J. of Bio. Res & Biotech. 1, (1), 29 - 34 (Jan/June, 2003)

Evaluation of the Larvicidal Activities of *Bacillus Sphaericus* on *Culex* Mosquito Found in Sokoto

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Abstract

Studies on the larvicidal activities of *Bacillus sphaericus,* against *Culex quinquefasciatus* mosquitoes found in (Sokoto), was carried out. The *B. sphaericus* (SPH 88) was obtained from Paseur Institute in Paris (France) in the form of a water dispersible powder. About 50mg of the powder was dissolved in 10ml sterile distilled water. From this stock, 1:100 dilution was made. Subsequent volumes of 15, 30, 60, 90, 100 and 120µg/l were fetched using micropipettes and added to beakers containing 150ml sterile distilled water. The mosquito larvae added to these beakers were obtained from reared mosquitoes in the laboratory. Twenty-five 4th instar larvae were placed in each beaker and left for 24 hours for larval mortality at room temperature. The larvicidal activities determined showed that *Culex quinquefasciatus* mosquito from this part of the country is highly sensitive to this bacterium. Lethal concentrations (LC_{50}) in µg/l determined from a plot of percentage mortalities against concentrations was 0.004µg/l while LC_{90} was 0.019µg/l indicating the *B. sphaericus* attacks the *Culex* mosquito with a consequent killing of over 90% of the population under test. Statistical analysis conducted using analysis of variance indicated that there was no significant difference in the overall larvicidal activities at 5% confidence limit (F>0.05).

Key Words: *Bacillus, Sphaericus, mosquitos, Sokoto*

Introduction

Culex mosquitoes are known as the vectors of the pathogens causing filariasis and certain encephalitis (Clements 1963). These are still among the most important worldwide health problems, despite progress made in their control (WHO, 1980a). Records on filariasis show that about one third of the African people are infected with filariasis. The global estimate of filariasis is one hundred and nineteen million. About 73 countries in Africa, Asia, Central America and Pacific Islands are endemic for filariasis (TDR, 1999). In 1990, about three million men and one million women lost their lives due to filariasis and at present filariasis has been designated by the WHO as a public health problem which should be for elimination by the year 2020 (TDR, 1999).

It is the intention of this paper to evaluate *Bacillus sphaericus* on the local population of *Culex quinquefasciatus* mosquitoes in other to determine the possibility of applying such bacteria

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in the control of such mosquitoes.

Materials and Methods

The *Bacillus sphaericus* SPH88 used is a water dispersible powder (Lot BSP247) produced by Pasteur institute in Paris, France. The test was carried out on the fourth instar larvae of the *Culex quinquefasciatus* mosquito (Umar, 1986; Anonymous, 1997).

Source of larvae

The adult *Culex quinquefasciatus* mosquitoes were collected from Sokoto township using insect hand nets and collection cage. The insects were separated using aspirator and identified according to Chandler and Read (1960) and placed in a rearing cage. The caged Mosquitoes were fed with 4% sucrose solution and grass infusion of *Pennisetum* sp. To serve as food and for egg laying. Adult female mosquitoes were blood fed by introducing a skinned Pigeon into the cage and left overnight twice weekly. Observation was made daily for oviposition. Laid eggs were transferred to larval rearing Pan flooded with water. Hatched larvae were allowed to reach the L4 stage (Dyers law) before use.

Preparation of various concentrations used

Fifty milligrams (50mg) of *B. sphaericus* powder was placed in universal bottles containing 10ml sterile distilled water (5000mg/1000ml). From this mixture a stock solution was made in a universal bottle by adding 0.1ml of the mixture to 9.9ml of sterile distilled water followed by agitation for one minute. From the stock solution (50mg/1), subsequent dilutions were prepared by fetching the following volumes using micropipettes; 15, 30, 60, 90, 100 and 120 micro-litre and added to six beakers each containing 150ml of water. The concentration of *Bacillus sphaericus* suspension in each beaker now becomes 0.05, 0.1, 0.2, 0.3, 0.33 and 0.4ug/l respectively. All concentrations were prepared in duplicates. A control beaker was set up having only water and larvae with no bacteria included.

Testing the larvicidal efficacy

To each set of beakers containing the concentrations of the *Bacillus sphaericus*, 25 fourth instar larvae of the *Culex quinquefasciatus* mosquitoes were added using a dropping pipette. The larvae exposed to *Bacillus sphaericus* concentrations were allowed to stand for 24 to 48 hours at Room Temperature. After 24 hours the number of dead larvae in each beaker were counted and the LC_{50} was obtained from a graph of mortality plotted against the concentrations. The potency of the bacteria in this formulation has been determined from the source of the powder as 1700ITU/mg.

Results and Discussion

The result of the larvicidal activities of various concentrations of *B. sphaericus* against the larvae of *Culex* mosquito is given in Table 1. Mortality was observed for a period of 12 hours. Larval mortality was observed for the initial period of 30 minutes of exposure. Mortality was recorded in the first 30 minutes through to the 12th hour. Larval mortality was seen to increase gradually from

lower concentrations to higher concentrations. The highest mortalities of 92% each were recorded at 60μg/l and 90μg/l concentrations. The lethal concentration (LC_{50}), graphically determined from the graph of percentage mortality and concentration was found to be 0.004 (figure 1). Statistical analysis conducted using analysis of variance indicated that there was no significant difference in the overall larvicidal activities (F>0.05) even though there was a significant difference in the means of certain concentrations (60, 90 and 120 μg/l), each having a mean of 1.64,1.64 and 1.50 respectively. The results of this research showed that *B. sphaericus* is highly active on *Culex* mosquito from this part of the world and it lends support to the practice in some countries like Europe, West Africa and Sri lanka where *B. sphaericus* has been used extensively for the field control of *Culex* mosquitoes (Wickremensinghe, 1980; Lacey and Singer, 1982; Obeta, 1986; Lacey et al., 1988; Thiery et al., 1992).

Table 1. **Mortality among larvae of *Culex* sp. Treated with various concentrations of standard powder of *B. sphaericus***

Time(min)	Conc. In μg/l					
	15	30	60	90	100	120
15	0	0	0	0	0	0
30	0	0	1	0	1	1
60	0	1	1	2	1	1
120	1	2	2	2	2	1
180	1	2	7	3	6	1
240	4	4	3	5	1	6
300	7	4	1	4	1	2
360	0	2	0	0	0	1
420	1	0	1	0	0	3
480	1	0	1	0	0	0
540	0	0	0	1	0	0
600	1	1	4	1	3	5
660	0	1	2	3	2	0
720	0	1	0	2	0	0
Total	17	18	23	23	17	21
Mort (%)	68	72	92	92	68	84

Conclusion

Lavicidal activities of *B. sphaericus* on *Culex* mosquito had been determined from this part of the country. The rate of mortality increased as the concentrations increased. The lethal concentration determined was 0.004µg/l. It is recommended to employ this *B. sphaericus* for the control of mosquito populations in Nigeria.

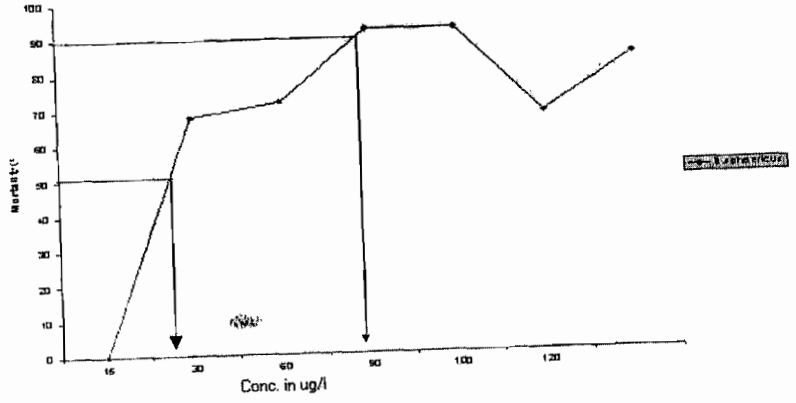

Fig.1 Mortality among larvae of Culex *quinquefasciatus* treated with *Bacillus sphaericus* Powder.

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