# Evaluation of some insecticides against *Culex pipiens*, the dominant mosquito species in Abha city

Khatir M. K. Ahmed<sup>1\*</sup>, Sufyan . H. Abdulah<sup>2</sup>, Nayla Mohammed<sup>3</sup>

<sup>1</sup>Dept. of Environmental Health, University of Bahri., Khartoum (Sudan).
 <sup>2</sup>Manistary of health South Kordofan states (Sudan)
 <sup>3</sup>Dept. of Epidemiology, University of Bahri., Khartoum (Sudan).
 \*Correspondence author: Khatir M. K. Ahmed

Abstract— The present study was planned to test on certain chemical insecticides from different groups such as organophosphates, pyrethroids and Bioinsecticide. Among these compounds two were chemical insecticides (Propetamphos and Cypermethrin), while the third one is Bioinsecticide (Baciloid 5000: Bacillus thuringiensis) against Culex pipiens, the dominant mosquito species in Abha city. Taking LC50 values (concentration which to kill 50% of mosquito larvae) into consideration, mosquito larvae of CX. Pipes against Propetamphos was effective (LC50 0.0162 ppm) against the 3rd instar of the laboratory strain comparing with a field strain which was 0.0442 ppm. At LC90 level, data indicated that Propetamphos (LC90 0.8109 ppm) was effective insecticide against the 3rd instar larvae of laboratory strain, while against field strain gave (LC90 3.31 ppm). Similarly, the results clearly showed that Cypermethrin was also very effective insecticide (LC50 0.0132 ppm) against the adult females against laboratory strain, while against field strain Cypermethrin gave (LC50 0.1192 ppm). On the other hand, the residual activity of Bacillus thuringiensis var. israelensis reached from 4 to 20 days of concentrations ranged between 0.001 to 100 p.m. in case laboratory strain, while reaching between 6-23 days of concentrations ranged between 0.001 to 100 p.m. in case field strain. Mortality percent was also found between 11.7 to 96.8% of concentrations ranged between 0.001 to 100 p.m. against laboratory strain in the 1st week, whereas ranged between 0.0 to 70.8% in the 2nd week against the same concentrations finally ranged between 0.0 to 12.7% in the 3rd week. Hence, the field collected larvae of Culex pipens were more susceptible and have prolonged residual effect as compared to laboratory reared.

Keywords— Evaluation of insecticides against Culex pipiens, in Abha city.

#### I. INTRODUCTION

Mosquitoes (Diptera: Culicidae) cause more human suffering than any other organism. Mosquitoes can be an annoying, serious problem in man's domain. They interfere with work and spoil hours of leisure time. Millions of people die from mosquito-borne diseases every year. Mosquito-borne diseases are responsible for a significant fraction of the global disease burden and have profound effects not only on health but also on the socioeconomic development of affected nations. An econometric model for malaria, which is responsible for more than 1 million deaths every year, suggests that countries with intensive malaria have income levels only 33% of that of those without malaria (WHO, 2004). Not only can mosquitoes carry diseases that afflict humans, such as malaria, yellow fever, dengue, filariasis and encephalitis, but they also transmit several diseases and parasites that cattle, dogs and horses are very susceptible to, and also affect humans. These include Rift Valley Fever (RVF), dog heartworm, West Nile virus (WNV) and Eastern Equine Encephalitis (EEE). Their attacks on farm animals can cause loss of weight and decreased milk production. RVF epidemic occurred in Saudi Arabia from August 2000 through September 2001 caused a total of 886 reported cases, and mortality rate was 13.9 % (Madani et al., 2003). In addition, mosquito bites can cause severe skin irritation through an allergic reaction to the mosquito's saliva - this is what causes the red bump and itching. Mosquitoes cause more human suffering than any other organism over one million people worldwide die from mosquito-borne diseases every year. Not only can mosquitoes carry diseases that afflict humans, they also transmit several diseases and parasites that dogs and horses are very susceptible to. These include dog heartworm, West Nile virus (WNV) and Eastern equine encephalitis (EEE). Mosquitoes are among the most serious insect pests of medical importance. They are vectors of various disease agents, some of which cause millions of cases of illnesses and deaths in man and animals each year. Among these diseases, malaria, yellow fever, dengue and dengue fever. hemorrhagic encephalitides, filariasis. dog heatworm, Rift Valley fever (RVF) and others prevail in endemic and epidemic areas in many countries in addition, mosquito bites can cause severe skin irritation through an allergic reaction to the mosquito's saliva - this is what causes the red bump and itching (WHO 1991 and Lerdthusnee et al., 1995). Countries in the Eastern Mediterranean Region of the World Health Organization (WHO), including the Kingdom of Saudi Arabia (KSA) bear ~11% of the world vector borne disease burden like malaria and arboviral diseases (WHO, 2004). Among mosquito species, Culex pipiens (Linnaeus) (the common house mosquito) is a species of blood-feeding mosquito of the family Culicidae. It is a vector of some diseases, such as Japanese encephalitis, meningitis, and urticaria and widely distributed species across Saudi Arabia has been incriminated as main vector of bancroftian filariasis (Southgate, 1979). Culex pipiens can be found in a fairly wide range of larval habitats but are generally associated with water that has a high organic content. The species utilizes temporary ground water that ranges from mildly to grossly polluted. The species also deposits its eggs in artificial containers including tin cans, tires and any refuse that allows stagnant water to puddle. The species is decidedly urban and reaches greatest numbers in large urban centers. Catch basins and storm drains provide ideal habitat for Cx. pipiens. The species becomes particularly abundant in areas where raw sewage leaks into subterranean drainage systems. Meat packing plants and slaughter house drainage ponds support hugh populations of this species. Culex pipiens can always be collected in the effluent from sewage treatment plants. Females feed on blood of birds or humans, and males feed on pollen, nectar, and the juice of plants. For an attempt to control such vectors, pesticides have been widely used and extensively produced. Accordingly the large scale use of toxicants against Cx. pipiens has frequently led to the development of strains of insects resistant to many insecticides which were designed for their eradication. Trials to study the effects of organphosphorus and synthetic pyrethroid insecticides in Cx. pipiens, the primary vector in Suadi Arabia, which breads in polluted water such as blocked drains and cesspits. It can bread in almost any kind of water collection. Chemical insecticides from different groups such as organophosphates, carbamates, pyrethroids and

insect growth regulators (IGR) which have good impact on Cx. pipiens.

**Objectives:** 

- To assess of some insecticides against Culex pipiens, in Abha city.

- To determine the dominant mosquito species in Abha city.

#### II. MATERIALS AND METHODS Rearing of Culex pipien

*Cx. pipiens* colony established from Laboratory (generation > F21) and maintained under a 14:10 (light: dark) cycle were used. Mosquitoes were reared using standard conditions (Richards et al. 2009) to generate similar-sized individuals. Adult mosquitoes were housed in 0.5 liter cardboard cages (Instawares, Kennesaw, GA) with mesh screening on top, provided a 20% sugar solution. The laboratory strain of Cx. pipiens used in this study was maintained under controlled conditions of temperature  $(27\pm2^{\circ}C)$ , relative humidity (70-80%) and a 16 hours photoperiod. Larval instars were maintained in enamel pans (30-35 cm in diameter and 8-10 cm in depth), half-filled with dechlorinated tap water, and provided with fish food. The amount of food was proportioned to the age of larvae. To avoid scum of formation in the rearing pans, larvae were poured daily into clean enamel pans. Developed pupae were transferred daily to plastic cups containing tap water that were introduced into 30 cm3 cages. Emerging adults were provided daily with 10% glucose solution, a piece of sponge hanging at the top of the cage by a thread. Females of Cx. pipiens were starved of sugar 24h prior to feeding on pigeon, each cage 27-95 mosquitoes/cage. Live pigeons are commonly used to maintain Cx. pipiens in colony then replace the pigeons every three months, this is artificial feeding methods are often used in our laboratory. Experiments may require using mosquitoes that have completed more than one gonotrophic cycle and the reproductive effects of these feeding regimens are currently unknown. Four days after the first blood feeding (mosquitoes 9 d old), 50 ml of tap water was added to each oviposition cup and mosquitoes were allowed to oviposit overnight. We and others have observed this time period to be sufficient for Cx. pipiens to complete the gonotrophic cycle prior to oviposition (Begum et al., 1985). Five days post-blood feeding, all adult mosquitoes were aspirated from cages and transferred according to treatment group to new cages containing an empty 100 ml plastic cup affixed to the bottom. As previously described, the plastic cup would be used later to contain an oviposition substrate . Larval sampling:

To find out the nature of the reproduction of species of medical mosquitoes important and geographical distribution of the hotbeds of reproduction in Abha city, the focus was on sites watersheds of swamps and pools, as well as the focus was on buildings under construction, animal shelters, tanks and water containers exposed in gardens, farms and brick factories. Samples were taken using standard dipping techniques with a plastic dipper (BioQuip Products, Inc. California, U.S.A.). Use hand dipper with 13 cm in diameter, made of reinforced plastic and equipped with wooden arm length of 160 is used to collect mosquito larvae from water in positive sites. Plastic jars 300 ml to put samples inside it and return it to the laboratory. The instars were preserved in 75% ethanol and identified morphologically to be defined (Edwards, 1941 & Gillies and Coetzee, 1987). GPS device to determine the coordinates of breeding spots and drop it on the maps to show areas with a high density of larvae in Abha city.

#### Adult mosquito sampling method:

Black Hole Mosquito Trap "Gangnam-gu, Seoul, 135-744, KORE " (Aburas, 2007) is basically used for collecting adult mosquitoes (can decoy and capture mosquitoes both in dark out doors and indoors in by a tripartite system). Electrically operated has been running the Black Hole Trap for 12 hours at least, where the bulbs fluorescent drop rays near ultraviolet (wavelength 352 nm) on a piece of painted Tio2, this process produces CO2 gas that attracts mosquitoes, also working the fan on the suction mosquitoes to net, which brings together the next day of fixing. Heat and near ultraviolet rays produced from the fluorescent lamps installed inside, and carbon dioxide produced when near ultraviolet rays are radiated onto titanium dioxide.Traps were placed in their positions before the sun and pool the next morning once every week, and is positioned samples were collected from traps in plastic cans (4x11x11 cm) blogger all the data from today's date and the name of the site. Then taken to the laboratory where they are placed in the freezer for an hour to kill insects live, then remove them from the freezer and left at room temperature for one hour in order to relax the muscles, is then loaded on scraps of paper and processed for the screening process and classification to be tested and to identify the species of mosquitoes and the results recorded.

#### **Test insects**

The laboratory strain of the mosquito, *Culex pipiens* (Linnaeus) was used as a baseline in insecticides. *Cx. pipiens* strains were collected from their drainages. These field colony strains exposed regularly to insecticidal applications for the pests control according to the routine

schedule program set annually. The 3rd instar larvae of *Cx. pipiens* were used to study the resistance ratio between the field and laboratory strains.

# Field colony strains

Strains of *Cx. pipiens* were collected from their drainages in areas treated with the recommended insecticides for mosquito control.

# Insecticides used

Commercial formulations of insecticides were used in this study representing three groups of insecticides commonly applied on mosquito control. These insecticides include organophosphorous (Propetamphos), synthetic pyrethroide (Cypermethrin) and *Bacillus thuringiensis* var. *israelensis*.

# a) Organophosphates

Common name: Propetamphos.

Trade name: Safrotin 20% MC.

Chemical name: (E) 0 - 2- isopropoxy – carbony l- lmethyl vinyl 0- methyl ethylphosphoramido thioatc.

# b) Synthetic pyrethroids

Common name: Cypermethrin 10% EC

Trade name: Exit 100

Chemical name: (RS)-alpha-cyano-3-phenoxybenzyl-(1RS,3RS,1RS,3SR)-3-(2,2-dichlorovinyl)-2,2-

dimethylcyclopropane carboxylate

#### c) Bioinsecticide

Common name: Bacillus thuringiensis var. israelensis.

Trade name: Baciloid 5000.

**Bioassays:** 

#### Larval bioassays

# **Dipping technique:**

The dipping method was applied according to the method described by Sukontason *et al.* (2004) with some modifications. All tests were run at 14:10 (light: dark) and maintained at  $28\pm2^{\circ}$ C and 70 – 80% humidity. The third instar larvae were used in the assays. Four replicates of twenty-five larvae each group were used at each concentration level. The experiments were repeated on subsequent days. The larvae of each group were gently dipped into insecticide solutions with a dip net, whereas those of the controls were dipped in tap water. After being dipped for exactly 30 sec, the larvae were transferred to the rearing jars containing food. After the larvae had been dipped, they were reared to determine the effect of different compounds of the life cycle also success of emergence on the other hand, the number of emerging flies was recorded.

#### Adult bioassay

CDC (complement mediated cytotoxicity) bottle bioassay. For the adult stage of Cx. *pipiens*, assays were adapted according to the WHO technique on the evaluation and testing of insecticides (WHO, 1996) Glass tubes were

treated singly with different concentrations of the chosen formulation of compounds. Solvent (acetone ethyl alcohol) were treated in test tubes (control). The treated test tubes were opened in a room kept at 25°C, 50-55% relative humidity and constant darkness and the door was kept closed due to no forced ventilation. Twenty five females were exposed to the treated surface for the one and 24 hours at the same temperature and relative humidity. Batches of 25 females were introduced into test tubes and allowed to a light and rest on the vertical treated surface. After the exposure period the mosquito were removed and transferred for observation and mortality count after one and 24 hours.

# Statistical analysis

#### Mortality counts:

Mortality counts were made after 24 hours. The dosage mortality data were subjected to probit analysis according to Finney (1952). Mortality percentages were corrected according to Abbott's (1925). Levels of resistance in the field colony strains of the two insects were calculated as follows:

		LC50 or LC90 of the field colony						
nesistailt	_	strains						
rauo( <b>k.k</b> )	_	LC50 of LC90 the laboratory strain						

The simple correlation and regression values were calculated to determine the relationship between the mean numbers of mosquitos captured and prevailing climatic conditions. The partial regression analyses were calculated to determine the effect of each weather factor alone on mosquito populations.

#### ANOVA

The data was subjected to analysis of variance (ANOVA) and the means were compared by LSD test at 0.05 levels, using SAS program (SAS Institute, 1988) Data were analyzed to find the relationship between mosquito densities and climatic factors and using correlation and multiple regression techniques. Similarly, the relationship between the climatic factors (temperature, relative humidity and rainfall) and mosquito density were analyzed.



III. RESULT AND DISCUSSION



Fig.1: %Mortality of the 3rd larval instars of laboratory and field strains of Culex pipiens after treated with different concentrations of Propetamphos 20% using dipping technique.

*Table.1: Susceptibility of Culex pipiens larvae (lab and field strains) to chemical insecticide Propetamphos 20% by dipping technique following continuous exposure for 24 h.* 

	Culex	Effective concentration s (ppm)	Larval mortali ty (%) <sup>a</sup>	Statistical parameters <sup>b</sup>								
Insecticide	pipie			LC50 (pp m)	LC90 (pp m)	Slop e	X <sup>2</sup> (Chi) <sup>2</sup>			R		
	<i>ns</i> strai n						С	Т	Р	С	Т	<b>RR</b> *
Propetamph	Lab St.	0.5-0.001	97-19	0.10 62	0.810 9	0.753 7	15.7 4	9. 5	0.000 4	0.92 7	0.81 1	27
os 20%	Field St.	0.5-0.001	90-15	0.04 42	3.31	0.683 8	12.1 5	9. 5	0.001 1	0.93 3	0.87 8	2.1

\* Resistant ratio (R.R.)

=

 $LC_{50}$  or  $LC_{90}$  for the field colony

Corresponding LC<sub>50</sub> or LC<sub>90</sub> of laboratory strain

a: Five replicates, 20 larvae each; control mortalities ranged from 0.0%-3.0%. b: Litchfield and Wilcoxon (1949). When tabulated (Chi)2 larger than calculated at 0.05 level of significance indicates the homogeneity of results C = Calculated T = Tabulated RR = Resistant ratio



Fig.2: Regression lines for Propetamphos 20% bioassay of larvae of laboratory and field strains of Culex pipiens using dipping technique.



Fig.3: Toxicity values of Propetamphos 20% against larvae of laboratory and field strains of Culex pipiens using dipping technique.





Fig.4: %Mortality of the adult females of laboratory and field strains of Culex pipiens after treated with different concentrations of Cypermethrin 10% using CDC bottle bioassay technique.



Fig.5: Regression lines for Cypermethrin 10% bioassay of adults females of laboratory and field strains of Culex pipiens using CDC bottle bioassay technique.

 Table.2: Susceptibility adult females of Culex pipiens (lab and field strains) to chemical insecticide Cypermethrin 100 following continuous exposure for 24 h using CDC bottle bioassay technique.

	Culex pipien s strain		Effectiv l e conc. morta (ppm) lity (%) <sup>a</sup>	Statistical parameters <sup>b</sup>								
Insecticide		Effectiv			LC90 (ppm)	Slope	X <sup>2</sup> (Chi) <sup>2</sup>			R		
		e conc. (ppm)		LC50 (ppm)			С	Т	Р	С	Т	RR
Cypermethri	Lab St.	0.5- 0.001	98-20	0.0132	0.6303	0.7631	20. 69	9. 5	0.0004	0.9 1	0.81	9.0
n 10%	Field St.	0.5- 0.001	89-19	0.1192	1.8522	1.076	19. 70	9. 5	0.0006	0.9 4	0.81	3

a: Five replicates, 20 larvae each; control mortalities ranged from 0.0%-3.0%. b: Litchfield and Wilcoxon (1949). When tabulated (Chi)<sup>2</sup> larger than calculated at 0.05 level of significance indicates the homogeneity of results C = Calculated T= Tabulated RR = Resistant ratio



Fig.6: Toxicity values of Cypermethrin 10% against adult females of laboratory and field strains of Culex pipiens using CDC bottle bioassay technique.

Conc.	Max. days of	%Mortality in No. of weeks							
(ppm)	residual activity	1 <sup>st</sup> week	2 <sup>nd</sup> week	3 <sup>rd</sup> week					
Laborato	ry strain	•							
0.001	4	11.7	0.0	0.0					
0.01	8	19.4	0.0	0.0					
0.1	10	61.9	7.4	0.0					
1	14	75.8	27.9	0.0					
10	17	93.7	51.5	2.7					
100	20	96.8	70.8	12.7					
Field strai	in	·		·					
0.001	6	9.2	0.0	0.0					
0.01	9	15.5	0.0	0.0					
0.1	11	58.4	0.0	0.0					
1	16	71.4	21.9	0.0					
10	19	90.8	42.7	0.0					
100	23	94.1	67.5	10.1					

Table.3: Residual activity (maximum no. of days) of Bacillus thuringiensis var. israelensis and percent mortality of Culex pipiens larvae/week



Fig.7: Residual activity (maximum no. of days) of Bacillus thuringiensis var. israelensis and percent mortality of Culex pipiens larva.

# Efficiency of organophosphrous insecticides against larvae of *Culex pipiens* by dipping technique under laboratory conditions:

In this study one organophosphorous insecticide used 'Propetamphos'. The tested toxicant was evaluated against field and laboratory strains of the 3rd instar larvae using the

dipping technique. The evaluation of insecticidal action was compared as follows:

# Comparison on basis of LC50 and LC90 values:

The used concentrations under laboratory conditions of Propetamphos was ranged between 0.5-0.001 ppm against laboratory and field strains. The mortality% of Propetamphos ranged between 19-97% against laboratory strain, whereas, against field strain the mortality% ranged between 15-90% {Table 1 and Fig. 1}. The required values, i.e. LC50's and LC90's are presented in {Tables 1 and Fig. 3]. Data given summarized the susceptibility of both field and laboratory strains of the 3rd instar larvae CX. Pipelines to Propetamphos. The results clearly showed that Propetamphos was effective insecticide (LC<sub>50</sub> 0.0162 ppm) against the 3rd instar of the laboratory strain comparing with a field strain which was 0.0442 ppm. At LC<sub>90</sub> level, data indicated that Propetamphos (LC<sub>90</sub> 0.8109 ppm) was effective insecticide against the 3rd instar larvae of laboratory strain, while against field strain gave (LC<sub>90</sub> 3.31 ppm). The slope of line is useful to know the homogeneity of stages of M. Domestic population, which reared under laboratory conditions. When the population of mosquitoes is similar in the homogeneity or the degree of resistance, meaning the slope is big or increase in regression, also, when tabulated (Chi)<sup>2</sup> larger than calculated at 0.05 level of significance indicates the homogeneity of results. Data in {Table 1 and Fig. 2} show that the slope of laboratory and field strains of larval stages of CX. pipiens population when using Propetamphos. Slope of laboratory strain was 0.7537 in case Propetamphos, while in filed strain was 0.6838. Results in  $\{Table - 1\}$  indicated that the tabulated X<sup>2</sup> (Chi) <sup>2</sup> was 9.5, while calculated X<sup>2</sup> (Chi) <sup>2</sup> was 15.74 of laboratory strains of larval stages of CX. pipiens when using Propetamphos, on the other hand, the tabulated (Chi) <sup>2</sup> was 9.5 and calculated (Chi) <sup>2</sup> was 12.15 against field strains, showed that the results are homogeneous. LC values that differ by 5-fold or less are not likely to reliably indicate resistance, and as a general guideline, differences of 10-fold or greater are necessary for proof of resistance. To determine the levels of resistance in field colonies of CX. Pipelines to the insecticidal action of Propetamphos, that mentioned previously at LC<sub>50</sub> level was 0.0162 of laboratory strains and was 0.0442 of field strains, respectively, while at LC<sub>90</sub> levels was 0.8109 of Propetamphos against laboratory strain and was 3.31 p.m. against field strain, with average of resistance ratio 2.73 fold, results indicated that the field strain of Propetamphos is sensitive. The obtained results agree with those obtained by Abed El-Samie and Abed El-Baset (2012) tested the efficacy of the most used insecticides belonging to different groups (organophosphate, carbamate, synthetic and pyrethroid) against four different field populations of CX. pipiens. Results obtained showed that the laboratory colony showed higher susceptibility to the tested insecticides than the mosquito populations collected from Sharkia and Assiut Governorates. Field populations of CX. pipiens from Sharkia were chosen to study the development of resistance

(resistant strain) in *CX. Pipelines* to chlorpyrifos toxicity. After 15 generations of selection pressure using chlorpyrifos against the 3rd instar larvae of *CX. pipiens*, resistance increased by 24.56-fold in the resistant strain as compared with the control. Fractionation of total soluble proteins using SDS–PAGE revealed some differences in the laboratory colony, field populations and resistant strain. The results may indicate that alkaline phosphatase and nonspecific esterases were probably responsible for the detoxification of chlorpyrifos in field populations.

# Efficiency of pyrethroid insecticides against adults of *CX. Pipelines* under laboratory conditions using the CDC bottle bioassay technique:

Pyrethroid compounds are prevented sodium gates from closing in nerves of insects and potent neuropoisons, their mechanism of action on the nervous system. The nerve excitation occurs as a result of changes in nerve membrane permeabilities to sodium and potassium ions, and therefore any effect of pyrethroids can be interpreted in terms of such permeabilities. In this study one pyrethroid insecticide used 'Cypermethrin'. The tested toxicant was evaluated against field and laboratory strains of the adult females using a CDC bottle bioassay.

# Comparison on basis of LC<sub>50</sub> and LC<sub>90</sub> values:

The used concentrations under laboratory conditions of Cypermethrin was ranged between 0.5-0.001 ppm against both laboratory and field strains. The mortality% of Cypermethrin ranged between 20-98% against the adult females of laboratory strain, while against field strain the mortality% of Cypermethrin ranged between 19-89% *[Table - 2 and Fig - 4]* The required values, i.e. LC50's and LC90's are presented in *[Tables - 2 and Fig - 6]*. Data given summarized the susceptibility of both field and laboratory strains of the 3rd instar larvae of CX. pipiens to the tested chemical. The results clearly showed that Cypermethrin was effective insecticide (LC50 0.0 ppm) against the 3rd instar larvae against laboratory strain, while against field strain Cypermethrin gave (LC50 0.1192 ppm). **The slope of toxicity lines:** 

Data in  $\{Table - 2\}$ : Show that the slope of laboratory and field strains of larval stages of CX. pipiens population when using Cypermethrin. Slope of laboratory strain was 0.7631 respectively, while in filing strain was 1.076. X<sup>2</sup> (Chi) <sup>2</sup> value:

Results in  $\{Table - 2\}$ : indicated that the tabulated X<sup>2</sup> (Chi) <sup>2</sup> was 9.5 while calculated X<sup>2</sup> (Chi) <sup>2</sup> of laboratory and field strains of larval stages of *CX. pipiens* population when using Cypermethrin was 20.69 and 19.70, respectively . Pyrethroids are now offered in a variety of commercial formulations available to ordinary consumers for use in the home. It is now estimated that approximately 80-90% percent of households in the United States use pesticides with pyrethroids comprising a considerable percentage of total use. To determine the levels of resistance in CX. pipiens of field colony to insecticidal effect, the adult specimens were collected from Abha city, in objective to compare the LC50 or LC90 values of the field colony with the corresponding values of the laboratory strain. The results given in  $\{Table - 2\}$ : illustrate monitoring resistance ratio in the 3rd instar larvae of CX. pipiens field colony strains collected from Abha city. Concerning the population of Abha city, the resistance ratios in the 3rd instar larvae to the toxicity of cypermethrin that mentioned previously at LC50 levels were 0.0132 and 0.1192 against laboratory and field strains, respectively .Our results some agree with obtained from Al-Sarar (2010) studied developing resistance against commonly used insecticides to control mosquitoes in Riyadh. Two populations from Wadi Namar (WN1 and WN2) were highly resistant to deltamethrin (187.1- and 161.4-folds respectively). The field population from AL-Wadi district (AL-W) showed low resistance to lambda-cyhalothrin (3.8-folds) and moderate resistance to beta-cyfluthrin and bifenthrin (14and 38.4-folds respectively). No resistance to fenitrothion was observed in WN1 population. Fenitrothion concentrations required to inhibit 50% of Acetylcholinesterase (AChE) activity in both WN1 population and the laboratory susceptible strain (S-LAB) were 786 and 0898ppm respectively. Piperonyl butoxide suppressed resistance to pyrethroid insecticides (> 90%) in field populations, indicating that oxidizes and/or esterases play an important role in the reduction of pyrethroids toxicity. These results should be considered in the current mosquito control programs in Riyadh. Four years earlier, resistance ratios of 10. And 8.5-folds of CX. pipiens populations from Iraq and EL-Nafl localities in Riyadh city, were recorded to deltamethrin (Al-Sarar et al., 2005). The following results In agreement with the present results, high resistance levels of Cs. pipiens pipiens (233 and 453 fold) to deltamethrin were reported in Tunisia (Daaboub et al., 2008). The field population AL-W displayed a low level of resistance to lambda-cyhalothrin and moderate levels of beta-cyfluthrin and bifenthrin. The low resistance level was also reported in CX. quingefasciatus field strains from Brazil and Malaysia against lambda-cyhalothrin. The AL-W population was susceptible to fenitrothion. This might be contributing to that the municipality of Riyadh does not use this compound regularly. The I50 values of AChE, the target site of fenitrothion, revealed that susceptibilities of S-LAB and WN1 populations were very

close. In Sri Lanka, AChE was also found to be sensitive to organophosphate insecticides in Kurunegala and Trincomalee An. culicifacies field populations.

# Efficiency of *Bacillus thuringiensis var. israelensis* against larvae of *Culex pipiens:* by dipping technique under laboratory conditions:

Bti have shown the highest level of biological activity with different levels of toxicity to mosquito species, i.e.; Culex and Aedes are highly susceptible while Anopheles is less susceptible (Charles et al., 1996). These biological insecticides have had a decimating effect on the use of chemical pesticides against mosquitoes during past 2-3 decades. Therefore, the aim of this point is efficiency of Bacillus thuringiensis var. israelensis against larvae of CX. Pipelines by dipping technique under laboratory conditions. Data in {*Table - 3 and fig - 7*} indicated that the residual activity of Bacillus thuringiensis var. israelensis reached from 4 to 20 days of concentrations ranged between 0.001 to 100 p.m. in case laboratory strain, while reaching between 6-23 days of concentrations ranged between 0.001 to 100 p.m. in case field strain. Also found the mortality percent ranged between 11.7 to 96.8% of concentrations ranged between 0.001 to 100 p.m. against laboratory strain in the 1st week, whereas ranged between 0.0 to 70.8 in the 2nd week against the same concentrations finally ranged between 0.0 to 12.7 in the 3rd week. On the other hand, the mortality percent ranged between 9.2 to 94.1% of concentrations ranged between 0.001 to 100 ppm against field strain in the 1st week, whereas ranged between 0.0 to 67.5 in the 2nd week against the same concentrations and finally ranged between 0.0 to 10.1 in the 3rd week. Obtained results agree with those obtained by Jahan et al. (2013)they found that field collected Culex quinquefasciatus larvae were more susceptible and have prolonged residual effect as compared to laboratory reared A. Stephens against Bsph while Bti have effect vice versa. Also, Zahran et al., (2013) found the most effective tools for CX. pipiens larvae eradication included B.t.i. Followed by Emamectin benzoate, Azadirachtin, Diflubenzuron then B. Business. The use of some binary mixtures of these tested control measures can get better control, save the amount and reduce control cost. Residual effect for field evaluation of Bti WDG under low treatment (0.2 mg/liter) against CX. quinquefasciatus lasted 14 days indicating more efficient for field bases as compared to laboratory. Biological control with Bti and Bsph larvicides proved highly effective yet selective in action (Charles and Nielsen, 2000) and therefore, environmentally safe to nontarget organisms as well as for human exposure (WHO, 1997). These Bacillus products are cost effective, can be produced locally and highly acceptable in the community.

# IV. CONCLUSION

- Some of the mosquitoes encountered are medically important as they are known vectors of various diseases, including malaria and dengue fever among others.
- Activities of immature stages and adults in outdoor areas should be started in late April-early May.
- Indoor control activities are very important during November and early May, because mosquito adults in the area do not leave their indoor areas and they do not lay eggs in November.
- In order to maintain historical records of relative vector population densities and seasonal population trends.
- Consequently, this study has provided information on temporal distribution and abundance. Since most of the species encountered are potential vectors of mosquito-borne diseases.

# REFERENCES

- Abd El-Samie & Abd El-Baset (2012). The efficacy of some insecticides on field populations of *Culex pipiens* (Linnaeus) from Egypt. The Journal of Basic & Applied Zoology. 65(1): 62-73.
- [2] Ali S Al-Sarar (2010). Insecticide resistance of *Culex pipiens* (L.) populations (Diptera: Culicidae) from Riyadh city, Saudi Arabia: Status and overcome. Saudi J Biol Sci. 6; 17(2):95-100.
- [3] Al-Sarar A.S., Hussein H.I., Al-Rajhi D., and Al-Mohaimeed, S. (2005), Susceptibility of CX. *pipiens* from different locations in Riyadh city to insecticides used to control mosquitoes in Saudi Arabia. J. Pest Cont.
- [4] Assar, A.A, Abo-El-Mahasen, M.M., Harba, N.M and Rady, A. A. (2012). Biochemical effects of chromosome on Culex pipiens larvae (Diptera: Culicidae). J. Am. SCI., 8 (5): 443-450.
- [5] Bekhiet, S.S. And Zayed, A.B. (1990). Laboratory evaluation of permethrin impregnated mosquito net against C. Pipiens. The Twelfth Scientific Conference, Cairo, Egypt.
- [6] Bisset, J., Rodriguez, M., Soca, A., Pasteur, N. And Raymond, M. (1997). Cross-resistance to parathyroid and organophosphorus insecticides in the southern house mosquito (Diptera: Culicide) from Cuba. J. MED. Entomol., 34 (2): 244-246.
- [7] Brookfield, J. F. Y. (1991). The resistance movement. Nature London, 350: 107-108.

- [8] Cetin and Yanikoglu, A. (2005). Evaluation of the naturally-derived insecticide spinosad against Culex pipiens L. (Diptera: Culicidae) larvae in septic tank water in Antalya, Turkey. J. Vector Ecol., 30 (1): 151-4.
- [9] Cetin, H. And Yanikoglu, A. (2006). Evaluation of tempos and chlorpyrifos-methyl against Culex pipiens (Diptera: Culicidae) larvae in septic tanks in Antalya, Turkey. J. MED. Entomol., 43 (6): 1195-9.
- [10] Chandre, F., Darriet, F., Darder. M., Cuany, A.; Doannio, J.M., Pasteur, N. And Guillet, P. (1998). Pyrethroid resistance in Culex Quinquefasciatus from West Africa. MED. Vet. Entomol., 12 (4): 359-366.
- [11] Charles, J. F. And C. Nielsen-LeRoux (2000) Mosquitocidal bacterial toxins: diversity, mode of action and resistance phenomena. Memories Do Institute Oswaldo Cruz, 95: 201–206.
- [12] Charles, J., Nielsen-LeRoux, F., and Delecluse, A. (1996). Bacillus sphaericus toxins. Molecular biology and mode of action. Annu. Rev. Entomol., 41: 451-472.
- [13] Corbel, V., Guessan, R. N., Brengues, C.; Chandre, F., Djogbenou, L., Martin, T., Akogbeto, T., Hougard, J. M. And, Rowland, M. (2007). Multiple insecticide resistance mechanisms in Anopheles gambiae and Culex quinquefasciatus from Benin, West Africa. Acta Tropica, 101 (3): 207-216.
- [14] Daaboub J., Ben Cheik R., Lamari A., Ben Jha I., Feriani M., Boubaker C., Ben Cheik H. 2008. Resistance to pyrethroids insecticides in Culex pipiens pipiens (Diptera: Culicidae) from Tunisia. Acta Trop. 107: 30–36.
- [15] Ganguly, S.S., Dutta-Gupta, K.K. And Dutta, P.K. (1994). Evaluation of susceptibility status of Culex quinquefasciatus larvae of organphosphorus insecticides based on logistic regression analysis Indian J. Public Health, 38 (1): 8-13.
- [16] Jahan, N., Jamali, E. H. And M. F. Qamar (2013). The residual activity of *Bacillus thuringiensis* var. *Israelensis* and *Bacillus sphaericus* against mosquito larvae. J. Anime. And Plant Sci., 23 (4): 1052-1059.
- [17] Li, A. Y., Guerrero, F. D. And Pruett, J. H. (2007). Involvement of esterases in designing resistance and biphasic effects of piperonyl butoxide on diazinon toxicity to Haematobia irritans (Diptera: Muscidae). J. Pesticide Biochem. And Physiol., 87: 147-155.
- [18] Mikhail, M. W., Al-Bursheed, K. M. And Allam, K.A. M. (2007). Susceptibility of Culex pipiens

complex to some insecticides in Qatar. J. Egypt. Soc. Parasite., 37 (3): 893-902.

- [19] Mostafa, A. (1990). Toxicological studies on the larvae of Culex pipiens (L.). PH. D. Thesis, Fac. Agric. Ain-Shams Univ.
- [20] Raghavendra, K., Singh, S.P., Subbarao, S. K. And Dash, A.P. (2009). Laboratory studies on mosquito larvicidal efficacy of aqueous &hexane extracts of dried fruit of *Solanum nigrum* Linn. Indian J Med Res 130, July 2009, pp 74-77.
- [21] Rodriguez, M.M., Bisset, J., Rodriguez, I. And Diaz, C. (1997). Determination of insecticide resistance and its biochemical mechanisms in 2 strains of *Culex quinquefasciatus* from Santiago de Cuba. Rev. Cabana MED. Trop., 49 (3): 209-214.
- [22] Selvi, S., Edah, M.A., Nazni, W.A., Lee, H.L. And Azahari, A.H. (2007). Characterization on malathion and permethrin resistance by bosses and the variation of esterase activity with the life stages of the mosquito *Culex quinquefasciatus*. Tropical Biomedicine, 24 (1): 63–75.
- [23] Sukontason, K.L., Boonchu, N. Sukontason, K. And Choochote, W. (2004). Effects of Eucalyptol on House Fly (Diptera: Muscidae) and Blow Fly (Diptera: Calliphoridae). MED. Troop. S. Paulo, 46: 97–100.
- [24] Tittiger, C. And Walker, V.K. (1997). Isolation and characterization of an unamplified ester B3 gene from malathion-resistant, *Culex tarsalis*. Biochem. Genet. 35(3-4): 119-138.
- [25] Wirth, M. C. And Geoghiu, G.P. (1996). Organophosphate resistance in *Culex trails* from Cyprus. J. Am. Mosque. Control Assoc., 12 (1): 112-18.
- [26] World Bank Report (2001). Malaria at a Glance.
- [27] Saran, Hossam, El. M., Kawanna, M. A. And Bosly, H. A. (2013). Larvicidal Activity and Joint Action Toxicity of Certain Combating Agents on *Culex pipiens* L. Mosquitoes. Annual Review & Research in Biology; 3 (4): 1055.
- [28] Zamburlini, R. And Bellantone, P. (1993).
  Resistance to the organophosphate insecticides tempos and Malathion in *Culex pipiens* L. (Diptera: Culicidae) from the Adriatic Coast near Friuli. Parasitologia, 35 (1-3): 1-15.