

Prospects of Microbial Larvicides for Mosquito Control Under Field Conditions

A THESIS

Presented to the Faculty of Science
Ain Shams University
for the award of the
Ph . D. Degree

595.771
A.K

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(1987)

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«وَقُتِلَ أَعْمَلُوا تَفْسِيرِي اللَّهِ عَلَيْكُمْ وَرَسُولِهِ وَالْمُؤْمِنُونَ»

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Special appreciation is due to the Research and Training Center on Vectors of Diseases for providing all the facilities needed during the study.

This investigation was fully supported financially by the World Health Organization (WHO) through a research project entitled "Entomological operational evaluation of formulation of Bacillus thuringiensis H-14 and Bacillus sphaericus as biological larvicides against mosquito vectors in Egypt" TDR program.

CONTENTS

I- Introduction	1
II- Literature Review	2
A- <u>Bacillus sphaericus</u> Neide and its pathogenicity to mosquito larvae	2
B- <u>Bacillus thuringiensis</u> H-14 and its pathogenicity to mosquito larvae	15
C- Factors affecting the activity of the microbial agents	29
III- Materials and Methods	36
Phase I : Laboratory investigation	36
A- Rearing technique of tested mosquito species	36
a- <u>Culex pipiens</u>	36
b- <u>Anopheles multicolor</u>	37
c- <u>Aedes caspius</u>	38
B- Larvicides tested	38
a- <u>Bacillus thuringiensis</u> H-14	38
b- <u>Bacillus sphaericus</u>	38
c- Fenithion	38
d- Dieldrin	39
C- Bioassay experiments	40
a- Orientation susceptibility tests	40
b- Susceptibility of <u>Culex pipiens</u> larvae to bacterial larvicides	40
c- Comparative larvicidal action of different larvicides during five days post- treatment	41
D- Factors influencing larvicidal action	42
a- Types of breeding water	42
b- Exposure to direct sun light	42
c- Interaction between chemical and bacterial larvicides.	43
E- Factors influencing <u>Bacillus sphaericus</u> larvicide action on <u>Culex pipiens</u> larvae	43
a- Effect of Crowding	43
b- Recycling of <u>Bacillus sphaericus</u> inside dead larvae	44

Phase II :- Field Studies	45
A- Small-Scale trials	45
a- Selected areas	45
1- El- Kashish village	46
2- El Gabal El- Asfar	46
3- Wadi El-Natroun	46
4- El-Shikha Salma	46
b- Materials used in the specification of breeding places.	47
c- Spraying equipments	48
d- Methods of applications	50
e- Sampling technique	53
i) Dipping technique	53
ii) Netting technique	54
B- Large-Scale trials	55
a- Collection of base-line data	56
i) Human baited trap	57
ii) Animal baited trap	57
b- Method of application	58
IV- Results	59
Phase I: Laboratory investigations	59
A- Screening the susceptibility of different mosquito species to bacterial and chemical larvicide	59
B- The susceptibility level of <u>Culex pipiens</u> larvae to bacterial larvicides	62
C- Comparative duration of toxic bacterial and chemical larvicides	65
D- Factors influencing larvicidal action	68
a- Types of breeding water	68
b- Exposure to direct sun light	73
c- Interaction between bacterial and chemical larvicides	78
E- Factors influencing <u>Bacillus sphaericus</u> larvicidal action on <u>Culex pipiens</u> larvae	82
a- Effect of crowding	82
b- Recycling of <u>Bacillus sphaericus</u> inside dead larvae	82

Phase II: Field Studies	88
A- Small-Scale trials	88
a- Testing larvicidal activity of periodically collected water samples from sprayed natural breeding places	88
1- El-Kashish village	88
2- El- Gabal El-Asfar	92
3- Wadi El-Natroun	95
4- El Shikha Salma	101
B- Mass application trials	105
1- At Wadi El Natroun	105
a- Abundance of mosquito species	105
b- Lethal effect of <u>Bacillus thuringiensis</u> H-14 on mosquito larvae	109
c- Effect of <u>Bacillus thuringiensis</u> H-14 on adult population as a result of mass application of larval breeding places.	115
2- At El Shikha Salma	121
a- Abundance of mosquito species	121
b- Larvicidal application at El Shikha Salma	124
c- Effect of <u>Bacillus thuringiensis</u> H-14 on the reduction of adult populations	128
V- Discussion	132
VI- Summary	147
VII- Literature Cited	150
VIII- Arabic Summary	

INTRODUCTION

INTRODUCTION 1

The failure of chemical pesticides to continue their control of insect disease vectors has caused scientists to turn their attention to search for alternative methods.

Due to the continued price increase of insecticides, vector resistance and the public concern about environmental contamination, anti-larval measures using biological agents are now being encouraged, either alone or in integrated control programmes against mosquito vectors.

Among the most promising biological agents for mosquito control are Bacillus thuringiensis H-14 and Bacillus sphaericus.

The present investigation aimed at the evaluation of these biological agents under natural field conditions.

Preliminary experiments were carried out under laboratory conditions to study the effect of some factors which might affect the efficacy of these biological agents before applying them in the natural breeding places.

The second aim of this study was to test these biological agents on small-scale field trials to evaluate their activity and persistence under natural conditions.

The last step was a large-scale mass application to study the effect of these larvicides on the reduction of newly emerged adults that survived larval treatment which are the stage responsible for disease transmission.

LITERATURE REVIEW

A. *Bacillus sphaericus* Neide and its pathogenicity to mosquito larvae :-

Kellen et al. (1965) isolated a strain of *Bacillus sphaericus* Neide from moribund fourth-instar larvae of *Culiseta incidens* (Thomson) collected near Fresno, California.

The bacterium was a gram-variable, aerobic, spore-former and was facultatively parasitic to mosquitoes in laboratory tests. Tests were performed with larvae of 10 species of mosquitoes, all of which were susceptible to the bacterium.

Davidson et al. (1975) determined numbers of viable bacteria in second-instar *Culex pipiens quinquefasciatus* Say larvae following ingestion of pathogenic strain SSII-1 and non-pathogenic *Bacillus sphaericus*. The non-pathogenic *B. sphaericus* recovered from larvae declined rapidly after cessation of feeding as did numbers of pathogenic SSII-1 cells fed at LD₂₀ dosage. Pathogenic cells when fed at LD₇₀ dosage, the number of *B. sphaericus* in larvae was increased following initial decline. When chloroform treated SSII-1 cultures, in which all bacteria except spores were dead, were fed at LD₇₀ and LD₉₈ dosages, no viable *B. sphaericus* was recovered from larvae. The authors added that, in all SSII-1 treatments, other bacterial flora multiplied rapidly in larvae following onset of mortality.

Ramoska et al. (1977) elucidated that Bacillus sphaericus strain 1593, 1404 and SSII-1 were assayed for infectivity against field collected larvae of Psorophora columbiae, [Dayer &Knab] Culex nigripalpus [Theobald] and Aedes taeniorhynchus [Lynch Arribalzaga] in southwest Florida.

Their results indicated that, all the three strains were highly active against the Psorophora and Culex species. Aedes taeniorhynchus was also found susceptible but required higher doses to achieve lethal responses. Tests were also conducted on the rate of infection and the differences in susceptibility of different instars to B. sphaericus. The authors stated that nearly 75% of the mortality occurred within 48 hrs. post-incubation with the bacterium.

Mulligan et al. (1978) found that Bacillus sphaericus Neide strain 1593-4 was virulent against the southern house mosquito, Culex quinquefasciatus Say, and possessed longer residual activity than strain SSII.1 in pond test at Fresno, California.

Further testing of strain 1593-4 provided excellent control of a natural population of Culex tarsalis Coquillett at 10^4 cells/ml

Infectivity of the bacterium to a mixed population of Cx. tarsalis and southern house mosquito was decreased in a treatment of a dairy drain. The authors also noticed effective control of a mixed population of Aedes melanimon Dyar and Ae. nigromaculis Ludlow was not obtained at 10^4 Cell/ml. in 0.02 ha pasture plots, although laboratory tests showed

that Ae. nigromaculis was high susceptibility to the bacterium compared to other Aedes species. They added that the field tests of strain 1593-4 showed no deleterious effect on non-target organisms at 10^5 Cells/ml.

Myers and Yousten (1978) related the pathogenicity of Bacillus sphaericus SSII.1 to be due to toxin. mediated rather than an infectious process. Chloroform or ultraviolet-light treatments that decreased the viable count of SSII.1 cells by 4 or 5 logs did not significantly alter the ability of the bacterial cells to kill larvae. They presented also that three lines of evidence indicated that toxic activity was not related to sporulation: 1) Cells grown in either a complex or a defined medium were toxic to all ages; 2) When supplemental Mn^{2+} was excluded from complex medium, the culture yielded few spores but was of equal toxicity to a culture containing many spores; and 3) Several early blocked oligo sporogenous mutants were isolated that had toxic activities comparable to that of the parent. The authors added that the toxin was shown to be relatively unstable because activity was destroyed by heat and decreased by refrigeration.

Myers et al. (1979) assayed two strains of Bacillus sphaericus SSII.1 and 1593 for toxic activity against second instar larvae of Culex pipiens quinquefasciatus. They found that strain 1593 developed a level of toxicity 3000 times that of strain SSII.1. The authors stated that although the toxic activity of B. sphaericus SSII.1 was relatively unchanged

throughout growth, an increase in activity of strain 1593 occurred as the bacteria began to sporulate; the toxin of strain 1593 was shown to be more stable than that of strain SSII-1.

Unlike the spores of strain SSII.1, the spores of B. sphaericus 1593 were found to be highly toxic.

Ramoska and Pacey (1979) found that the effectiveness of Bacillus sphaericus (Strain 1593) for the control of mosquito larvae in the laboratory was inversely related to the amount of food available to the larvae. The rate of bacterial uptake differed between larvae of Culex quinquefasciatus and Anopheles albimanus.

Larvae of the former species consumed lethal quantities rapidly (5 min. onwards) after bacterial application, whereas, those of the latter species did so less rapidly, resulting in low mortality even after incubation for 90 min. with a high concentration (7×10^7 Cells/ml.)

Balaraman (1980) compared two strains of Bacillus sphaericus isolated indigenously from sick moribund mosquito larvae in Pondichary to a strain of B. sphaericus which was commercially produced in U.S.A and supplied by WHO, regarding their efficacy in killing larvae of three species of mosquito vectors of diseases (Culex fatigans, Anopheles stephensi and Aedes aegypti).

The author found that the indigenous isolated strain was as efficient for use as a larvicide as the strain supplied by the WHO.