steinernema* sp. and *H. marilatus* were successfully cultured on a large scale using Bedding three-dimensional

culture technique. *S. riobrave* achieved the highest offspring production on the two tested media.

Culture methods effects (*in vivo* Vs. *in vitro*) on virulence and propagation ability of EPNs (*S. carpocapsae* BA2 and *H. bacteriophora* BA1) were carried out. *In vivo* produced nematodes showed almost the same virulence and propagation ability as *in vitro* produced ones.

Aiming at improvement of the Egyptian EPNs efficacy (*S. carpocapsae* BA2 and *H. bacteriophora* BA1) for foliar application against two of important foliar pests (*S. littoralis* and *A. ipsilon*) infesting corn plants. The effect of adding some of potential adjuvants "surfactants, polymers and oil" to sprays containing infective juveniles were examined in the laboratory on foliage against fourth larval instar of both *S. littoralis* and *A. ipsilon.*

For *S. littoralis,* some adjuvants (surfactant-polymer formulation) SPF significantly increased the nematode efficacy and thus increased % mortality of the pest. However, for *A. ipsilon,* data showed no significant differences between treatments used.

In field experiment, one of the treatments was chosen due to its available cheap constituents (Carboxymethyl cellulose (CMC) + tween 80 + paraffin oil). This treatment significantly increased the effect of both *S. carpocapsae* BA2 and *H. bacteriophora* BA1 against *S. littoralis* larvae. In case of *A. ipsilon* larvae, the same treatment significantly increased performance of *H. bacteriophora.*

Key words: *Galleria mellonella, Steinernema carpocapsae, Heterorhabditis bacteriophora, Steinernema riobrave, Heterorhabditis marilatus, Steinernema scaptrisci*, mass production, formulation *Spodoptera littoralis, Agrotis ipsilon*. surfactant-polymer formulation.

CONTENTS

LIST OF ABBREVIATIONS

EPNs	Entomopathogenic Nematodes
IJ(s)	Infective Juveniles
DJ(s)	Dauer juveniles
FC	Food consumption
CI	Consumption index
g	Gram
Kg	Kilogram
M	Medium
T	Treatment
Dist.	Distilled
Hrs.	Hours
cm	Centimetre
ml.	Millilitre
L	Litre
µl	Micro litre
TTC	Triphenyl Tetracolium Chloride
UV	Ultra violet
DBM	Diamond back moth
CIB	Cereal leaf beetle
CMC	Carboxy methyl cellulose
TCA	Trichloroacetic acid
NBTA	Bromothymd blue-agar
SPF	Surfactant polymer formulation
BSA	Bovin serum albumin

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