Evaluation of Biolarvicides

Biolarvicides

Over the last few decades, there is growing realization that alternate methods to chemical control need to be studied and perfected. In the last decade the bacilli based mosquito larvicides popularly known as biocides or biolarvicides are becoming popular in vector control. Many commercial formulations are available and can be used in large-scale mosquito control operations. The major advantages of biolarvicides are—reduced application costs, safety to environment, human beings, animals and other nontarget organisms. Various formulations of *Bacillus sphaericus* and *B. thuringiensis* var. *israelensis* have been evaluated during the last two decades.

Laboratory and Field Evaluations

Biocide-S (*Bacillus sphaericus* 1593M) developed by Madurai Kamraj University was the first biocide tested at MRC in 1983, later three experimental formulations of Biocide-S–HIL-8, 9 and 10 wettable powder and dust formulations were developed and tested in laboratory and small-scale field trials (Mittal *et al*., 1985). These formulations were found to be effective against both *Culex* and *Anopheles* larvae in laboratory and small pits in field. However, further development of various aqueous formulations, MKU-1, 2 AU, etc. based on Biocide-S were not much effective against *Anopheles* sp. particularly *An. culicifacies*. In addition to Biocide-S various other formulations of *B. sphaericus* obtained through WHO and various other laboratories in India and abroad were tested both in laboratory and small-scale field trials (Ansari *et al*., 1989, 1995). All these formulations were however, effective only against culicines and some anopheline sp. particularly against *An. stephensi* and *An. subpictus* and not against *An. culicifacies*.

Solvay, a liquid and Abotts granular formulations of *B. sphaericus* 2362 obtained through WHO, were tested against *An. stephensi*, *An. culicifacies* and *Cx. quinquefasciatus* both under laboratory and field conditions (Ansari *et al*., 1989). Later two more formulations of *B. sphaericus* 2362 namely Spherimos and Vectolex 2.5 AS obtained through WHO were tested (Ansari *et al*., 1995). It was found that *Cx. quinquefasciatus* was more susceptible to all these formulations followed by *An. stephensi* and *An. culicifacies*. In the field condition, absolute mortality in *Culex* larvae was maintained with Spherimos @ 2 ml/m^2 for 1 week in pools and 99% reduction for 3 weeks in the wells. The activity was enhanced for three weeks in pools when the dose was increased to 10 ml/m^2. On the other hand, against anophelines the absolute impact was seen for 1 week @ 2 ml/m^2 and up to 90.8% for two weeks in the pools. In some pools the impact was moderate. In the wells, the impact was seen for 1 week @ 2 ml/m^2 whereas @ 10 ml/m^2 it was prolonged for 2 weeks. A good impact of Vectolex was observed on *Culex* sp. in the field which lasted for 4 weeks in the pools @ 2–5 ml/m^2 and for 6 weeks in unused wells @ 5–10 ml/m^2 (Fig. 36).

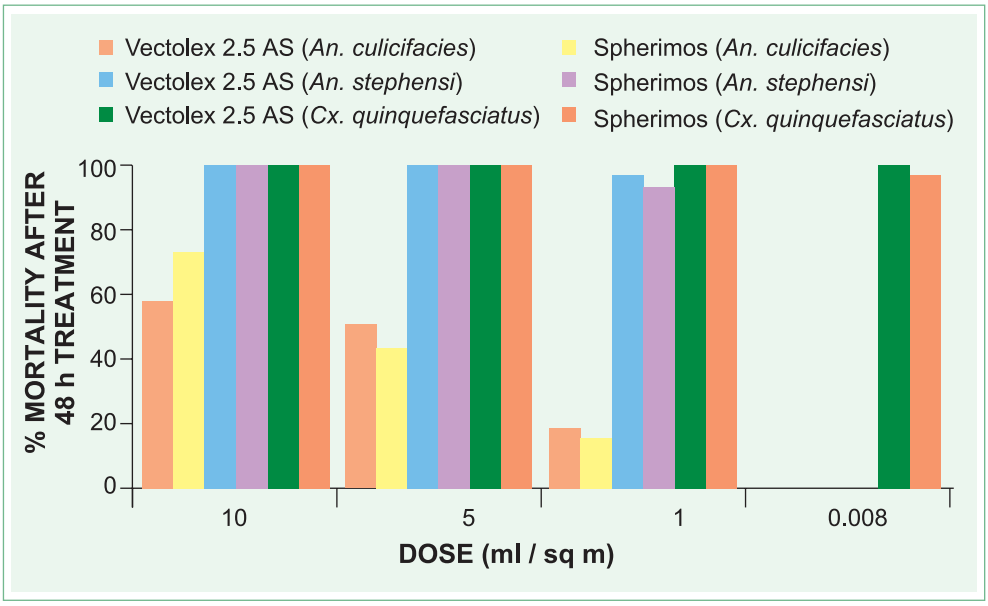

Fig. 36: **Laboratory evaluation of *B. sphaericus* 2362 flowable formulations against larvae**

In the year 1992, National Malaria Eradication Programme (now NAMP) procured two Russian formulations of biolarvicides—Spherix (*B. sphaericus*, serotype H 5a5b, strain B-101) and Bacticide (*B. thuringiensis* var. *israelensis*, serotype H-14, strain 164). NAMP provided 40 MT of Spherix and 10 MT Bacticide to MRC for scientific evaluation by conducting multicentric field trials in different eco-climatic situations in India.

Prior to this, the laboratory bioassays with these two formulations were conducted in Delhi from July to October 1991. It was observed that both Spherix and Bacticide were effective against *An. culicifacies*, *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti* larvae (LC_{50} values of Spherix ranged from 0.19 to >40 mg/l and Bacticide from 0.034 to 0.16mg/l) (Fig. 37). It was found that Spherix was most effective against *Cx. quinquefasciatus* and the least against *Ae. aegypti*, whereas Bacticide (named as Bactoculicide) was most effective against *Ae. aegypti* and the least against *An. culicifacies.* Among the two malaria vectors tested, *An. stephensi* was much more susceptible than *An. culicifacies* to both formulations, this observation was also corroborated later by the findings of the field trials (Mittal *et al*., 1993).

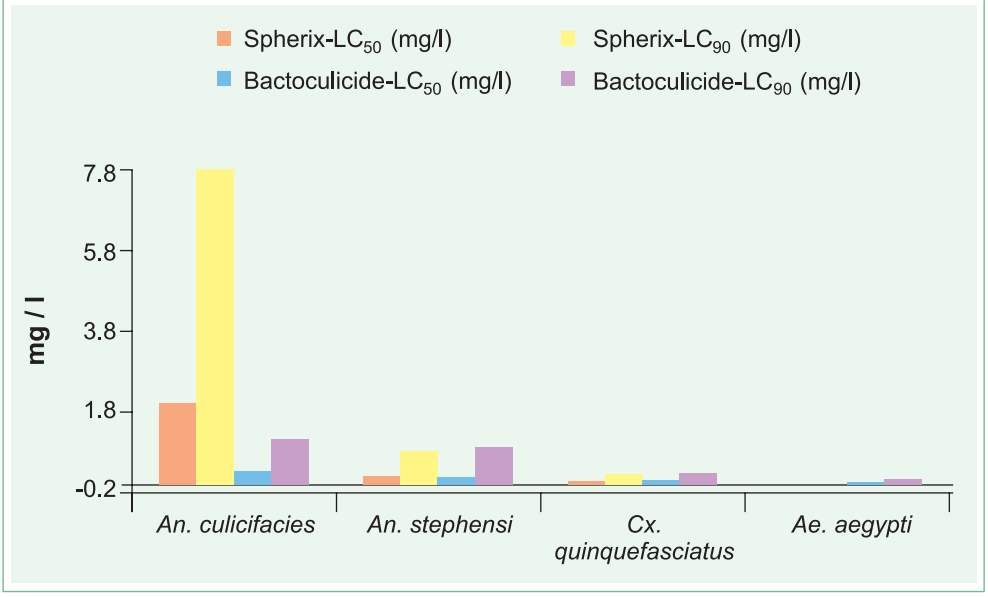

Fig. 37: **Comparative toxicities of spherix and bactoculicide against III instar mosquito larvae**

The safety profile of these formulations to nontarget organisms (NTOs) such as *Gambusia affinis*, *Poecelia reticulata*, frog tadpoles, notonectid bugs (*Enithares indica* and *Anisops sardae*) and Cyclops (Copepods) was also established prior to large-scale field trials in the country. The LC_{50} values for above NTOs ranged from 50 to > 2000 mg/l, which were many fold higher than the recommended field dose for larval control.

Simultaneously, studies on effect of temperature showed that a 10 °C rise in temperature from 21 to 31 °C increased efficacy of Spherix and Bacticide by 2–100 fold against different vector species (Fig. 38) (Mittal *et al*., 1993). This indicated that biolarvicides would lose efficacy in cold climatic conditions and prove very useful in warmer months or tropical climatic conditions which exist in India for most of the year (Fig. 39).

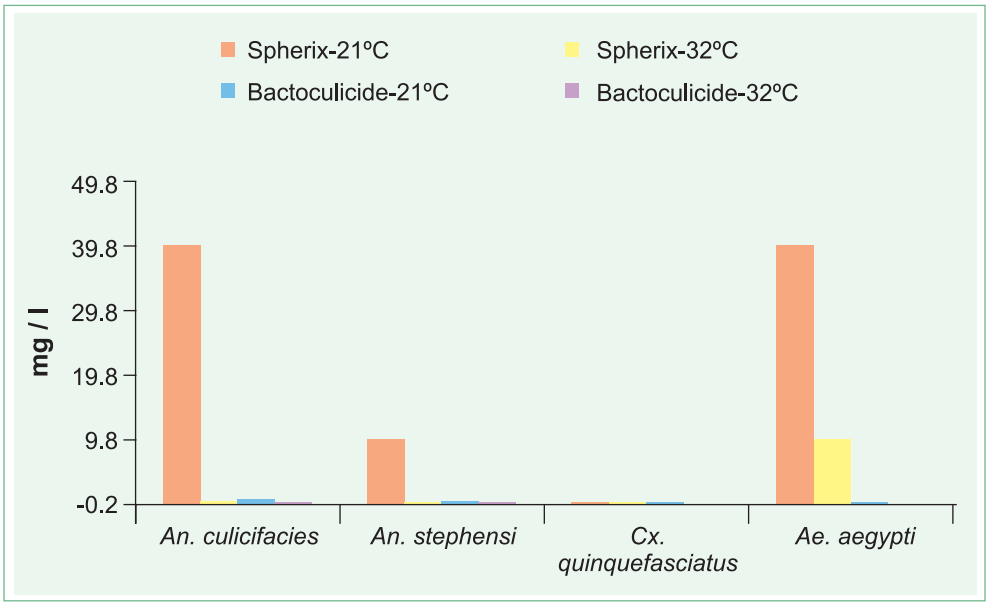

Fig. 38: **Effect of temperature on larvicidal activity (LC_{50}) of spherix and bactoculicide against different vector species**

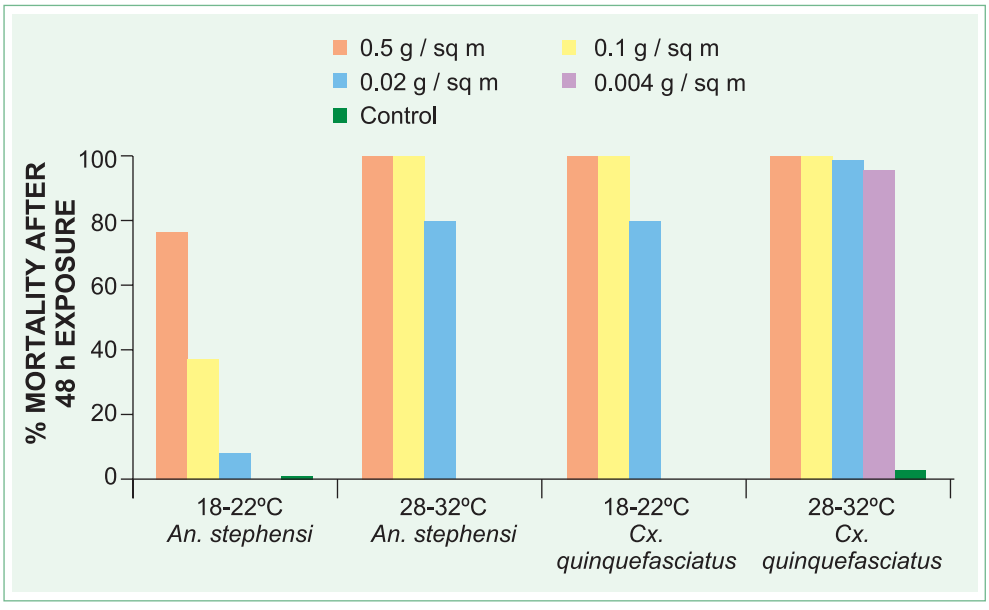

Fig. 39: **Studies on the effect of temperature on the efficacy of spherix on *An. stephensi* and *Cx. quinquefasciatus***

Similarly it was observed that beyond pH 9.5 the efficacy of both Spherix and Bacticide was reduced drastically (Mittal *et al*., 1995). There was however, no significant difference observed in the activity between pH 3.5-9.5.

Multicentric trials were carried out with Spherix in different MRC field stations, namely at Farrukhabad, Shahjahanpur, Hardwar, Mathura, Ghaziabad, Delhi, Panaji and Vasco da Gama, Goa, Chennai, Mandla, Nadiad, Shankargarh, Rourkela and Car Nicobar Islands. From the results of these multicentric studies it became evident that both the Russian formulations—Spherix and Bacticide were effective against anophelines and culicines including disease vectors in different ecological conditions (Fig. 40). The residual activity of biocides depended upon vegetation, algae and organic pollution. The larval control was longer when breeding habitats had less pollution, less or no vegetation as effect could be seen for up to 4 weeks in them, whereas the impact lasted for 3–7 days in others (Figs. 41 & 42).

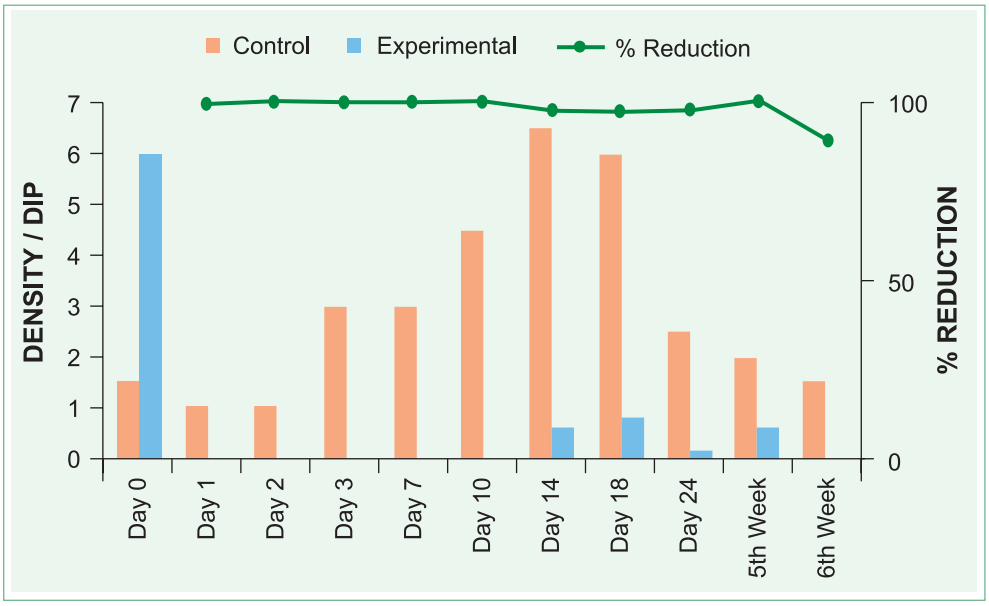

Fig. 40: **Efficacy of bactoculicide against mosquito larvae in industrial scraps at BHEL, Hardwar, Uttaranchal**

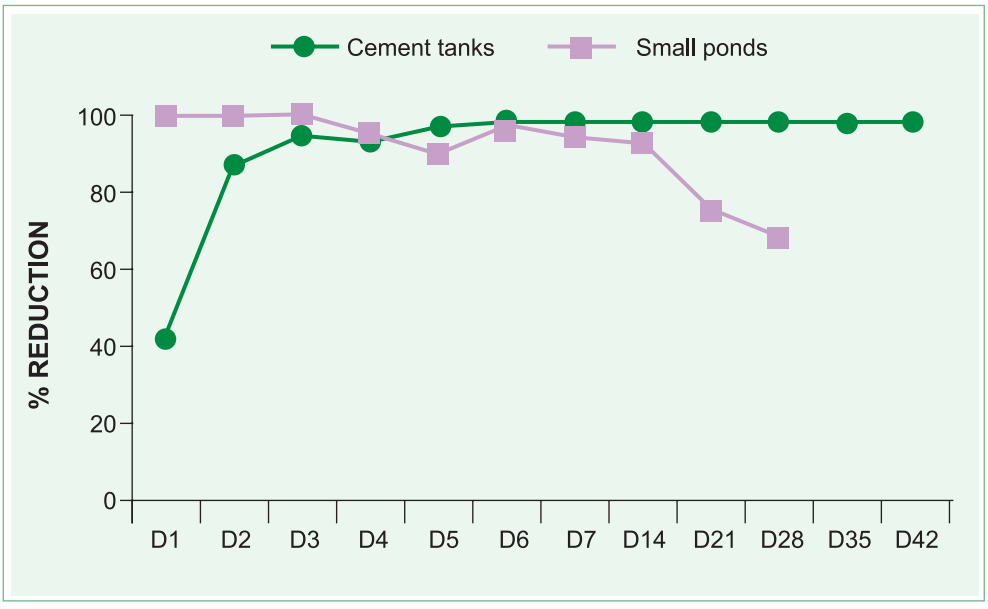

Fig. 41: **Larvicidal efficacy of spherix against anopheline larvae in cemented tanks and small ponds after a single application in Bhabhar area, District Nainital, Uttaranchal**

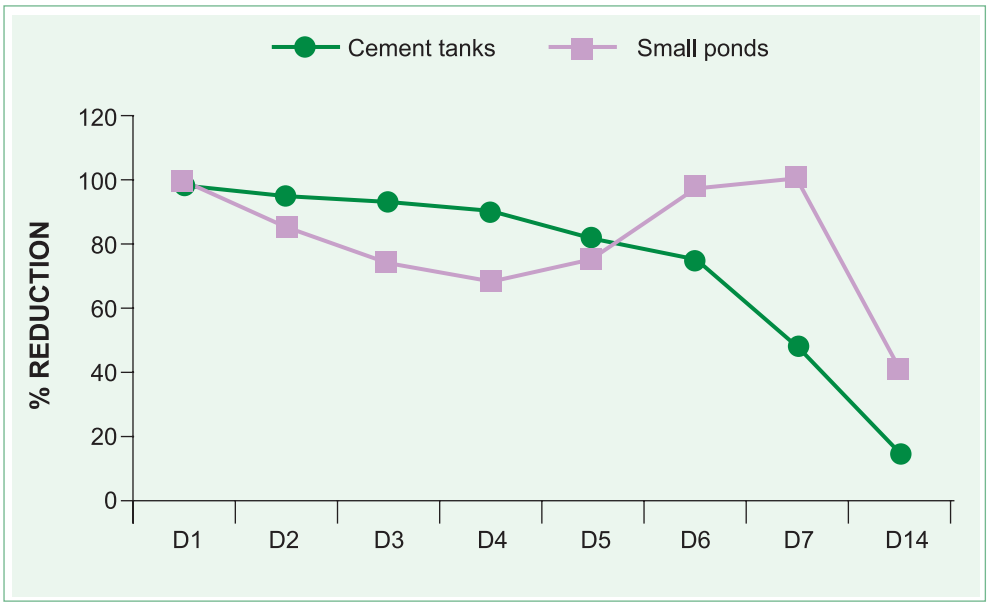

Fig. 42: **Larvicidal efficacy of bactoculicide against anopheline larvae in cemented tanks and small ponds**

Later extensive field trials were also conducted by MRC to assess the efficacy of the above formulations. Later various other formulations of *Bti* tablet, granule and wettable powder formulations against different disease vector species and in urban and rural areas were tested (Batra *et al*., 2000; Dua *et al*., 1993; Kar *et al*., 1997; Kumar *et al*., 1994, 1995, 1996, 1998; Mittal *et al*., 2000; Shukla *et al*., 1997 & Yadav *et al*., 1997).

Recently a commercial formulation (Wockhardt *Bti*) of *B. thuringiensis* var. *israelensis* H-14 50% WP was evaluated in field in three field stations at Goa, Haldwani (Uttaranchal) and Shahjahanpur (U.P.). Field efficacy was tested in different breeding habitats namely paddy fields, river bed pools, pokhars, pits and drains, septic tanks, ornamental fountains and cement tanks. The formulation was tested at 2 doses, 0.5 and 1 g/m^2. Over 90% reduction in larval density was observed in most of the breeding habitats at a dose of 0.5 g/m^2 except in septic tanks. In septic tanks 1 g/m^2 was effective. Residual effect lasted for 1 week in most of the breeding sites except in ornamental fountains and cement tanks. In paddy fields the efficacy was observed for only 2 days (Fig. 43). n

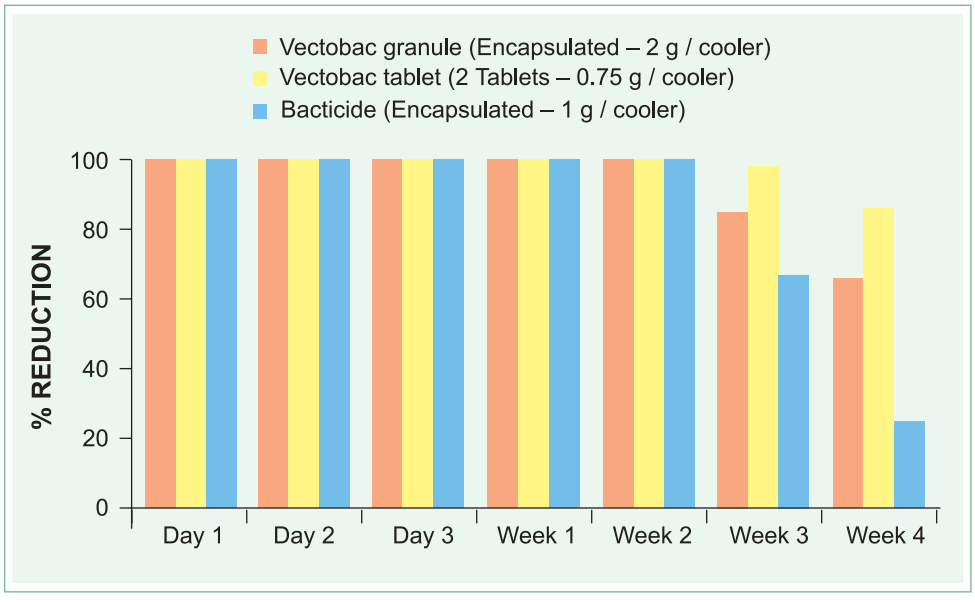

Fig. 43: **Efficacy of different formulations in containing *Ae. aegypti* breeding in treated desert coolers**