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Relative susceptibility of some common mosquito vector larvae to synthetic insecticidal compounds in north-western Rajasthan

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Abstract: *Relative susceptibility of three important mosquito vector larvae viz., Anopheles stephensi, Aedes aegypti and Culex quinquefasciatus, prevalent in the arid region was determined against four organophosphates (malathion, fenitrothion, fenthion, temephos) and three synthetic pyrethroid (alphamethrin, deltamethrin and fanvalerate) compounds. Studies were carried out on late 3rd or early 4th instar larvae of these species using standard WHO technique. Based on concentration mortality data* LC_{50} *and* LC_{90} *values along with their fiducial limits, regression equation, chi-square* (x^2)*/ heterogeneity of the response have been determined by log probit regression analysis.* LC_{50} *values as observed for the above seven insecticides were 0.8097, 0.0398. 0.0432, 0.0035, 0.0025, 0.0092, 0.1006; 1.2370, 0.0531, 0.0655, 0.0076, 0.00004, 0.00004, 0.0046 and 1.4980, 0.0719, 0.0817, 0.0056, 0.00021, 0.00073, 0.0112 mg/l for the above three mosquito species respectively. Among the four organophosphates tested temephos was the most effective followed by fenitrothion, fenthion and malathion. In general, Anopheles was found more susceptible as compared to the other two culicines to the above four compounds. The results also showed that larvae of Ae. aegypti were most susceptible followed by Cx. quinquefasciatus and An. stephensi to all the three pyrethroids tested. Among the three compounds tested alphamethrin was found to be the most toxic followed by deltamethrin while fanvalerate was the least toxic. The study would be of great importance while planning use of these insecticides for the control of different vector species in this area.*

Key words: *Organophosphates, Pyrethroids, Vector mosquitoes, Desert Rajasthan*
PDF file of full length paper is available with author

Introduction

Vector control, which includes both anti-larval and anti-adult measures constitutes an important aspect of any mosquito control programmes. Mosquitoes, the best known group among dipteran insects, are of great importance to man as vectors of many communicable diseases such as malaria, filaria, dengue/dengue hemorrhagic fever (DHF) *etc.* Among the anophelines, *Anopheles stephensi* and *An. culicifacies* are the important vector of malaria while among the culicines, *Culex quinquefasciatus* and *Aedes aegypti* are the vectors of filaria and dengue/DHF respectively. All these mosquito species have been identified as primary vectors of the above communicable diseases in this region of Rajasthan (Bansal and Singh, 1993; Bansal *et al*., 1994; Prakash *et al*., 2005). Hence, their control either by biological or chemical means is the basic requirement for planning an effective vector control strategy. Although the use of insecticides poses the threat of environmental pollution and insect resistance, yet in the developing countries including India control of tropical diseases like malaria, filaria and dengue is solely dependent upon chemical control. Although several studies have been done on the susceptibility status of adult mosquitoes in India (Sahu *et al*., 1990; Chand and Yadav, 1991; Baskar, 1992; Bansal and Singh, 1995; Chaudhry and Anand, 2005; Bansal and Singh, 1996; Bansal and Singh, 2006), yet the level of susceptibility of its larval stages to different synthetic larvicides or adulticides is lacking in this area in spite of the fact that susceptibility differs from region to region.

Materials and Methods

Fully fed female mosquitoes of all the three species were collected early in the morning from different areas of Jodhpur city. Collection was made from inside the human dwellings and cattle sheds with the help of an aspirator supplied by WHO and kept in Barraud cages provided with cotton pads soaked in 10% glucose solution and inside with an enamel water tray for laying eggs. Different larval stages from 1st to 4th instar were reared in the laboratory and used for the tests. During this period the larvae were fed on finely powdered dog biscuits and yeast powder in the ratio of 2:1. Various test concentrations were prepared by adding standard insecticide solution of all the insecticides to 249 ml. of tap water in a 500-ml. beaker. Control tests were also conducted by adding the same amount of solvent to 249 ml. of water. To each of the beakers containing different test concentrations, 25 healthy late 3rd instar larvae were released. Percent mortalities were calculated 24 hr later by counting both dead and moribund larvae as per instructions given by WHO (1981). Larvae were considered moribund if they failed to flex head to siphon when provoked with a glass rod. All tests were carried out at a controlled room temperature of 28±2°C and RH at 75±5%. All the tests were repeated four times to investigate variations and average was taken. The data were corrected by using Abbott's (1925) formula if mortality was between 5-20% in control experiments. The LC_{50} and LC_{90} values were computed using log probit regression analysis (Finney, 1971).

Results and Discussion

The results of the relative susceptibility of larvae of all the three mosquito species *viz*., *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus,* to the seven synthetic insecticidal compounds are given in the Table 1-3. From the tables it is very clear that alphamethrin and deltamethrin used in the present study are very effective at very low concentrations for each mosquito species, however, fanvalerate was found very less effective as compared to the above two pyrethroids tested. Among the organophosphorous tested temephos was the most effective while malathion the least with all the three vector species tested. The efficacy of fenitrothion and fenthion to all the three species was found in between malathion and temephos and are about 10-15 times less effective as compared to temephos, which is the most effective larvicide. Based on the LC_{50} values, *An. stephensi* was found more susceptible to malathion LC_{50}-0.8097) as compared to *Ae. Aegypti* (LC_{50}-1.2370) and *Cx. quinquefasciatus* (LC_{50}-1.4980 mg/l) respectively. Studies done at Gwalior (Gopalan *et al*., 1996) with malathion showed its LC_{50} in susceptible *Cx. quinquefasciatus* being 0.03 mg/l, but continuous exposure of the larvae up to 25th generation raised the LC_{50} to 2000 times from 0.03 to 61.09 mg/l. LC_{50} for malathion, however, in the present study was found from 0.81 to 1.50 mg/l for larvae of all the three mosquito species showing that a high degree of resistance is being built up in the population of this semi-arid area. However, with temephos *An. stephensi* (LC_{50}-0.0035) was most susceptible followed by *Cx. quinque- fasciatus* (LC_{50}-0.0056) and *Ae. Aegypti* (LC_{50}-0.0076 mg/l) and a 100% kill at the standard diagnostic concentration. A 100% kill with temephos has also been observed in the field collected and laboratory populations of these species at Panaji, Goa (Thavaselvam *et al*.,1993). Studies carried out with *Cx. quinquefasciatus* in other parts of India also revealed that temephos is a very effective larvicide, the LC_{50} being about 0.0015 at Pune (Ganguly *et al*., 1994) 0.0017 (Thomas *et al*., 1991) and 0.0022 (Mittal *et al*., 1994) at Delhi and 0.0076 mg/l at Bikaner (Bansal and Singh, 2002). However, test carried out at Rajahmundary town (Patnaik *et al*., 1997) showed that temephos 50% EC was not much effective in drastically reducing the larval and pupal density even at dosages four times higher than the recommended ones. Tests carried out with fenitrothion and fenthion also showed them to be quite effective larvicides and 96-100% mortality was observed at the standard diagnostic concentrations, the LC_{50} values being 0.0398 and 0.0432; 0.0531, 0.0655 and 0.0719, 0.0817 mg/l with all the three mosquito species respectively. These values are quite high when compared with results obtained with the larvae of *Cx. quinquefasciatus* in other parts of India (Thomas *et al*., 1991; Ganguly *et al*., 1994; Mittal *et al*., 1994; Gopalan *et al*., 1996) with the same insecticides. These results clearly indicate that larvae have developed resistance towards many of the organophosphate compounds tested in the present study. These results suggest that the resistance is slowly building up in the population of this area and its level has grown many times. High resistance might be due to the strain variations and their adaptability to the harsh desert climatic conditions.

A perusal of the Table 1-3 showed that larvae of *Ae. aegypti* were most susceptible followed by *Cx. quinquefasciatus* and *An. stephensi* towards all the pyrethroids tested in the present investigation. *Ae. aegypti* was found more susceptible (LC_{50}-0.00004) for alphamethrin as compared to *Cx. quinquefasciatus* (0.00021) and *An. stephensi* (LC_{50}-0.0025 mg /l) which showed that larvae of *Aedes* are 62.75 and of *Culex* 11.95 times more susceptible than *Anopheles* to alphamethrin. Similarly for deltamethrin larvae of *Ae. aegypti* were found more susceptible (LC_{50}-0.000041) followed by *Culex quinquefasciatus* (LC_{50}-0.00073 mg/l) and *An. stephensi* (LC_{50}-0.0092 mg/l) which showed that larvae of *Aedes* are 224.4 and of *Culex* 12.6 times more

Table - 1: Probit regression analysis of the mortality data of larvae of *An. stephensi*

Insecticides	Regression coefficient (slope)	Regression equation	Intercept	Chi-square heterogeneity (D.F.)	LC_{50} ± S.E. (Fiducial limits)	LC_{90} ± S.E. (Fiducial limits)
Malathion	1.82	Y=1.82x+1.54	1.54	2.14(2)	0.8097 ± 0.0117 (0.5927 - 1.1060)	4.1050 ± 0.0139 (2.1520 - 7.8300)
Fenitrothion	2.79	Y=2.79x - 0.53	0.53	1.23(2)	0.0398 ± 0.0011 (0.0319 - 0.0498)	0.1114 ± 0.0012 (0.0748 - 0.1750)
Fenthion	3.56	Y=3.56x - 0.82	-0.82	3.15(2)	0.0432 ± 0.0011 (0.0369 - 0.0506)	0.0990 ± 0.0012 (0.0724 - 0.1354)
Temephos	2.23	Y=2.23x+ 1.56	1.56	1.25(2)	0.0035 ± 0.0001 (0.0026 - 0.0047)	0.0131 ± 0.0001 (0.0065 - 0.0261)
Alphamethrin	1.23	Y=1.23x +3.29	3.29	3.72(2)	0.0025 ± 0.0001 (0.0016 - 0.0040)	0.0277± 0.0002 (0.0094 - 0.0822)
Deltamethrin	1.18	Y=1.18x +3.86	3.86	3.25(2)	0.0092 ± 0.0013 (0.0058 - 0.0145)	0.1118 ± 0.0018 (0.0340 - 0.3675)
Fanvalerate	0.90	Y=0.90x +4.09	4.09	2.49(2)	0.1006 ± 0.0134 (0.0564 - 0.1794)	2.6218 ± 0.0231 (0.5063 - 3.5738)

All values of LC_{50} and LC_{90} along with their fiducial limits are in mg/l

Table-2 : Probit regression analysis of the mortality data of larvae of *Ae. aegypti*

Insecticides	**Regression coefficient (slope)**	**Regression equation**	**Intercept**	**Chi-square heterogeneity (D.F.)**	**LC_{50} ± S.E. (Fiducial limits)**	**LC_{90} ± S.E. (Fiducial limits)**
Malathion	2.14	Y=2.14x + 0.52	0.52	4.23(2)	1.2370 ± 0.0117 (0.9022 - 1.6950)	4.8930 ±0.0143 (2.4170 - 9.9000)
Fenitrothion	2.93	Y=2.93x – 0.05	-0.05	2.11(2)	0.0531 ± 0.0011 (0.0440 - 0.0640)	0.1452 ± 0.0012 (0.0949 - 0.2221)
Fenthion	3.35	Y=3.35x – 1.09	-1.09	3.59(2)	0.0655 ± 0.0011 (0.0543 - 0.0789)	0.1579 ± 0.0012 (0.1027 - 0.2428)
Temephos	1.76	Y=1.76x + 1.68	1.68	3.93(2)	0.0076 ± 0.0001 (0.0054 - 0.0109)	0.0407 ± 0.0002 (0.0177 - 0.0935)
Alphamethrin	1.49	Y=1.49x +4.10	4.10	0.20(2)	0.00004 ± 0.00001 (0.00003 -0.00006)	0.0003 ± 0.0000 (0.0001 - 0.0007)
Deltamethrin	1.93	Y=1.93x +3.83	3.83	0.77(2)	0.00004 ± 0.00001 (0.00003 - 0.00006)	0.0002 ± 0.0000 (0.0001 - 0.0004)
Fanvalerate	2.41	Y=2.41x +3.53	3.53	0.23(2)	0.0046 ± 0.0012 (0.0034 - 0.0061)	0.0169 ± 0.0014 (0.0091 - 0.0315)

All values of LC_{50} and LC_{90} along with their fiducial limits are in mg/l

Table-3: Probit regression analysis of mortality data of larvae of *Cx. quinquefasciatus*

Insecticides	**Regression coefficient (slope)**	**Regression equation**	**Intercept**	**Chi-square heterogeneity (D.F.)**	**LC_{50} ± S.E. (Fiducial limits)**	**LC_{90} ±S.E. (Fiducial limits)**
Malathion	1.86	Y=1.86x + 0.96	0.96	3.69(2)	1.4980 ± 0.0115 (1.1290 - 1.9860)	7.3100 ± 0.0142 (3.6970 - 14.450)
Fenitrothion	3.57	Y=3.57x - 1.62	-1.62	6.66(2)	0.0719 ± 0.0011 (0.0622 - 0.0831)	0.1642 ± 0.0012 (0.1166 - 0.2312)
Fenthion	3.03	Y=3.03x - 0.79	-0.79	6.92(2)	0.0817 ± 0.0011 (0.0684 - 0.0977)	0.2164 ± 0.0013 (0.1391 - 0.3367)
Temephos	2.23	Y=2.23x+ 1.10	1.10	4.64(2)	0.0056 ± 0.0001 (0.0044 - 0.0071)	0.0208 ± 0.0001 (0.0122 - 0.0354)
Alphamethrin	1.18	Y=1.18x+3.45	3.45	3.08(2)	0.00021 ± 0.00001 (0.00013 - 0.00034)	0.0025 ± 0.00001 (0.0007 - 0.0087)
Deltamethrin	1.27	Y=1.27x +3.90	3.90	2.83(2)	0.00073 ± 0.00012 (0.00051 - 0.00106)	0.0075 ± 0.0002 (0.0030 - 0.0187)
Fanvalerate	2.17	Y=2.17x +2.72	2.72	0.83(2)	0.0112 ± 0.0012 (0.0089 - 0.0142)	0.0436 ± 0.0013 (0.0255 - 0.0746)

All values of LC_{50} and LC_{90} along with their fiducial limits are in mg/l

susceptible than *Anopheles* to deltamethrrin. Verma *et al*. (1983) evaluated the efficacy of cypermethrin to the larvae of above three mosquito species and found *Culex quinquefasciatus* to be more susceptible (LC_{50} -0.00032) instead of *Ae. Aegypti* (LC_{50}- 0.00037 and *An. stephensi* (LC_{50}-0.0054 mg/l). Verma and Rahman (1984), also evaluated the efficacy of synthetic pyrethroids over natural pyrethrins and DDT and found the formers to be much more effective for the above three mosquito species. Experiments carried out with fanvalerate in the present study found the LC_{50} values for *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* as 0.1006, 0.0046 and 0.0112 mg/l respectively which showed that larvae of *Aedes* were 21.9 and of *Culex* 9.0 times more susceptible than *An. stephensi*. The above experiments clearly revealed that fanvalerate was least effective in comparison to rest of the two pyrethroids tested. Experiments carried out in triple insecticide resistant areas of Gujarat and Maharashtra (Yadava *et al*., 1996) on cyfluthrin, a synthetic pyrethroid, showed it to be an effective and safe insecticide for control of *An. stephensi* and *Cx. quinquefasciatus* with consequent decrease in the vector density and cases of malaria. Mohapatra *et al*. (1999), also evaluated the efficacy of cyfluthrin on these mosquito species and showed it to be ovicidal on *An. stephensi* and *Ae. aegypti* while checked the complete hatchability of eggs in *Cx. quinquefasciatus*. Complete susceptibility of many mosquito species towards many synthetic pyrethroids have also been observed by many authors (Vijayan and Revanna, 1993; Vijayan and Pushpalatha, 1997; Bansal and Singh, 2004). However, resistance to these pyrethroids is also inevitable when mosquito generations either in adult or larval form will be continuously exposed to the selection pressure of these insecticides (Thomas *et al*., 1991; Rajasree and Shetty, 1998). Results of the present study clearly suggest that resistance is developing both in the larval and adult population of different disease vectors. A routine susceptibility monitoring is required in different geographical areas to know the status of resistance and its biochemical basis based on differential insecticidal tolerance exhibited by these mosquito species for a judicious use in the integrated vector management programme.

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