

# THE SUSCEPTIBILITY OF MOSQUITO LARVAE TO EIGHTEEN INSECTICIDES IN CZECHOSLOVAKIA

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**ABSTRACT.** During 1971–1974 the susceptibility of the IVth larval instar of *Aedes cantans*, *Ae. vexans*, *Ae. punctor*, *Ae. excrucians*, *Ae. communis*, *Ae. sticticus*, *Culex pipiens pipiens*, *Cx. pipiens molestus* and *Culiseta annulata* to some organophosphates, carbamates and chlorinated hydrocarbons was estimated by means of the WHO method. The susceptibility to organophosphates such as chlorpyrifos, temephos, fenitrothion, fenitrothion, pirimiphos-methyl and bromophos was very high (LC 50 0.001–0.01 ppm), whereas dichlor-

vos, malathion, diazinon and trichlorphon were less toxic (LC 50 0.03–0.24 ppm). Carbamates such as carbaryl, propoxur and bendiocarb were of even lower toxicity (LC 50 0.09–0.38 ppm).

No resistance to chlorinated hydrocarbons used in mosquito control in Czechoslovakia, such as DDT and lindane, was detected in the larvae studied. Of the chlorinated hydrocarbons methoxychlor was found in general to be less toxic than the rest.

**INTRODUCTION.** Mosquitoes, regardless of their medical importance, are a great nuisance to humans in low lying areas near the larger rivers of Czechoslovakia (Trpiš 1962, Palička 1967, Hájková and Minář 1970, Rettich 1971a). *Aedes cantans* (Meig.), *Ae. communis* (Deg.), *Ae. sticticus* (Meig.) from flood-plain forests, *Ae. punctor* (Kirby) from coniferous forests and peat-bogs and *Ae. vexans* (Meig.) and *Ae. excrucians* (Walk.) from flooded meadows are the most important species. *Culex pipiens molestus* (Forsk) causes nuisance in urban areas. The females of *Cx. pipiens pipiens* (L.) hibernate "en masse" in cellars and *Culiseta annulata* (Schrk.) females are a nuisance to livestock.

Until the late sixties DDT was used widely for killing culicine mosquitoes. Apprehension that resistance might develop in frequently treated territories (Rettich 1971a) and especially toxicological reasons led to the rapid introduction of new insecticides. For control of mosquito larvae fenitrothion and pirimiphos-methyl are important recent introductions.

The aim of the present study was to ascertain the baseline susceptibility of mosquito larvae to insecticides currently used and in development. The results of the study should serve both as an aid in selecting the most toxic insecticides for mosquito larvae and for evaluating the possible future development of resistance.

**MATERIALS AND METHODS.** Larvae of the

mosquito species to be tested were caught in various localities of Central Bohemia (CB), East Bohemia (EB), South Bohemia (SB) and South Moravia (SM) during 1971–1974. *Ae. cantans* larvae were caught in temporary and permanent breeding places in the meadow forest of the flooded regions of the rivers Elbe, Lužnice and Morava in Poděbrady (CB—April 1973, April 1974), Velký Oske (CB—April 1971), Kolin (CB—April 1974), Vlkov (SB—May 1972), Lomnice (SB—May 1973), Hodonín (SM—April 1972) and Lanžhot (SM—April 1974). Larvae of *Ae. communis* (Hodonín—April 1973), *Ae. sticticus* (Poděbrady—June 1971) and *Cs. annulata* (Poděbrady—September 1974) were caught in similar breeding places. *Ae. vexans* larvae were caught in flooded meadows of the localities Hradec Králové (EB—June 1974), České Budějovice (SB—June 1972), Hodonín (SM—April 1972) and Nové Mlýny (SM—May 1973). *Ae. excrucians* larvae were caught in flooded meadows around Pavlov (SM—May 1973). *Ae. punctor* larvae were caught in breeding places in peatbogs in the coniferous forest near Červené Blato (SB—May 1972). *Cx. pipiens pipiens* larvae were obtained from rain water tanks and puddles on roads in Praha (CB—August 1972), July 1973), Zdíby (CB—September 1971), Nová Rabyň (CB—September 1973), Hradec Králové (EB—August, September 1974), Vlkov (SB—September

1971) and Val (SB—September 1974). Larvae of *Cx. pipiens molestus* were found in a sewage water tank and then bred in the laboratory. The areas around Poděbrady and Lanžhot were treated with DDT and lindane from time to time.

A wire strainer was used to collect 4th instar larvae in the field. The larvae were carried to the laboratory in cooled PVC flasks. On arrival they were immediately tested using the WHO standard method (WHO 1963). The large number of larvae of one species from one locality allowed many tests to be done over more than 1 day. Larvae of *Ae. cantans*, *Ae. vexans*, *Ae. punctator*, *Cx. pipiens pipiens*, *Cx. pipiens molestus* and *Cs. annulata* from the localities mentioned above were tested against the organophosphates chlorpyrifos, temephos (Abate), fenitrothion, fenthion, primiphos-methyl, bromophos, diazinon, dichlorvos and trichlorphon; the carbamates carbaryl, propoxur (Baygon) and bendiocarb and the chlorinated hydrocarbons DDT, lindane (gamma BHC), dieldrin, endrin and methoxychlor. Larvae of *Ae. communis*, *Ae. excrucians* and *Ae. sticticus* were tested against the insecticides as given in tables 2 and 3.

Minor modifications of the WHO standard test were introduced. Instead of glass jars, 250 ml polystyrene cups were used. Ethanol solutions of pure or technical grade insecticides (w/v) were prepared just before the tests were carried out. In each test 5 or 6 concentrations forming a geometrical series with a quotient of 1.5 were usually prepared. Each concentration with 25 larvae was run in triplicate. The temperature of distilled water (in field laboratories that of tap water) was maintained at 20–23°C in the course of a 24 hours' test. The mortality counts were made according to instructions of the WHO method (WHO 1963).

RESULTS AND DISCUSSION. The results obtained are summarized in Tables 1–5, which show, in the case of *Ae. cantans*, *Ae. vexans* and *Cx. pipiens pipiens* the average LC 50's and LC 90's and the spread of results obtained.

*Ae. cantans* (Table 1). A total of 18 insecticides was tested against larvae of *Ae. cantans*. The larvae were found to be most susceptible to chlorpyrifos and fenthion (average LC 50:0.0011 and 0.0027 ppm respectively). The larvae were very susceptible to temephos, lindane and fenitro-

Table 1. The susceptibility of 4th instar larvae of *Aedes cantans* (Meig.) from 8 localities to several insecticides. Average LC 50 and LC 90 values and (in parentheses) spread of results obtained in ppm.

Insecticide	LC 50	LC 90
chlorpyrifos	0.0011(0.0007–0.0017)	0.0020(0.0011–0.0042)
fenthion	0.0027(0.0005–0.0057)	0.0043(0.0012–0.0078)
temephos	0.0043(0.0019–0.0060)	0.0067(0.0027–0.0129)
lindane	0.0047(0.0024–0.0092)	0.0096(0.0063–0.0195)
fenitrothion	0.0059(0.0023–0.0093)	0.0101(0.0063–0.0187)
bromophos	0.0086(0.0040–0.0130)	0.0133(0.0066–0.0200)
dieldrin	0.0091(0.0046–0.0117)	0.0190(0.0146–0.0270)
DDT	0.0102(0.0063–0.0174)	0.0253(0.0111–0.0724)
primiphos-methyl	0.0103(0.0036–0.0144)	0.0165(0.0088–0.0242)
endrin	0.0107(0.0079–0.0153)	0.0187(0.0133–0.0272)
methoxychlor	0.0315(0.0221–0.0570)	0.0530(0.0354–0.1038)
diazinon	0.0356(0.0090–0.0521)	0.0511(0.0147–0.0671)
malathion	0.0488(0.0249–0.0741)	0.1622(0.0524–0.7181)
dichlorvos	0.0504(0.0157–0.0779)	0.0749(0.0366–0.1186)
trichlorphon	0.0724(0.0219–0.1423)	0.1264(0.0642–0.2055)
propoxur	0.1112(0.0452–0.1827)	0.2102(0.0709–0.3917)
bendiocarb	0.1751(0.0841–0.2409)	0.3192(0.1996–0.4032)
carbaryl	0.3766(0.2099–0.6826)	0.8361(0.335–2.2325)

thion (average LC 50s: 0.0043, 0.0047, 0.0059 ppm respectively). Bromophos, pirimiphos-methyl, dieldrin, DDT and endrin were also appreciably toxic (LC 50 range: 0.0086–0.0107 ppm). Diazinon, malathion, dichlorvos and trichlorphon were distinctly less toxic (LC 50 range: 0.036–0.072 ppm). The susceptibility of *Ae. cantans* larvae to the carbamates propoxur, bendiocarb and carbaryl was very low (LC 50 range: 0.11–0.38 ppm). The susceptibility to chlorinated hydrocarbons and fenitrothion correspond with those determined earlier (Rettich 1974), the susceptibility of these larvae to DDT, dieldrin, methoxychlor, chlorpyrifos, temephos, fenthion, diazinon and propoxur is similar to those for *Ae. taeniorhynchus* larvae found by Armstrong in America. (1971).

*Aedes vexans* (Table 2). Of 16 insecticides tested *Ae. vexans* larvae were most susceptible to chlorpyrifos (average LC 50: 0.001 ppm). They were found also to be highly susceptible to DDT, dieldrin, temephos, fenitrothion, fenthion, lindane, pirimiphos-methyl and endrin (average LC 50: 0.0019, 0.0021, 0.0027, 0.0039, 0.0041, 0.0047, 0.0052 and 0.0072 ppm respectively). Bromophos, dichlorvos, malathion and diazinon were less

toxic (LC 50 range: 0.01–0.04 ppm). Susceptibility of the larvae to trichlorphon, propoxur and carbaryl was very low (LC 50 range: 0.1–0.32 ppm). The susceptibility of these larvae to chlorinated hydrocarbons and fenitrothion corresponds with our earlier findings (Rettich 1974), and the susceptibility to malathion, fenthion, chlorpyrifos and temephos conforms with the findings of Gillies et al. (1971) concerning *Ae. vexans* in U.S.A.

*Ae. excrucians* (Table 3). Of 10 insecticides tested the larvae were found to be almost equally susceptible to fenthion, chlorpyrifos, temephos, fenitrothion, bromophos, dieldrin and DDT (LC 50 range: 0.0032–0.0063 ppm). Malathion and trichlorphon were distinctly less toxic (LC 50: 0.03–0.042 ppm respectively). Carbaryl proved to be the least toxic insecticide (LC 50: 0.15 ppm).

*Ae. communis* (Table 3). Larvae of *Ae. communis* were tested against 9 insecticides and were very susceptible to chlorpyrifos, fenthion, temephos and fenitrothion (LC 50: 0.0012, 0.0035, 0.0046 and 0.0047 respectively). The LC 50's of DDT, bromophos and malathion were 0.009, 0.0013 and 0.038 ppm respectively. Susceptibility to trichlorphon and carbaryl was low (LC 50: 0.14 and 0.17 ppm respec-

Table 2. The susceptibility of 4th instar larvae of *Aedes vexans* (Meig.) from 4 localities to several insecticides. Average LC 50 and LC 90 values and (in parentheses) the spread of results obtained in ppm.

Insecticide	LC 50	LC 90
chlorpyrifos	0.0010(0.0004–0.0018)	0.0021(0.0011–0.0029)
DDT	0.0019(0.0013–0.0030)	0.0054(0.0025–0.0096)
dieldrin	0.0021(0.0014–0.0026)	0.0035(0.0023–0.0048)
temephos	0.0027(0.0020–0.0037)	0.0045(0.0032–0.0063)
fenitrothion	0.0039(0.0021–0.0052)	0.0060(0.0028–0.0086)
fenthion	0.0041(0.0020–0.0064)	0.0089(0.0028–0.0205)
lindane	0.0047(0.0024–0.0076)	0.0089(0.0051–0.0147)
pirimiphos-methyl	0.0052(one test only)	0.0123
endrin	0.0072(0.0032–0.0109)	0.0141(0.0105–0.0216)
bromophos	0.0103(0.0034–0.0165)	0.0219(0.0075–0.0443)
dichlorvos	0.0261(0.0171–0.0308)	0.0397(0.0248–0.0514)
malathion	0.0261(0.0238–0.0285)	0.0481(0.0430–0.0533)
diazinon	0.0379(0.0266–0.0493)	0.0612(0.0569–0.0656)
trichlorphon	0.1047(0.0440–0.2673)	0.1807(0.0731–0.4687)
propoxur	0.1640(0.0964–0.2316)	0.3034(0.1839–0.4229)
carbaryl	0.3226(0.0739–0.6331)	0.8699(0.4270–1.3521)

Table 3. The susceptibility of 4th instar larvae of *Aedes excrucians* (Walk.), *Ae. communis* (Deg.) and *Ae. sticticus* (Meig.) to several insecticides. LC 50 and LC 90 values in ppm.

Insecticide	<i>Aedes excrucians</i>		<i>Aedes communis</i>		<i>Aedes sticticus</i>	
	LC 50	LC 90	LC 50	LC 90	LC 50	LC 90
fenthion	0.0032	0.0049	0.0035	0.0058	0.0005	0.0007
chlorpyrifos	0.0033	0.0046	0.0012	0.0021	0.0005	0.0007
temephos	0.0035	0.0053	0.0046	0.0058	0.0038	0.0053
fenitrothion	0.0039	0.0069	0.0047	0.0074	0.0011	0.0018
bromophos	0.0043	0.0091	0.0127	0.0169	0.0028	0.0044
dieldrin	0.0047	0.0172	—	—	0.0016	0.0023
DDT	0.0063	0.0155	0.0090	0.0129	0.0026	0.0037
malathion	0.0303	0.0594	0.0382	0.0660	0.0155	0.0230
trichlorphon	0.0423	0.0680	0.1397	0.1824	—	—
carbaryl	0.1455	0.6167	0.1679	0.5898	—	—

tively). Our figures for susceptibility of *Ae. communis* larvae to DDT are distinctly lower than those indicated by Ungureanu and Theodorescu (1961) from Roumania.

*Ae. sticticus* (Table 3). *Ae. sticticus* larvae were more susceptible to all the 8 insecticides tested than the other species of *Aedes*. Chlorpyrifos and fenthion were the most toxic insecticides for *Ae. sticticus* larvae (LC 50: 0.0005 ppm). The susceptibility to fenitrothion, dieldrin, DDT, bromophos, and temephos was very high (LC 50: 0.0011, 0.0016, 0.0026, 0.0028

and 0.0038 ppm respectively). The LC 50 value for malathion was 0.0155 ppm. Ungureanu and Theodorescu (1961) found somewhat lower values of susceptibility to DDT (LC 50: 0.006 ppm) for these larvae.

*Ae. punctor* (Table 5). The susceptibility of larvae of this species to the 18 insecticides tested resembles roughly that of *Ae. cantans*. Larvae were found to be most susceptible to chlorpyrifos (LC 50: 0.0027 ppm). Susceptibility to dichlorvos, malathion and diazinon was distinctly lower (LC 50 range: 0.044–0.07 ppm) than the

Table 4. The susceptibility of 4th instar larvae of *Culex pipiens pipiens* (L.) from 7 localities to several insecticides. Average LC 50 and LC 90 values and (in parentheses) spread of results obtained in ppm.

Insecticide	LC 50	LC 90
chlorpyrifos	0.0016(0.0005–0.0029)	0.0028(0.0007–0.0052)
temephos	0.0016(0.0005–0.0053)	0.0028(0.0012–0.0089)
dieldrin	0.0036(0.0016–0.0115)	0.0101(0.0025–0.0421)
pirimiphos-methyl	0.0041(0.0021–0.0074)	0.0126(0.0034–0.0411)
fenthion	0.0042(0.0011–0.0079)	0.0060(0.0020–0.0107)
bromophos	0.0053(0.0031–0.0117)	0.0119(0.0060–0.0280)
DDT	0.0057(0.0015–0.0107)	0.0117(0.0030–0.0236)
fenitrothion	0.0068(0.0020–0.0144)	0.0113(0.0051–0.0227)
endrin	0.0084(0.0021–0.0203)	0.0172(0.0037–0.0455)
methoxychlor	0.0089(0.0046–0.0162)	0.0155(0.0075–0.0292)
lindane	0.0180(0.0075–0.0439)	0.0337(0.0151–0.0859)
diazinon	0.0243(0.0077–0.0657)	0.0357(0.0144–0.0862)
dichlorvos	0.0303(0.0154–0.0562)	0.0468(0.0250–0.0870)
malathion	0.0322(0.0088–0.0656)	0.0513(0.0144–0.1106)
bendiocarb	0.0570(0.0518–0.0633)	0.0896(0.0721–0.1083)
propoxur	0.0894(0.0465–0.1180)	0.1903(0.0781–0.3853)
trichlorphon	0.0946(0.0564–0.1526)	0.1467(0.1084–0.2482)
carbaryl	0.3339(0.0677–0.8898)	0.5908(0.1220–1.5073)

other organophosphates (LC 50 range: 0.006–0.009 ppm). The carbamates bendiocarb, propoxur and carbaryl and the organophosphate trichlorphon had a very low toxicity (LC 50 range: 0.09–0.3 ppm).

*Cx. pipiens pipiens* (Table 4). From among the 18 insecticides tested against *Cx. pipiens* larvae, chlorpyrifos and temephos were the most toxic insecticides with an average LC 50 value of 0.0016 ppm. Larval susceptibility to dieldrin, pirimiphos-methyl, fenthion, bromophos, DDT, fenitrothion, endrin and methoxychlor was also very high, the average LC 50 value ranging from 0.0036 to 0.0089 ppm. The toxicity of diazinon, dichlorvos, malathion, and especially trichlorphon was markedly lower (average LC 50: 0.024, 0.03, 0.032 and 0.095 ppm respectively). Curiously enough the average susceptibility to lindane was also lower (LC 50 0.018 ppm). Of the three carbamates tested bendiocarb and propoxur were distinctly more toxic than carbaryl (average LC 50: 0.057, 0.089 and 0.33 ppm respectively). Susceptibility values of organophosphates such as chlorpyrifos, temephos, bromophos, fenthion, feni-

trothion and diazinon are very close to those found by Gras and Rioux (1968) for larvae of this species in France. These authors, however, found much lower susceptibility to DDT, trichlorphon and carbaryl. The susceptibility of *Cx. pipiens* larvae to fenthion, fenitrothion and malathion resembles also the findings of Tomasucci and Michieli (1968) with the same species in Italy, but their susceptibility levels for DDT and dieldrin were lower.

*Cx. pipiens molestus* (Table 5). The susceptibility of *Cx. p. molestus* larvae to all organophosphates and chlorinated hydrocarbons tested was almost equal to that of *Cx. p. pipiens* larvae. Naturally low susceptibility to lindane resembles that of *Cx. p. pipiens* larvae. The susceptibility of *Cx. molestus* to carbamates was lower than that of *Cx. p. pipiens* (LC 50: 0.1–0.4 ppm). The susceptibility of *Cx. p. molestus* larvae to the chlorinated hydrocarbons DDT, dieldrin and lindane was almost similar to that of susceptible strains of this species in Israel reported by Barkai et al. (1967). On the other hand these authors found higher susceptibility to malathion and diazinon.

*Cx. annulata* (Table 5). The susceptibility

Table 5. The susceptibility of 4th instar larvae of *Aedes punctor* (Kirby), *Culex pipiens molestus* (Forsk.) and *Culiseta annulata* (Schrk.) to several insecticides. LC 50 and LC 90 values in ppm.

Insecticide	<i>Aedes punctor</i>		<i>Culex pipiens molestus</i>		<i>Culiseta annulata</i>	
	LC 50	LC 90	LC 50	LC 90	LC 50	LC 90
chloryprifos	0.0027	0.0050	0.0012	0.0016	0.0035	0.0053
bromophos	0.0056	0.0122	0.0033	0.0055	0.0139	0.0251
temephos	0.0057	0.0085	0.0013	0.0021	0.0058	0.0112
fenthion	0.0057	0.0078	0.0038	0.0059	0.0050	0.0073
fenitrothion	0.0059	0.0113	0.0073	0.0110	0.0079	0.0142
pirimiphos-methyl	0.0088	0.0143	0.0061	0.0125	0.0022	0.0031
lindane	0.0104	0.0265	0.0306	0.1420	0.0188	0.0528
endrin	0.0141	0.0236	0.0097	0.0238	0.0116	0.0195
dieldrin	0.0163	0.0302	0.0040	0.0074	0.0136	0.0240
DDT	0.0280	0.0773	0.0061	0.0115	0.0206	0.0577
malathion	0.0441	0.0811	0.0342	0.0530	0.0245	0.0398
dichlorvos	0.0458	0.0616	0.0335	0.0602	0.0662	0.1290
methoxychlor	0.0631	0.1096	0.0189	0.0311	0.0383	0.0661
diazinon	0.0695	0.1227	0.0308	0.0635	0.0623	0.1046
bendiocarb	0.0887	0.3361	0.0965	0.1599	0.1782	0.3425
trichlorphon	0.1782	0.2972	0.0582	0.0872	0.2436	0.3373
propoxur	0.2065	0.3529	0.2396	0.4198	0.2247	0.8137
carbaryl	0.2983	0.7458	0.4188	0.8574	0.1795	0.3352

to the 18 insecticides tested is roughly similar to that of *Ae. cantans* and *Ae. punctor*, the susceptibility to pirimiphos-methyl being outstandingly higher (LC 50:0.0022 ppm—0.01 ppm for *Ae. cantans*, 0.009 ppm for *Ae. punctor*). The LC 50 values of chlorpyrifos, temephos, fenthion and fenitrothion were 0.0004–0.008 ppm. The LC 50 values for chlorinated hydrocarbons, bromophos and malathion were 0.012–0.038 ppm. The LC 50 values of diazinon and dichlorvos were about 0.06–0.07. The toxicity of the carbamates and trichlorphon was low (LC 50:0.18–0.24 ppm.).

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