

Larvivoracious Potential of Different Stages of *Culex tigripes* (Diptera, Culicidae) in the Prospective of its Use in Biological Control of Malaria Vectors.

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Key words : Larvivoracious potential - *Culex tigripes* - Biological control - Malaria.

Summary

Carried out in the laboratory, the present study tends to assess the larvivoracious potential of different stages of *Culex tigripes* against malaria vectors. The four stages of *C. tigripes* have effectively a larvivoracious activity and may be used in the biological control of mosquito larvae, vector of malaria. Neither lower or higher larval density, large or small size of larvae nor any position pattern of prey larvae from the water surface do inhibit the larvivoracious behaviour of *C. tigripes*.

Résumé:

La présente étude en laboratoire évalue le potentiel larvivoire des différents stades de *Culex tigripes* contre les vecteurs du paludisme. Les quatre stades larvaires de *C. tigripes* ont effectivement une activité larvivoire et peuvent être utilisés dans la lutte biologique contre les larves de moustiques transmetteurs du paludisme. Ni la densité, ni la taille, ni même la position des larves proies sur la surface de l'eau n'inhibent le comportement larvivoire de *C. tigripes*.

Introduction

Malaria is one of the most mortal complaints in tropical areas. It has presently attained upsetting proportions with 270 million of infected persons among which more than one million die yearly (7).

The chemoresistance phenomena have rendered inefficacy the malaria control program : the main mortal malaria parasite *Plasmodium falciparum* has grown into resistant to the most widespread antimalarial drug, in this case, the Chloroquine and its by-product (2, 6).

On the other hand, mosquito vectors resist more and more to insecticides. In addition, these synthetic drugs are very expensive and pollute the environment. Weiser (10) advocates the biological mean as the reasonable and environment friendly alternative for control of diseases vectors.

The increase of malaria infection, necessitates a steady search for the discovery of new organisms which may destroy the vectors of this mortal endemic disease without disturbance in the environment.

Larvae of some Culicidae genera are known to be predators of Anophelines larvae and may be used in biological control of malaria vectors .

Among the mosquito species which larvae feed on other mosquito aquatic stages only *C. tigripes* usually extends its regime to Anopheles larvae. In addition, *C. tigripes* ranges in the same larval breeding sites as Anopheles species (fish ponds, temporary rainwater pools, shallow of streams and rivers, etc.).

Adult female of *C. tigripes* feeds on birds and very rarely on man (8 , 9).

Considering this triple advantage, *C. tigripes* is the single species of mosquito with predaceous larvae

which may be used in biological or integrated control of malaria vectors without any danger to human being.

The predaceous efficiency of *C. tigripes* larvae against larvae of mosquitoes have already been pointed out (1, 3, 4, 5).

This paper aims to contribute to the understanding of the larvivoracious potential of *C. tigripes*, mainly to study the undermentioned aspects, not yet elucidated, such as:

1. the larval stages on which *C. tigripes* is more effective in larvae eating ;
2. the influence of the larval density on the larvivoracious behaviour of *C. tigripes*;
3. the impact of prey larvae position in the water on the larvivoracious activity of *C. tigripes* : the predation of prey larvae of *Anopheles* genus (which adopt horizontal position on the water surface) is compared with the predation of prey larvae of *Culex* (whose position is almost vertical, hanging down from the water surface by the tip of their long respirator siphon).

Material and Methods

The biological material is constituted by larvae of *C. tigripes* used as predator larvae and by prey larvae belonging to Anopheles and *Culex* (other than *C. tigripes*) species. The three groups are distinguished from each other by the characteristics presented below :

Anopheles larvae do not have respirator siphon so that, in breathing, take up a horizontal position on the surface of water.

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Larvae of *C. tigripes*, larger than the former, have a short siphon ; in breathing, they stay almost parallel on water surface in the attitude recalling those of *Anopheles* larvae.

The other larvae of *Culex* genus are of varied size according to the species and have a long siphon. They hang down from the water surface by the tip of their long siphon through which they breath.

To estimate the larvivorous potential of *C. tigripes*, we exposed one single predaceous larvae with fifty prey larvae composed of twenty - five individuals of *Culex* spp. and twenty - five of *Anopheles* spp.

Due to the cannibalism observed in the feeding behaviour of *C. tigripes* only one larva of this larvivorous species was exposed at once with prey larvae in sight to have standard experiments. The larval density varied with the two kinds of container surface (37.50 cm² and 1130 cm²) used for expositions. So, two larval densities were considered:

Density 1 = 1.36 larvae/cm² and Density 2 = 0.045 larvae/cm² (in this regards the larval density varied from simple to third).

Exposed larvae were sorted according to their developing aquatic stages. Previous observations on the manner mosquito larvae cast their skins before growing into larger stages allowed us to conciliate size of larvae and its developing stage according to the species. A micrometer ocular mounted on a 10 x objective of a microscope was used for the mensuration of the 4 larval stages.

Especially for prey larvae constituted by several species, the mensuration was done according to the 4 larval stages of *Anopheles gambiae* and *Anopheles funestus* (the main vectors of malaria in Africa) identified in the laboratory.

Thirty-two combinations of predation experiments with defined larval stages (1 to 4) for prey and *C. tigripes* larvae were carried out in the laboratory. Each combination was repeated ten times (320 predation experiments done).

The examination of each experiment occurred 24 hours after the exposition.

Statistical tests used in the present paper were as well one-tailed as two-tailed.

Results and Discussion

Results of larvae mensuration are mentioned in Tables 1 and 2. Few differences in length were observed as for prey larvae as for *C. tigripes* larvae used in this experimental study.

Table 3 presents the results of the predation experiments carried out in the laboratory.

Using the z-test for comparison of means, we noticed that larvae of *C. tigripes* maintain a predaceous activity over the 4 stages as indicated on the Fig. 1a.

When the larval density is higher (1.36 larvae/cm² in this study), the stages II, III and IV are not significantly different in their larvivorous activity ($p > 0.05$). The predation of larval stage III increases lightly without

being significantly different to those of stages II and IV. Nevertheless, when the larval density is lower (0.045 larvae/cm² in this study), predation activity of larval stages III and IV are statistically the same ; larval stage III becomes less performant in its larvivorous activity.

The light subsidence of predation (statistically non significant) observed to larval stage IV may be explained by the fact that the larvae getting ready to enter in a nonfeeding period (pupal stage) diminishes its diet.

The position of prey larvae in water does not influence the larvivorous behaviour of *C. tigripes*. *Anopheles* larvae have been eaten as well as *Culex* prey larvae (Table 3) even if they do not adopt a same position in the water. There is no significant difference between predation of *Anopheles* larvae and those of *Culex* prey larvae as well in the density 1 (Kruskal Wallis test $H = 1.64$; $p > 0.05$) as in the density 2 (Kruskal Wallis test $H = 0.43$; $p > 0.05$).

The positive correlation ($t = 4.15$; $p < 0.05$) found between the predation within both densities proves that larvivorous reflex of *C. tigripes* does not depend with the larval density, though the performance (number of larvae eaten by any time unit) depends on it (Table 3, Fig. 1a,b).

As for the prey larvae, the larval stages I and II are the most vulnerable (Fig. 1b) and their destruction by *C. tigripes* larvae is significantly different from those of stages III and IV ($p < 0.05$) in both experimental densities. The size of prey larvae, varying with larval stage has a significative impact on the predation inflicted by *C. tigripes*. Prey larvae of short length are more eaten than those of long length. However, this is not necessary question of preference. For, to satisfy its daily diet, *C. tigripes* has to consume much (in number) prey larvae of short length than it does with prey larvae of long length.

Table 1
Mensuration of the 4 larval stages of *Culex tigripes*

Larval Stages	Mean lenght (mm)	N
	X \pm SD	
Stage I	2.96 \pm 0.54	30
Stage II	4.65 \pm 1.06	30
Stage III	6.60 \pm 1.70	30
Stage IV	8.96 \pm 0.83	30

X : Mean
SD : Standard deviation
N : Samples on which mensuration have been taken.

Table 2
Mensuration of prey larvae referring to the four larval stages of *Anopheles gambiae* and *Anopheles funestus*

Group	Length spectre (mm) of different stages of <i>A. gambiae</i> and <i>A. funestus</i>	Mean length (mm)		N
		X	SD	
1	Stage I (1.00* - 2.00)	1.71 \pm 0.22		60
2	Stage II (2.10 - 3.00)	2.46 \pm 0.23		60
3	Stage III (3.10 - 4.00)	3.57 \pm 0.35		60
4	Stage IV (4.10 - 5.00)	4.41 \pm 0.39		60

X: Mean
SD : Standard deviation
N : Samples on which mensurations have been taken . 30 larvae of *A. gambiae* and 30 larvae of *A. funestus*
* Larvae which length was less than 1.00 mm have been rejected off the study , their manipulation being very delicate.

Table 3
Results of predation experiments carried out in the laboratory.

Experiment	% predation in Density 1 (1.36 larvae/cm ²)				% predation in Density 2 (0.045 larvae/cm ²)			
	Anopheles X ± SD	Culex X ± SD	Total prey X ± SD	N	Anopheles X ± SD	Culex X ± SD	Total prey X ± SD	N
PI x PI	29.20 ± 5.35	18.80 ± 13.54	48.00 ± 16.92	10	8.00 ± 5.66	9.00 ± 6.34	17.00 ± 11.52	10
PI x PII	11.60 ± 8.53	17.80 ± 10.22	29.40 ± 13.79	10	11.40 ± 7.23	8.40 ± 7.23	19.80 ± 9.50	10
PI x PIII	11.40 ± 6.19	12.80 ± 10.96	24.20 ± 13.68	10	8.00 ± 5.50	3.00 ± 3.30	11.00 ± 5.43	10
PI x PIV	4.80 ± 2.86	10.60 ± 5.42	15.40 ± 5.25	10	5.60 ± 3.63	5.00 ± 2.54	10.60 ± 4.43	10
PII x PI	25.80 ± 14.03	36.80 ± 15.75	62.60 ± 27.42	10	14.40 ± 5.48	19.40 ± 9.29	33.80 ± 7.86	10
PII x PII	27.60 ± 10.66	25.60 ± 11.65	53.20 ± 18.02	10	10.40 ± 12.28	10.40 ± 7.59	20.80 ± 12.12	10
PII x PIII	17.20 ± 7.55	21.40 ± 9.14	38.60 ± 12.54	10	12.00 ± 5.81	9.80 ± 5.37	21.80 ± 9.63	10
PII x PIV	13.40 ± 10.16	18.80 ± 7.13	32.20 ± 14.37	10	10.00 ± 6.93	8.20 ± 6.56	18.20 ± 10.73	10
PIII x PI	34.00 ± 10.29	36.80 ± 15.44	70.80 ± 25.00	10	26.00 ± 10.02	26.80 ± 11.59	52.80 ± 13.99	10
PIII x PII	34.60 ± 6.11	40.60 ± 6.26	75.20 ± 6.75	10	19.80 ± 6.56	24.40 ± 10.70	44.20 ± 14.89	10
PIII x PIII	18.40 ± 10.01	23.00 ± 9.76	41.40 ± 15.55	10	16.60 ± 6.99	10.60 ± 9.33	27.20 ± 13.17	10
PIII x PIV	15.80 ± 8.76	19.80 ± 9.95	35.60 ± 14.35	10	8.00 ± 5.33	10.40 ± 6.65	18.40 ± 7.29	10
PIV x PI	19.20 ± 12.51	24.40 ± 17.43	43.60 ± 27.09	10	25.20 ± 9.94	31.00 ± 7.67	56.20 ± 16.20	10
PIV x PII	26.00 ± 12.47	30.80 ± 12.90	56.80 ± 24.13	10	22.40 ± 9.13	22.60 ± 6.47	45.00 ± 6.75	10
PIV x PIII	15.60 ± 9.83	20.00 ± 8.48	35.60 ± 14.78	10	11.00 ± 4.83	10.20 ± 8.66	21.20 ± 10.63	10
PIV x PIV	14.60 ± 7.83	20.20 ± 10.47	34.80 ± 13.57	10	12.20 ± 9.82	5.60 ± 5.23	17.80 ± 14.09	10

P . Predaceous larvae p . Prey larvae
I, II, III, IV . Larval stages of mosquito
X ± SD . Mean ± Standard deviation of percentage of predation
N . Number of carried out experiments

It results from the study that all the 4 larval stages of *C. tigripes* have effectively a larvivorous activity and may provide a new natural control mean for malaria vectors. Yet, the real larvivorous power of *C. tigripes*, could be specified only after field experimentation.

The interest of the present study was to understand better the larvivorous behaviour of *C. tigripes*, predator of mosquito larvae and the impact of this species on larval density in one breeding place (laboratory experiment). Other studies are required to under-

stand the impact of this species on mosquito density in natural conditions by field studies.

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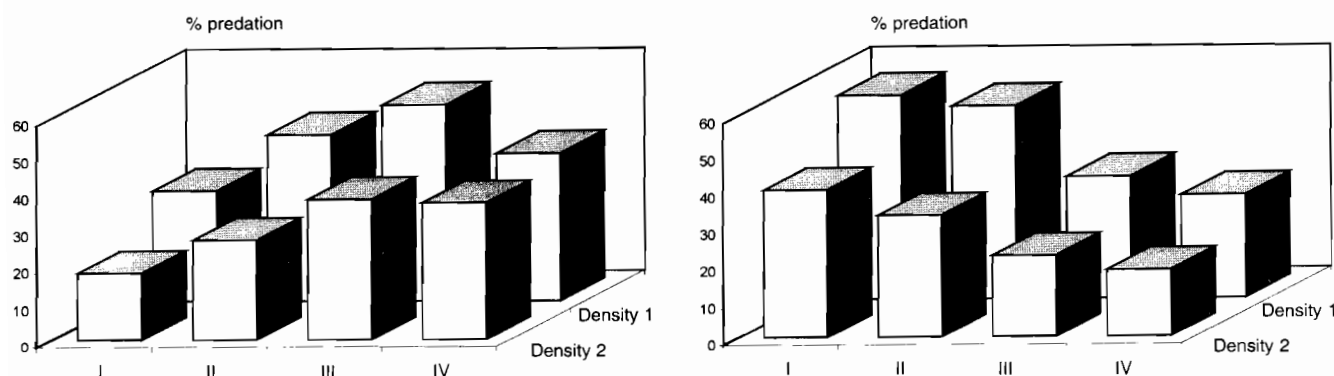


Fig. 1. Larvivorous potential of *Culex tigripes*
I, II, III, IV : Larval stages of mosquito
Density 1 : 1.360 larvae / cm²
Density 2 : 0.045 larvae / cm²

A . Predation of different stages of *C. tigripes*

B . Predation of different stages of prey larvae by *C. tigripes*

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