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Short Communication

Larvicidal Potential of Microbial Agents *Bacillus thuringiensis* var. *israelensis* and *Bacillus sphaericus* against Mosquito Vectors Prevalent in Arid Areas

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In arid conditions the mosquito control can effectively be carried-out by applying control measures against the immature stages rather than the adult population, since in arid areas the water resources are very limited and so are the mosquito breeding habitats (Mulla *et al.*, 2001). In such situations use of microbial agents appears the most suitable method (Mittal *et al.*, 2001). They do not show cross-resistance to chemical larvicides and are suitable for areas showing resistance to organo-phosphates or pyrethroids (BCN, 1998). Keeping this in view laboratory studies were carried out to determine the larvicidal efficacy of microbial agents *Bacillus thuringiensis* var. *israelensis* and *Bacillus sphaericus* against important mosquito vector species prevalent in arid areas.

Laboratory evaluation of *Bacillus thuringiensis* var. *israelensis* (*Bti*) was carried-out against three important vector species, viz., *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*, collected from different urban and peri- urban areas of Jodhpur city, while, *Bacillus sphaericus* (*Bs*) was evaluated only against *Cx. quinquefasciatus*. The microbial agents, Sphericide WP and Bacticide WP, the wetable powder formulations (16% WP) of *Bs* (800 IU/mg) and *Bti* (5000 IU/mg), respectively, procured from Biotech International Ltd., were used in the experiments. All the experiments were conducted as per the recommendations of WHO for evaluation of larvicidal activity of an insecticide/ larvicide in laboratory (WHO, 1975). Late third and early fourth instar larvae of the mosquito species were used in the experiments. Batches of 25 larvae were exposed to 250 mL of each test concentration. The temperature and relative humidity (RH) during the experiments were 28±2°C and 75±5%, respectively. The observations were recorded after 24 hours. Each vector species had 4-5 replicates of each concentration to obtain concentration- mortality response data. Lethal concentrations at 50 (LC_{50}) and 90 (LC_{90}) per cent levels were determined using log-probit analysis (Finney, 1971).

All the three vector mosquito species were found susceptible to the *Bti* formulation used in the experiments (Table 1). The concentrations tried were from 0.05 to 1, 0.01 to 0.1 and 0.01 to 0.5 mg L^{-1} for *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus*, respectively. Based on the concentration-mortality response, the lethal concentrations at 50% level (LC_{50}) and 90% level (LC_{90}), were calculated by log-probit analysis. *Bti* at LC_{50} level was 3.3 and 3.5 times more effective against *Ae. aegypti* and *Cx. quinquefasciatus*,

Table 1. Laboratory evaluation of the efficacy of Bacillus thuringiensis var. israelensis against three vector mosquito species

Mosquito species	Concentrations (mg L^{-1})	Mortality (%)	Regresion coefficient	Regression equation	Chi-square (df)	LC_{50} (Fiducial limits)	LC_{90} (Fiducial limits)
Anopheles stephensi	Control	00.0	1.46	Y = 3.64+1.46x	4.09 (3)	0.085 (0.063-0.0117)	1.134 (0.301-1.375)
	0.05	33.3 (4.5)*					
	0.10	64.2 (5.3)					
	0.50	79.8 (5.8)					
	1.00	97.3 (6.9)					
Aedes aegypti	Control	00.0	2.01	Y = 2.15+2.01x	5.87 (4)	0.026 (0.019-0.036)	0.113 (0.054-0.241)
	0.05	24.7 (4.3)					
	0.10	37.8 (4.6)					
	0.50	78.9 (5.8)					
	0.75	86.6 (6.1)					
	1.00	96.0 (6.7)					
Culex quinquefasciatus	Control	00.0	1.45	Y = 2.99+1.45x	1.56 (3)	0.024 (0.018-0.035)	0.186 (0.077-0.488)
	0.01	30.0 (4.4)					
	0.05	65.3 (5.3)					
	0.10	89.8 (6.2)					
	0.50	100.0 (8.7)					

* Figures in parenthes show empirical probits.

respectively, in comparison to *An. stephensi*. However, at LC_{90} level *Bti* was found 10.0 and 6.1 times more effective against *Ae. aegypti* and *Cx. quinquefasciatus* in comparison to *An. setphensi.*

Cx. quinquefasciatus was found susceptible to *Bs.* (Table 2). The concentrations used in the experiments were from 0.005 to 1.00 mg L^{-1}. Based on the concentration-mortality response, the lethal concentrations at 50% level (LC_{50}) and 90% level (LC_{90}), calculated by log-probit analysis, were 0.06 and 1.251 mg L^{-1}. The chi-square value calculated during the analysis did not show any heterogeneity in the response.

The laboratory evaluation of these microbial agents has been carried out against different strains of vector mosquito species with particular reference to *An. stephensi* and *Ae. aegypti*, which are established vectors of malaria and dengue/DHF in urban situations and breed in different habitats being used for storing the water for domestic use. Sharma *et al.* (1982) determined LC_{90} of *Bti* (0.22 mg L^{-1}) against *Ae. aegypti*, which was 4.23 times more effective than present study, whereas, studies of Malaria Research Center (MRC, 2000) determined LC_{50} values of *Bti* against vector mosquito species between 0.034 to 0.16 mg L^{-1}, which are 1.4 to 1.9 times more effective than estimated. In the present study. In another study (Mittal *et al.*, 2001) the LC_{50} values of *Bti* against *An. stephensi* (0.14 mg L^{-1}) and *Ae. aegypti* (0.06 mg L^{-1}), were 1.6 and 2.45 times more effective than that obtained in the present study. Barua and Das (1994) in Assam evaluated *Bti* (Teknar) against *Cx. quinquefasciatus* and determined LC_{50} and LC_{90} values 13.5 and 7.8 times

Table 2. Laboratory evaluation of the efficacy of Bacillus sphaericus against Culex quinquefasciatus

Conc. (mg L^{-1})	Mortality (%)	Regression coefficient	Regression equation	Chi-square (df)	LC_{50} (Fiducial limits)	LC_{90} (Fiducial limits)
Control	0.3.3	0.97	Y =	1.81	0.060	1.251
0.005	13.3 (3.89)*		3.28+0.97x	(4)	(0.037-0.097)	(0.432-3.628)
0.01	26.7 (4.38)					
0.1	56.0 (5.13)					
0.5	78.7 (5.79)					
1.0	94.7 (6.62)					

* Figures in parenthes show empirical probits.

more effective than in our estimation. It is clear from the above observations that the mosquito species tested from arid areas are more susceptible to these microbial products in comparison to the vector populations prevalent in other areas.

Studies have revealed *Bti* was more effective against the larvae of *Aedes* and *Culex* and *Anopheles* species (Mittal *et al.*, 2001). Same trend was observed in the present study. Based on the larvicidal efficacy of these microbial agents can be recommended for mosquito control in arid situations.

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