The Effect of Different NaCl and pH Levels on Survival of *Culex sp.* (Diptera) Larvae in Basrah

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Abstract: To investigate the effect of NaCl or pH levels on survival of *Culex sp.* larvae , a sample of these larvae was collected from some pools in Basrah city and exposed to 5,10,15 and 20 ppt of NaCl or 2,3 and 4 pH levels . Lethal concentration for 50% of exposed larvae (LC50) in NaCl and pH treatments were 6.2892 ppt and pH 2.98 respectively .All of the larvae were died at first few hours of exposure to either 20 ppt of NaCl or pH =2 . The results of this study suggest that NaCl levels higher than 6.289 ppt and pH levels higher than 2.98 (in *Culex sp.* larvae environment) are very effective on larvae survival, thus these levels could be used in mosquitoes control efforts.

Keywords: *Culex* , larvae , pH , NaCl , toxicity .

Introduction

Toxicity tests are desirable in water quality evaluations because chemical and physical tests alone are not sufficient to assess potential effects on aquatic biota . Two of toxicity tests purposes are determining (1) suitability of environmental conditions for aquatic life and (2) favorable and unfavorable environmental factors such as DO , pH ,temperature , salinity or turbidity (1) .

Mosquitoes are flies in the family Culicidae . There are more than 3000 known species exist worldwide (2) .Furthermore mosquitoes differ from other biting Diptera in having a long slender body , long legs and long needle-shaped mouth parts (3) .

Mosquitoes are poikilothermic (cold-blooded) , thus their rate of development and other aspects of their physiology are temperature-dependent ,as temperature increases , their development time shortens (2).

The real danger ,that ,mosquitoes vector diseases and capable of transmitting disease-causing viruses , protozoa and filarial nematodes (4). There are four distinct developmental stages in mosquitoes life cycle : egg, larva, pupa and adult, the immature stages require water to complete their life cycle (5).

Larvae emerge from eggs within 2-3 days in optimal conditions . They can be found in a wide variety of habitats, including temporary flood water and snowmelt pools. Larvae eat a variety of died and living organisms including detritus, algae, bacteria and fungi (2). T he larval stage is typically completed in 5-6 days (5).(6)Found that *Toxorhynchites splendens* larvae could live in turbid and distilled water .

The purpose of this study was to investigate the effects of fluctuations in pH and salinity on survival of mosquitoes' larvae, and to determine the lower and upper limits of larvae tolerance to these factores as a part of mosquitoes' breeding ecology.

Materials and Methods

Sample collection : larvae of mosquitoes *Culex sp*. were collected from some temporary rainwater pools in Aljubaila region, identified and isolated in biology department, college of education, university of Basrah.

Preparation of test solutions : NaCl test concentrations (5,10,15 and 20 ppt) prepared by weighing the required quantity (gm) for each concentration of pure NaCl and dissolved it in one liter of distilled water, with three replicates for each treatment. pH solutions (pH 2,3 and 4) were prepared by using nitric acid (HNO3), a pH meter was used to determine pH level over all the time of experimentation, with the same number of replicates as in NaCl treatments.

Procedure : Larvae were isolated and distributed into different salinity concentrations and pH levels(25 larvae in each replicate). Another 25 larvae were kept in the original sample water, the number of died larvae were recorded every 24 hours. Statistical treatment was conducted according to (7) and(8).

Results

Fig.1 and 2 show the toxicity lines of NaCl and pH levels to *Culex sp.* larvae, which obtained by plotting log. of NaCl or pH levels against percentage probit of mortality, the equations of linear regression were obtained by least squares method.

The lethal concentrations for 50% (LC50)of *Culex sp.* larvae used as base for comparison between the toxicity of NaCl or pH treatments. During the exposure period (96 hours) the LC50's of NaCl and pH were 6.2892 ppt and 2.98 respectively (table 1), the difference between upper and lower limits of confidence of NaCl concentrations was greater than this of pH level. All of the larvae were died at first few hours of exposure to either 20 ppt of NaCl or pH 2. Least squares method was used to conduct the relationship between time of exposure and mortality of *Culex sp*. larvae (Fig.3 and 4). Time-Mortality relationship was represented by plotting log. of experimentation time against percentage probit of mortality (Fig.3 and 4).

Table 2 shows the lethal time for 50% of *Culex sp.* larvae (LT50) in each concentration of NaCl and pH level, when NaCl or pH level was higher, the difference between upper and lower limits of confidence was higher too. The lowest LT50 was in the highest NaCl concentration and in the lower one of pH levels.

Discussion

environmental factors operate to create favorable or unfavorable conditions for mosquito breeding (9), since breeding habitats of different mosquito species are specific to particular habitats, the determination of breeding habitats is of great benefit to mosquito control method (10). Table 1 shows that pH LC50 was lower than that of NaCl, same result was found by (11) when *Aedes aegypti* and *Culex quinquefasciatus* larvae exposed to distilled water or high NaCl media, the hemolymph ion levels did not affect, but pH3 caused significant decrease in hemolymph Na^+ and Cl^- levels in both species.

The results of present study (table 1) refer that NaCl concentrations upper than 6.2 ppt and pH level less than 3 are lethal for more than 50% of exposed *Culex sp.* larvae in 96 hours, beside

	pH	NaCl ppt
a	8.7107	8.256
b	-7.8239	-1.5931
LC16	3.988	4.7718
LC50	2.98	6.2892
LC84	2.227	8.2893
S (Slop)	0.747	1.318
F (Slop coefficient)	0.886	1.12076
Upper limit of confidence	2.6422	167.455
Lower limit of confidence	3.361	111.232

Table 1

	pH=4	pH=3	15 ppt	10ppt	5 ppt
a*	1.328	2.2479	4.8546	2.1885	1.7332
b*	1.85	0.673	1.747−	1.5027	1.2995
LT16 (hour)	42.3161	30.5138	15.3421	13.9852	36.6328
LT50 (hour)	235.4984	84.1216	24.5366	39.631	136.479
LT84 (hour)	1310.5991	231.909	39.2415	112.305	508.47
S (Slop)	5.5652	2.7568	1.5993	2.8337	3.7256
F (Slop coefficient)	4.5024	1.8748	1.2682	1.7256	2.6494
Upper limit of confidence	1060.308	157.711	31.1173	68.387	361.587
Lower limit of confidence	52.3051	44.8696	19.3475	22.9665	51.513

* Constant in linear regression equation y =b+ax

Table 2

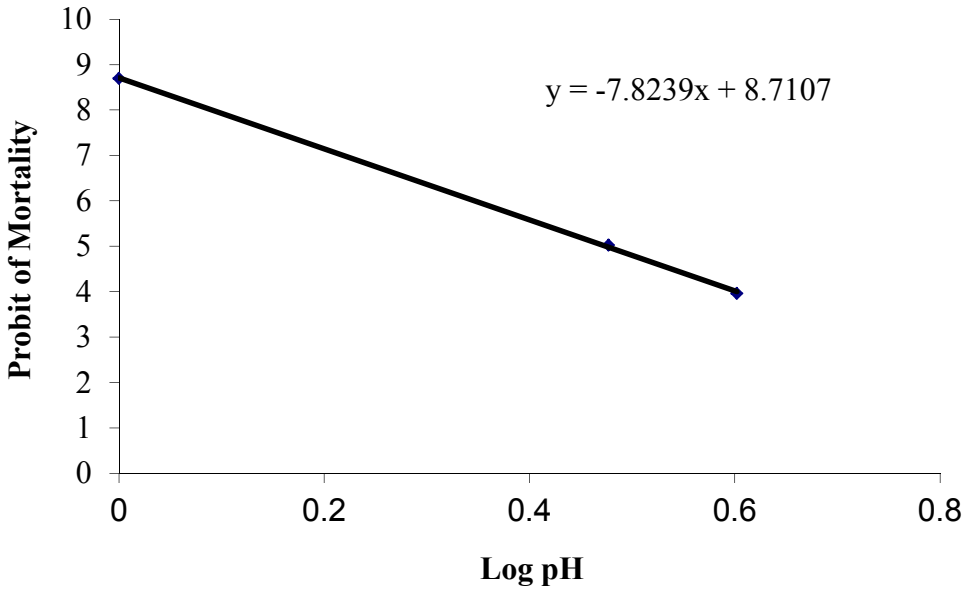
y = -7.8239x + 8.7107
Probit of Mortality
Log pH
10
9
8
7
6
5
4
3
2
1
0
0
0.2
0.4
0.6
0.8

Figure 1

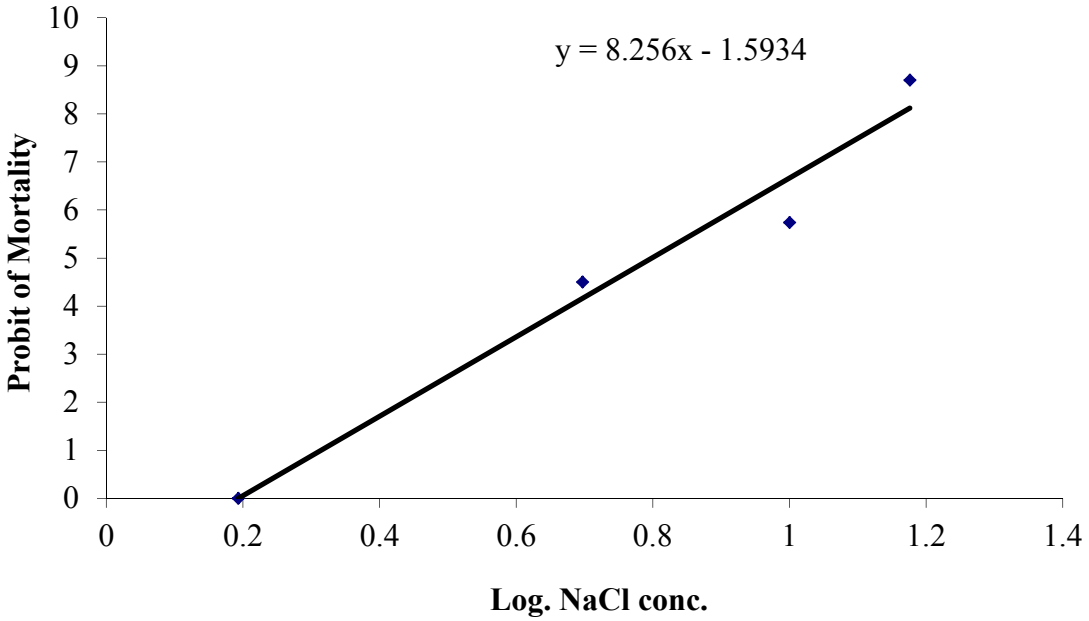
y = 8.256x - 1.5934
Probit of Mortality
10
9
8
7
6
5
4
3
2
1
0
0
0.2
0.4
0.6
0.8
1
1.2
1.4
Log. NaCl conc.

Figure 2

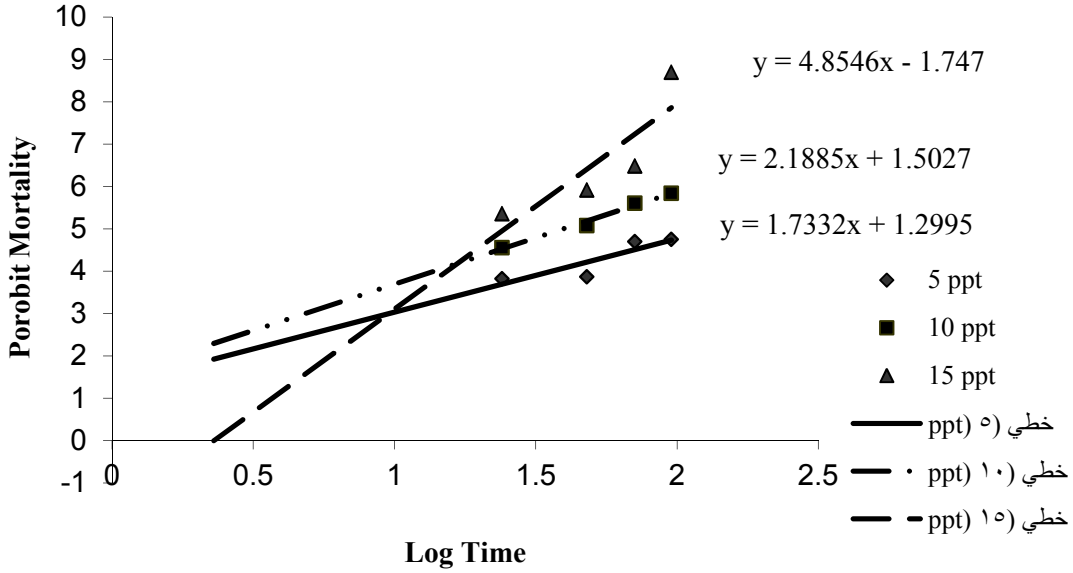
Porobit Mortality
Log Time
y = 4.8546x - 1.747
y = 2.1885x + 1.5027
y = 1.7332x + 1.2995
5 ppt
10 ppt
15 ppt
خطي (٥ ppt)
خطي (١٠ ppt)
خطي (١٥ ppt)

Figure 3

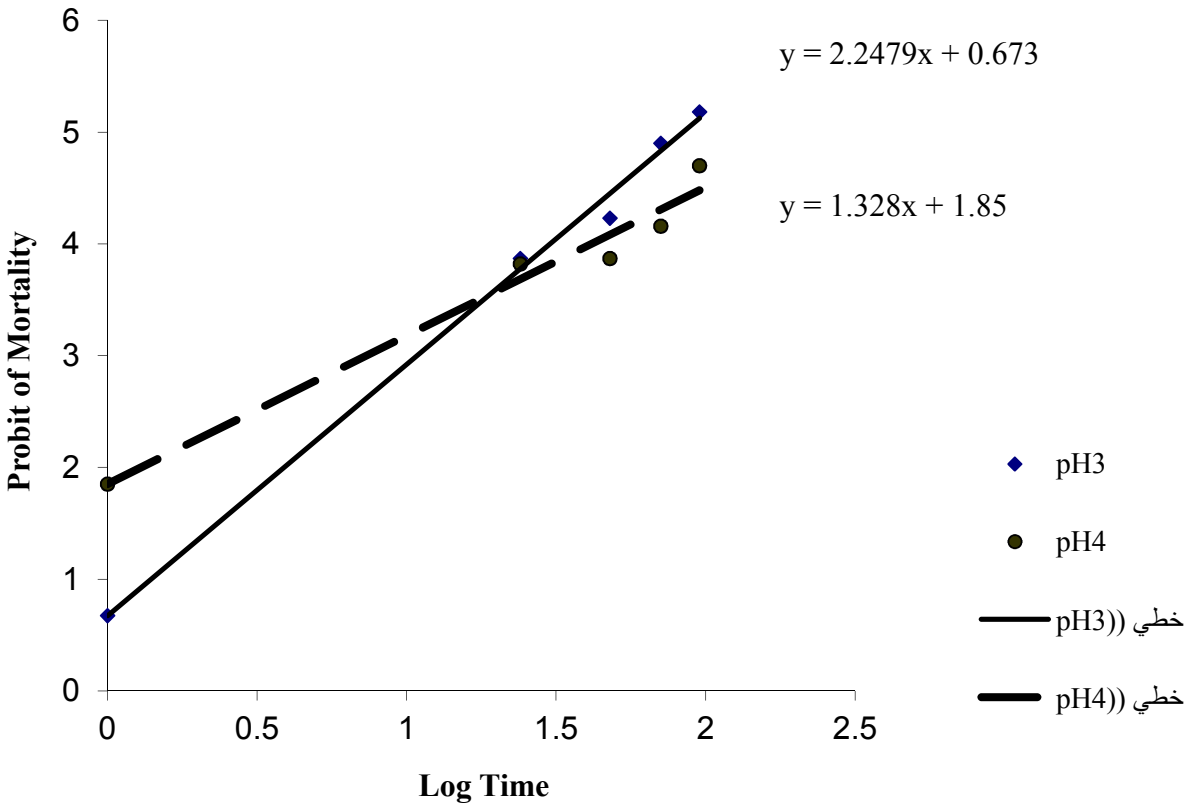
y = 2.2479x + 0.673
y = 1.328x + 1.85
Probit of Mortality
Log Time
0
1
2
3
4
5
6
0
0.5
1
1.5
2
2.5
pH3
pH4
pH3)) خطي
pH4)) خطي

Figure 4

Figure 1: Mortality of *Culex* larvae due to exposure to pH=3&4 during 96 hours with linear regression equation .

Figure 2 : Mortality of *Culex* larvae due to exposure to 5,10&15 ppt of NaCl during 96 hours with linear regression equation.

Figure 3 : Mortality of *Culex* larvae during 24,48,72 &96 hours of exposure to 5,10 &15 ppt of NaCl with linear regression equations.

Figure 4 : Mortality of *Culex* larvae during 24,48,72 &96 hours of exposure to pH=3&4 with linear regression equations.

Table 1: NaCl concentration & pH levels caused in mortality of 16,50&84% of *Culex* larvae with upper & lower limits of confidence.

Table 2 : Time in hours caused in mortality of 16,50&84% of *Culex* larvae with upper & lower limits of confidence for NaCl concentrations & pH levels.

death salinity may cause other effects on larvae as in (12) study who found that with increasing salinity, growth rates of *Aedes aegypti* decreased, percent body water was constant across salinity and duration of larval stage increased, but this increase cannot compensate for decrease in growth rate, resulting in an overall decrease in both wet and dry pupal mass at high salinity.

pH3 LC50 was 84.12 hours (table 2), is as a result to disturbance in uptake of ions (11) found that Rio Negro water with pH 3.5 (an organically rich but ion poor body of water) caused inhibition of Na+ uptake and stimulation of Cl- uptake in *Culex quinquefasciatus* and only a significant reduction of Na+ uptake in *Aedes aegypti* .

LT50 for highest used NaCl concentration (15ppt) was 24.53 hours (table 2) this tolerance of larvae may result from modulation of Na+ efflux and Cl- influx as mentioned by (13) as a result this modulation allowed *Culex tarsalis* to avoid apotential salt load and ionic disturbance in the hemolymph during an acute increase in salinity .

The slope of pH levels was higher than that of comparable NaCl concentrations (table 2), high slope value refers that the tested group is homogenous and susceptible to factors which effect on it during experimentation (7).

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تأثير مستويات مختلفة من NaCl أوpH في بقاء يرقات بعوض (Diptera) *Culex sp.* في البصرة

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الخلاصة : لدراسة تأثير مستويات ملح NaCl او دالة الحموضة pH في بقاء يرقات بعوض *Culex sp.*، تم جمع عينة من هذه اليرقات من بعض برك مدينة البصرة وعرضت الى تركيز 5،10،15او 20 ppt من NaCl او 2،3 او pH 4 ، وجد ان التركيز القاتل لنصف اليرقات (LC50) في معاملات NaCl و pH كان 6.2892 ppt و 2.98 pH على التوالي. ماتت اليرقات في الساعات القليلة الاولى من التعرض ل 20 ppt من NaCl او pH= 2. تقترح نتائج هذه الدراسة ان مستويات NaCl الأعلى من 6.2892 ppt ومستويات pH الأعلى من 2.98 مؤثرة جدا في بقاء يرقات *Culex sp.* ، وعليه فأنها يمكن ان تكون مفيدة في جهود مكافحة البعوض.