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Development of organophosphorus and carbamate-resistance in Indian strains of *Anopheles stephensi* Liston

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Abstract. Larvae of two Indian strains of *Anopheles stephensi* were highly susceptible to chlorpyrifos, temephos, dichlorvos, fenitrothion, fenthion and malathion but not to carbamates, DDT and γ-HCH. Both strains upon continuous and intense larval selection under laboratory conditions, developed high level of resistance to malathion and moderate levels to fenitrothion, fenthion, and temephos. However, selection with propoxur did not produce resistance in both strains. Larval selection could not induce any tolerance in adults. In general, selection with op-compounds induced cross-tolerance to other op-compounds, organochlorines and carbamates. Malathion-selection caused high cross resistance to fenitrothion while, fenitrothion selection produced high cross-resistance to malathion.

Keywords. *Anopheles stephensi*; larval selection; op-compounds; cross-tolerance; cross-resistance.

1. Introduction

Recent resurgence of malaria in many parts of the world including India is largely attributed to the development of organochlorine-resistance by major vectors of malaria. In India, many strains of *Anopheles stephensi*, an important vector of malaria in urban areas were reported to have become resistant to DDT and γ-HCH since 1955 (Bhombore *et al* 1963; Rajagopalan, *et al* 1956; Rao and Sitaraman 1964; Roy *et al* 1978). Prolonged use of malathion in mosquito control operations has resulted in malathion-resistance in *A. stephensi* in Iran (Manouchehri *et al* 1975, 1976), Iraq (Manouchehri *et al* 1980), Pakistan (Rathor *et al* 1980; Rathor and Toquir 1980) and in *A. culicifacies* from India (Rajagopal 1977; Herath and Davidson 1981a). First instance of a vector of malaria developing multi-resistance came from Central America where field populations of *A. albimanus* became resistant to many op-compounds and carbamates as a result of extensive use of these chemicals in cotton insect control (Ariaratnam and Georghiou 1971). Reports on development of resistance in anopheline mosquitoes are now on an increase and many of them have also developed cross-tolerance to related and unrelated compounds (WHO 1980).

A control programme of malaria involving use of insecticides has to rely largely on the op and carbamate insecticides as no other potent alternatives are available due to many constraints in the development of newer insecticides. Therefore, it was interesting to evaluate how far the op- and carbamates would be useful in controlling populations of *A. stephensi* on a long term basis as and when they are employed in mosquito abatements. With this in view laboratory studies on the speed of selection of two field-collected strains of *A. stephensi* for resistance to malathion, fenitrothion, fenthion, temephos and propoxur and their cross-resistance patterns to other compounds were carried out and the results are reported here.

2. Materials and methods

Strains of *A. stephensi* employed in the present study originated from engorged adult females collected from cattle sheds in Basantpur village in Haryana state and Okhla village in New Delhi during April–May, 1979 and since then maintained in the laboratory at 28 ± 2°C, 80 ± 5% RH and 14 hr of artificial daylight and 10 hr of darkness. The adults were held in cages and the eggs were allowed to hatch in dechlorinated tapwater in enamel bowls. The larvae were reared in large enamel trays (36 × 30 × 5 cm) containing water and provided with finely powdered dog biscuits and yeast in the ratio of 3:2. The rearing water was changed on alternate days until pupation. Under laboratory conditions, pupation was completed in 8 to 15 days. The pupae were collected in small enamelled bowls with water and kept in cloth cages (30 × 30 × 30 cm) for adult emergence. Adults were fed on freshly water-soaked, split raisins. Water was provided in the form of a wet cotton pad kept at the top of the cage. The adults were given 3 days for mating and on the fourth day, females were given blood meal using albino rat. Prior to each blood meal, females were starved completely for 24 hr by removing the raisins from the cages.

The insecticides used were 95 to 98% pure. Chemical names of some of the insecticides employed in the present studies are:

(i) fenthion:	0,0-dimethyl 0-(4-methylthio)-*m* tolyl phosphorothioate.
(ii) temephos:	0,0,0′,0′-tetramethyl 0,0′ thiodi-*p*-phenylene phosphorothioate.
(iii) fenitrothion:	0,0-dimethyl 0-(4-nitro-*m*-tolyl) phosphorothioate.
(iv) chlorpyrifos:	0,0-diethyl 0-(3,5,6-trichloro-2-pyridyl) phosphorothioate.
(v) dichlorvos:	2,2-dichlorothenyl dimethyl phosphate.
(vi) propoxur:	0-isopropoxy phenyl methyl carbamate.
(vii) carbaryl:	1-naphthalenyl methyl carbamate.
(viii) carbofuran:	2,3-dihydro-2,2-dimethyl 7-benzofuranyl methyl carbamate.

The susceptibility of the larvae to various insecticides was determined using early fourth instar larvae (WHO 1981a). In all the tests, larvae of the F_3 generation of the field collected strains were used. All the tests were replicated four times each and the correct percentage mortality was calculated according to Abbott's formula. The LC_{50} and LC_{90} values were determined from dosage-mortality lines plotted on log-probit paper (Codex 320-76). The adult susceptibility was also determined using the standard method for testing adult susceptibility (WHO 1981b). The adults were exposed for different time intervals, *i.e.* for 30, 60, 120, 240 min to diagnostic doses of DDT (4%), dieldrin (0.4%), malathion (5%), fenthion (2·5%), fenitrothion (1%) and propoxur (0·1%) and the mortality was recorded after 24 hr. The LT_{50} and LT_{90} values were calculated from the *d–m* lines plotted on log-probit paper. Tests were performed only on 3-day old, fully blood-fed adult females. Each test had 4 replicates of 20 mosquitoes each with similar controls.

Larval selection was performed by exposing 100 to 150 early fourth instar larvae, in 10 to 12 batches for 24 hr, in glass jars (450 ml) containing 249 ml of water and 1 ml of the required concentration of the insecticide solution in ethanol. The solution was thoroughly stirred and kept for 24 hr. The dosage was adjusted in every generation so as to cause 80 to 90% mortality of the larvae. The surviving larvae were separated and thoroughly rinsed with water and transferred to water-filled trays with food. The adults were blood fed, eggs were collected, hatched and thus the cycle continued. Thus 20 successive generations were selected with propoxur, malathion, fenthion, fenitrothion and temephos. The data were subjected to computer analysis of the regression of probit mortality on log-dosage and the LC_{50}, LC_{90}, slope about the linear regression line were computed using complex mathematical formula (Finney 1971) programmed on an HP 21 mx computer and 100 iterations were performed for each group. Resistance-ratio was calculated by dividing the final LC_{50} value of the insecticide selected strain by LC_{50} of the susceptible parental strain. Cross-resistance of larvae of each of the F_5, F_{10}, F_{15} and F_{20} insecticide selected generations to other insecticides was also determined. The cross-resistance ratio was calculated similarly as the resistance ratio. The data were subjected to regression analysis as described earlier.

3. Results and discussion

Larval susceptibility of Delhi and Haryana strains of *A. stephensi* to 11 insecticides is given in table 1. Moderate tolerance to both DDT and γ-HCH may be due to pre-existing resistance as these compounds are extensively used in mosquito control programmes in India. Among the organophosphates, chlorpyrifos was most toxic followed by temephos. Chlorpyrifos is not commonly used as spray in India but resistance to chlorpyrifos has been reported in *A. sacharovi* from Turkey (Ramsdale *et al* 1980). Temephos though proved lethal to larvae of Delhi and Haryana strains, gave less percentage-mortality in one field strain of *A. stephensi* collected from Madras, India (Roy *et al* 1978). Malathion and fenthion were equitoxic to both Indian strains and

Table 1. Larval LC_{50} and LC_{90} levels (in ppm) of Delhi and Haryana strains of *A. stephensi*.

	Delhi			Haryana		
Insecticide	LC_{50}	LC_{90}	Slope	LC_{50}	LC_{90}	Slope
Malathion	0·004 (0·003–0·005)	0·01 (0·008–0·017)	3·038	0·010 (0·008–0·030)	0·030 (0·022–0·050)	2·856
Fenitrothion	0·002 (0·001–0·002)	0·005 (0·004–0·009)	2·420	0·0025 (0·002–0·003)	0·006 (0·004–0·008)	3·693
Fenthion	0·004 (0·003–0·005)	0·014 (0·009–0·025)	2·290	0·002 (0·001–0·002)	0·005 (0·004–0·007)	3·378
Temephos	0·0008 (0·0006–0·001)	0·002 (0·001–0·003)	3·533	0·0008 (0·0006–0·001)	0·002 (0·001–0·003)	3·202
Chlorpyrifos	0·0002 (0·0002–0·0003)	0·0006 (0·0005–0·001)	2·633	0·0009 (0·0007–0·002)	0·002 (0·001–0·003)	1·800
Dichlorvos	0·003 (0·002–0·007)	0·018 (0·013–0·031)	2·380	0·005 (0·004–0·007)	0·019 (0·014–0·034)	2·277
Propoxur	0·146 (0·117–0·181)	0·379 (0·287–0·577)	3·091	0·207 (0·160–0·273)	0·759 (0·521–1·370)	2·278
Carbaryl	0·212 (0·175–0·255)	0·420 (0·335–0·607)	4·304	0·256 (0·200–0·329)	0·885 (0·635–1·445)	2·382
Carbofuran	0·250 (0·207–0·302)	0·502 (0·400–0·723)	4·221	0·361 (0·307–0·426)	0·607 (0·501–0·846)	5·694
γ-HCH	0·104 (0·078–0·139)	0·498 (0·329–0·931)	1·881	0·134 (0·100–0·184)	0·681 (0·433–1·383)	1·813
DDT	0·175 (0·134–0·227)	0·648 (0·458–1·105)	2·259	0·282 (0·227–0·350)	0·711 (0·542–1·077)	3·191

Figures in parentheses indicate upper and lower fiducial limits.

fenitrothion was 2 × and 4 × as toxic as malathion to Delhi and Haryana strains respectively. Field-resistance to malathion and fenitrothion has appeared in many anopheline species in different areas of the world (Ramsdale *et al* 1980; Ariaratnam and Georghiou 1971; Georghiou 1972; Herath and Davidson 1981c; Rajagopal 1977). Potency of propoxur was greater compared to other two carbamates and DDT but lesser than γ-HCH.

On exposure to discriminating doses of malathion, fenthion, fenitrothion and propoxur adult females of both the strains showed complete mortality in just 30 min of exposure. However, exposure to DDT followed by 24 hr of recovery period gave LT_{50} values as 42 min for both the strains (table 2). LT_{50} values with 0·4 % dieldrin were 50 and 60 min for Delhi and Haryana strains (table 3). It is clear from the present data that the two strains are equally susceptible to the various compounds tested as larvicides and adulticides.

Malathion selection pressure exerted on larvae produced 234 and 86-fold resistance in 20 generations of selection in Delhi and Haryana strains (figures 1 and 2, table 4). The F_{20}-selected generation showed nearly 1·5 × increase in slope value compared to susceptible parental strain. A somewhat similar situation was reported in adults of *A. stephensi* from Pakistan where selection in laboratory induced 20-fold resistance in adults (Rathor and Toquir 1980). Larval-resistance could not produce any appreciable tolerance in adults as 100 % mortality to 5 % malathion was observed in adult females from selected generation. On the other hand, it was observed in *A. arabiensis* and *A.*

Table 2. Susceptibility of adult females of *A. stephensi* to 4 % DDT impregnated paper (diagnostic dose) of the susceptible and insecticide-selected Delhi and Haryana strains (expressed as LT_{50} and LT_{90} in minutes).

Status of females	Delhi			Haryana		
	LT_{50}	LT_{90}	Slope	LT_{50}	LT_{90}	Slope
Parental (susceptible)	42·43	108·74	3·135	42·42	91·68	3·829
F_{20} malathion-selected	62·03	159·84	3·117	14·08	292·14	1·560
F_{20} fenitrothion-selected	52·29	120·90	3·515	68·51	196·92	2·795
F_{20} fenthion-selected	43·06	133·11	2·615	56·98	183·55	2·522
F_{20} temephos-selected	36·53	82·31	3·633	48·09	136·58	2·827
F_{20} propoxur-selected	58·11	147·39	3·170	47·37	130·32	2·196

Table 3. Susceptibility of adult females of *A. stephensi* to 0·4 % dieldrin impregnated paper (diagnostic dose) of the susceptible and insecticide-selected Delhi and Haryana strains (expressed as LT_{50} and LT_{90} in minutes).

Status of females	Delhi			Haryana		
	LT_{50}	LT_{90}	Slope	LT_{50}	LT_{90}	Slope
Parental (susceptible)	56·12	251·53	1·967	50·55	232·57	1·933
F_{20} malathion-selected	80·03	195·73	3·299	42·43	185·34	2·001
F_{20} fenitrothion-selected	37·43	258·16	1·527	33·83	204·08	1·642
F_{20} fenthion-selected	33·38	217·06	1·576	34·40	174·81	1·815
F_{20} temephos-selected	24·86	176·44	1·506	39·01	205·84	1·774
F_{20} propoxur-selected	43·01	222·27	1·796	41·78	111·02	3·019

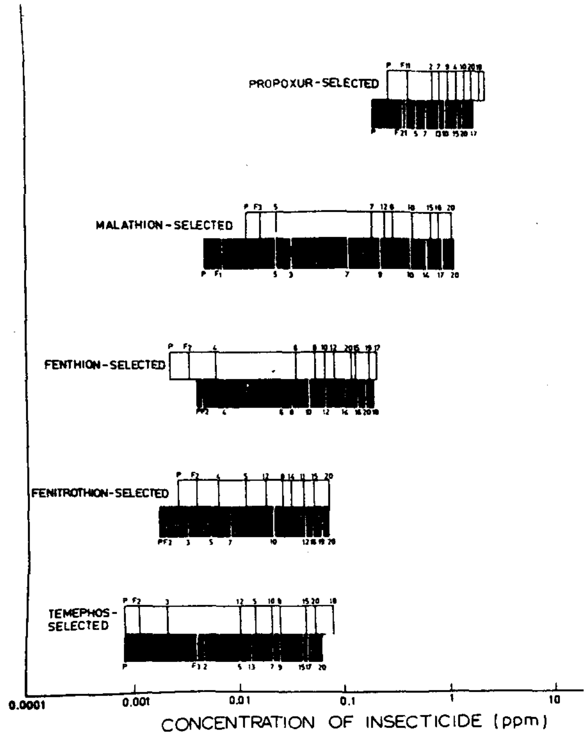

Figure 1. Larval LC_{50} levels of certain generations of Delhi and Haryana strains of *A. stephensi* selected with organophosphorus compounds and carbamates in successive generations in larval stage (▬ Haryana strain; ▭ Delhi strain).

Table 4. F_{20} LC_{50} levels, slope and resistance-ratios of Delhi and Haryana strains of *A. Stephensi* larvae selected with insecticides.

	Delhi			Haryana		
Insecticide	LC_{50} (ppm)	*RR*	Slope	LC_{50} (ppm)	*RR*	Slope
Malathion	0·938 (0·772–1·151)	234·50	3·972	0·866 (0·718–1·050)	86·60	4·250
Fenitrothion	0·073 (0·059–0·091)	36·50	3·309	0·068 (0·055–0·084)	27·20	3·494
Fenthion	0·150 (0·120–0·189)	37·50	3·157	0·143 (0·097–0·161)	71·50	4·326
Temephos	0·052 (0·042–0·066)	65·00	3·193	0·054 (0·043–0·067)	67·50	3·667
Propoxur	1·050 (0·879–1·260)	7·19	4·557	1·160 (0·971–1·400)	5·60	4·387

Figures in parantheses indicate upper and lower fiducial limits.

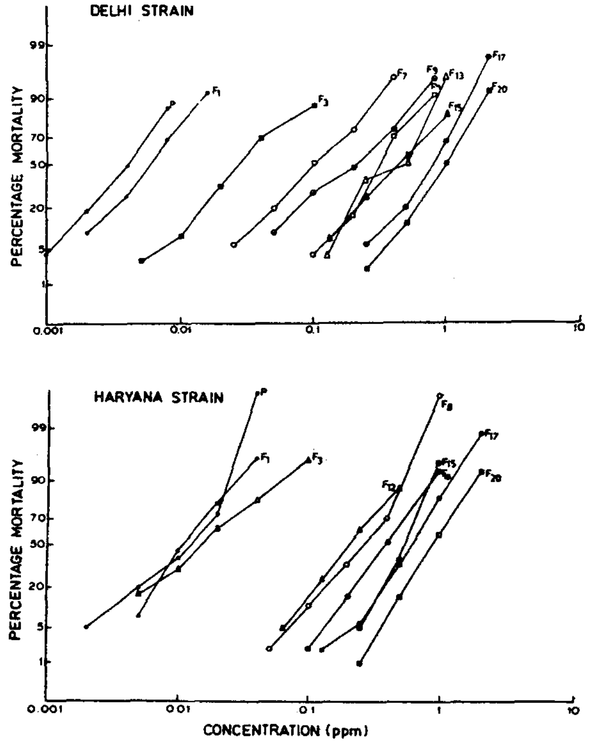

Figure 2. Dosage-mortality relationships of certain generations of *A. stephensi* selected with malathion pressure in larval stage.

atroparvous that malathion-resistance in adults was not extendable to larvae (WHO 1980). Adult females of the F_{20} malathion selected strains were moderately tolerant to 4% DDT ($LT_{50} = 62$ min) and slightly susceptible to 0·4% dieldrin ($LT_{50} = 42$ min) (tables 2 and 3).

Malathion-selection for 20 generations caused 13–14 fold cross-resistance to fenitrothion, 11-fold to γ-HCH and 7-fold to DDT in both strains. No appreciable level of cross-resistance to chlorpyrifos, dichlorvos and carbamates was observed (table 5). This type of cross-resistance to fenitrothion has been observed in malathion-resistant strains of *A. culicifacies* from India (Herath and Davidson 1981d) and *A. stephensi* from Iran (Herath and Davidson 1981b). The co-existence of malathion and fenitrothion-resistance in malathion-selected strains is comparable to natural populations of *A. albimanus* from El Salvador where the larvae acquired 178-fold resistance to malathion and 45-fold to fenitrothion though the latter was less frequently used in the area (Georghiou *et al* 1973).

Delhi and Haryana strains on selection with fenitrothion showed increased LC_{50} levels attaining 36·5 × and 27 × respectively (figure 1, table 4). Marked fluctuations in slope of the dosage-mortality regression lines were observed (figure 3). Adults were completely susceptible to 0·1% fenitrothion and moderately tolerant to 4% DDT (table 2). In strains of *A. albimanus* from Haiti and Panama, intense laboratory selection with fenitrothion could result only in 3 × increase in tolerance (Georghiou and Calman 1969). Fenitrothion-resistance has also been demonstrated in adults of *A. culicifacies*

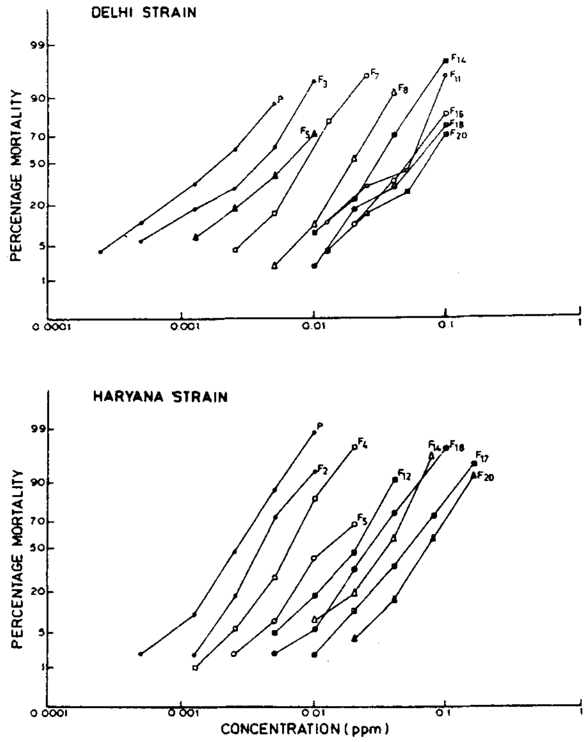

Figure 3. Dosage-mortality relationships of certain generations of *A. stephensi* selected with fenitrothion pressure in larval stage.

from Maharashtra and Gujarat states of India (Herath *et al* 1981; Herath and Davidson 1981a).

Fenitrothion-selection induced 198-fold cross-resistance to malathion for Delhi strain and 90-fold for Haryana strain (table 5). Such a type of cross-resistance was also obtained for *A. culicifacies* from India resistant to fenitrothion (Herath *et al* 1981; Herath and Davidson 1981d). Also, a remarkable degree of resistance to fenthion but low to moderate levels to other op-compounds and carbamates and organochlorines was observed (table 5). Small increases in tolerance to fenthion were observed in *C.p. fatigans* and *A. albimanus* subjected to fenitrothion-selection pressure in the laboratory (Georghiou and Calman 1969).

The Delhi strain when subjected to selection pressure with fenthion for 20 generations picked up 51·5-fold resistance by F_{18} generation (figure 4, table 4). However, the level of resistance slightly declined becoming 37·5 × in F_{20} generation (figures 1 and 4). A very similar pattern was obtained for the Haryana strain also (figure 4, table 4). Fluctuations in slope were evident throughout selection, showing a final increase in F_{20} generation. Adult females were susceptible to diagnostic doses of fenthion, and dieldrin but tolerant to 4 % DDT (tables 2 and 3). Tolerance to fenthion has been reported in a multi-resistant strain of *A. albimanus* from El Salvador (Georghiou *et al* 1972). Tadano and Brown (1966) and Thomas (1970) showed that fenthion-selection in laboratory in *C.p. fatigans* increased the LC_{50} level only by 2 to 4-fold in 22 generations and 2-fold in 10 generations respectively. It is interesting to note from table 5

Table 5. Cross-resistance ratios of Delhi and Haryana strains of *A. stephensi* larvae selected for 20 generations with op-compounds and carbamates. Generation of selection-F_{20}.

Strain	Selecting agent	Compounds tested for cross-resistance										
		Mala-thion	Fenitro-thion	Fenthion	Temephos	Chlor-pyrifos	Dichlor-vos	Pro-poxur	Carbaryl	Carbo-furan	-HCH	DDT
Delhi	Malathion	—	13·52 (4·008)	5·35 (4·366)	11·85 (4·366)	4·85 (3·454)	6·33 (4·032)	4·85 (3·748)	4·62 (3·844)	1·63 (4·588)	11·9 (3·697)	7·05 (2·894)
Haryana		—	14·4 (4·210)	12 (4·334)	23·41 (4·638)	2·05 (3·626)	1·96 (3·252)	3·06 (4·251)	3·89 (4·084)	3·25 (3·857)	5·34 (4·302)	7·57 (3·341)
Delhi	Fenitrothion	198·5 (4·261)	—	54·86 (4·262)	8·89 (3·716)	9 (6·014)	7 (4·426)	4·36 (5·921)	6 (3·782)	2·02 (4·802)	9·86 (3·906)	9·35 (3·573)
Haryana		90·2 (2·858)	—	106·5 (4·664)	9·51 (4·093)	2·11 (3·177)	3·54 (3·349)	3·14 (4·112)	5·07 (3·231)	1·79 (3·101)	8·17 (3·856)	8·45 (3·856)
Delhi	Fenthion	116·25 (2·658)	17·64 (3·950)	—	9·26 (3·567)	12·5 (2·505)	8·9 (3·550)	4·96 (3·964)	4·95 (3·121)	1·87 (4·371)	19·6 (3·758)	8·94 (3·339)
Haryana		35·7 (3·707)	11·6 (4·812)	—	10·01 (3·188)	3·11 (4·792)	4 (4·802)	2·72 (3·588)	2·63 (3·272)	2·23 (4·246)	19·84 (3·303)	5·35 (2·929)
Delhi	Temephos	42·5 (2·946)	12·94 (1·785)	67·02 (4·005)	—	4·3 (4·695)	2·76 (2·612)	0·17 (4·095)	4·7 (4·802)	1·36 (2·571)	2·28 (2·457)	21·64 (5·610)
Haryana		25·6 (4·422)	10 (6·021)	196·5 (3·586)	—	0·87 (3·698)	1·6 (5·070)	2·65 (3·918)	3·06 (3·101)	1·1 (4·610)	5·09 (5·513)	12·32 (3·753)
Delhi	Propoxur	38·25 (1·676)	11·64 (3·471)	67·83 (4·261)	57·31 (4·681)	6·35 (3·713)	3·2 (2·061)	—	6·23 (3·628)	1·6 (4·135)	4·9 (3·790)	29·73 (5·821)
Haryana		19·6 (4·547)	7·08 (3·739)	104 (3·836)	34·14 (5·334)	1·33 (3·244)	4 (4·025)	—	3·2 (3·681)	0·95 (4·628)	5·3 (4·514)	14·46 (5·338)

Figures in parantheses indicate slope.

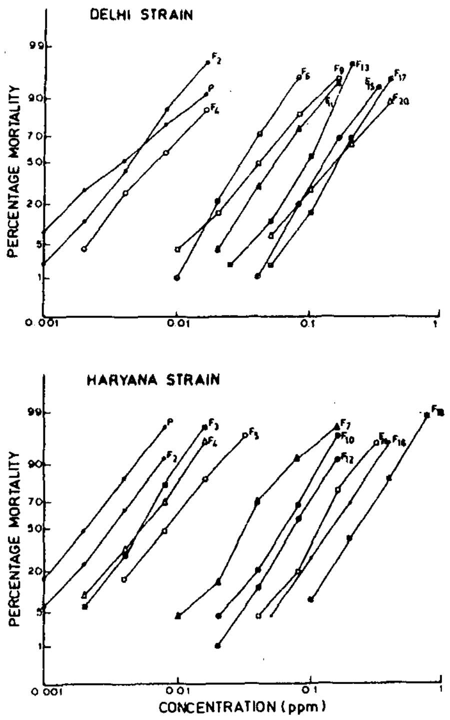

Figure 4. Dosage-mortality relationships of certain generations of *A. stephensi* selected with fenthion pressure in larval stage.

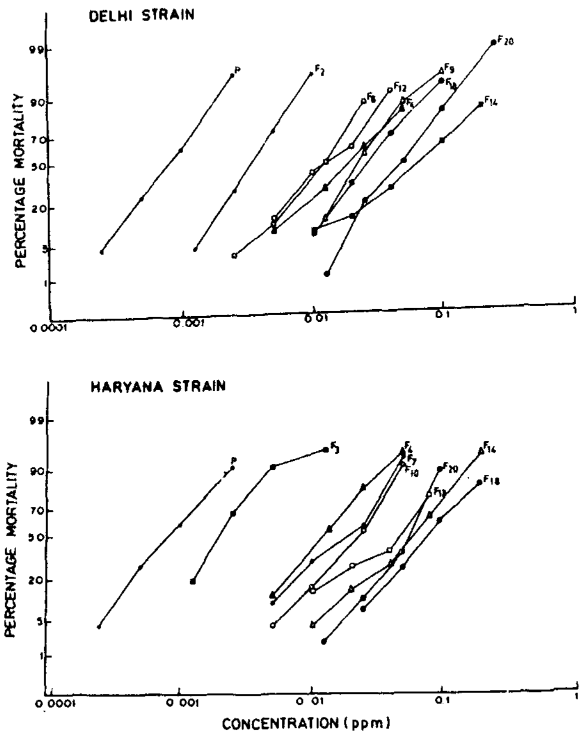

Figure 5. Dosage-mortality relationships of certain generations of *A. stephensi* selected with temephos pressure in larval stage.

that fenthion-resistance induced maximum cross-resistance to malathion as compared to any other op-compound tested. A high rate of tolerance to γ-HCH and moderate levels to DDT was observed for both strains.

From the dosage-mortality regression lines of temephos-selected strains (figure 5) it is evident that the lines of successive selected generations moved to the right thereby the level of resistance increased to 65-fold for Delhi strain and 67·5-fold for Haryana strain (table 4). Slope values showed fluctuations with less appreciable difference in the F_{20} generation compared to parental generation of both the strains (table 4). A much higher level of resistance resulted from laboratory selection on larvae of *C.p. quinquefasciatus* up to 322-fold in 9 generations was obtained by Ranasinghe' and Georghiou (1976). Adult females were more susceptible to diagnostic dose of dieldrin and less to DDT (tables 2 and 3). Temephos-selection induced very high tolerance to fenthion and malathion followed by fenitrothion and DDT indicating development of resistance to compounds with non-identical structures (table 5).

As compared to the levels of resistance induced by op-compounds, propoxur could not induce noticeable tolerance in larvae of two strains tested. Variations in slope were not significant (figures 1 and 6, table 4). On the contrary, intense selection with propoxur has resulted in 100 × increase in larval LC_{50} in a population of *A. albimanus* from El Salvador (Ariaratnam and Georghiou 1971) and in *C.p.fatigans* (Georghiou *et al* 1966), but not in Haiti strain (Georghiou and Calman 1969) and *C. tarsalis* from Coachella valley, California (Georghiou *et al* 1974). Propoxur-selection induced high cross-resistance to fenthion, malathion, fenitrothion, temephos and DDT but not to

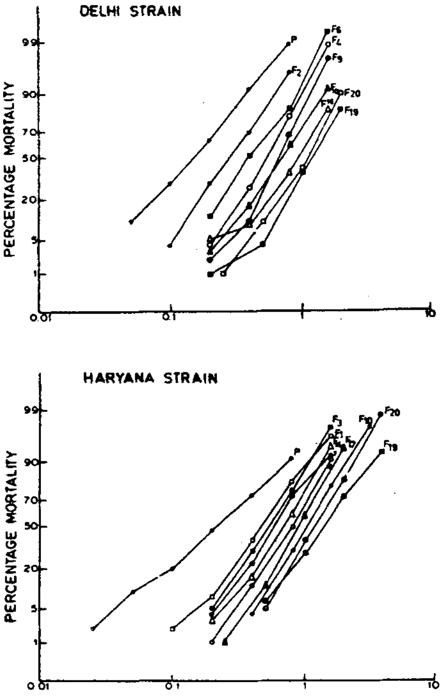

Figure 6. Dosage-mortality relationships of certain generations of *A. stephensi* selected with propoxur pressure in larval stage.

other carbamates. In contrast to present observations, propoxur-selected strains exhibited a broad spectrum of tolerance to other carbamates in a El Salvador population of *A. albimanus* (Ariaratnam and Georghiou 1971) and in *C.p. fatigans* (Georghiou *et al* 1966).

It is evident from the present data that the Indian strains of *A. stephensi* are potentially capable of developing high levels of resistance to malathion under laboratory conditions and moderate levels of tolerance to fenitrothion, fenthion and temephos, a situation well in agreement with multiple-field resistance reported. From dosage-mortality regression lines it is clear that the slope values of successive generations of selection with op and carbamate compounds fluctuated with no appreciable increase in the F_{20} selected generation. The probable reason may be that the population is heterozygous and may not be uniformly responding to the selecting agent. The observed changes may be due to slow reorganization of genetic factors involved in the development of resistance. The response to increasing doses of toxicant in successive generations is slow and the factors involved may not be all that powerful to induce rapid resistance.

Another factor is the non-extendability of larval-resistance to adults as the female adults of the selected strains were susceptible to discriminating doses of the compounds tested. This may be due to stage-specific resistance mechanisms or simply the adults and larvae may have different mechanisms of resistance operating. This point is of great interest in field control of mosquitoes. The third important factor that emerges from our studies is the various types of cross-resistance patterns obtained. Most remarkable is the cross-resistances between fenitrothion and malathion which are structurally unrelated thus reducing the choice of one of them being used as alternative in mosquito control. Besides, it was also observed that strains selected with propoxur, fenthion, fenitrothion and temephos showed remarkable tolerance to malathion and the reverse was not true. A similar situation where compounds with disparate structures showing cross-resistance to each other has been encountered in parathion-selected strains of *A. albimanus* showing unusual tolerances to other op compounds (Ayad and Georghiou 1979).

It may be concluded from the present studies that the Indian strains of *A. stephensi* have the innate ability to develop resistance to op-compounds but not to propoxur. The resistance develops very slowly in larvae and does not extend to adults. These selected strains are capable of developing cross-tolerances to other similar compounds. These findings also suggest that the long term efficacy of these op compounds as alternatives in mosquito control programmes is very limited.

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