Impact of feacal coliforms and interspecific cohabitation on the productivity of culicidae breeding sites, littoral-Cameroon

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ABSTRACT

The demographic structure and emergence dynamics of Culicidae depend on biotic and abiotic interactions in the environment. The objective of this study is to determine the influence of bacteriological parameters, abundance, and interspecific cohabitation on the development of the pre-imaginal stages of Culicidae. Larvae were sampled using the “dipping” method and identified using dichotomous keys after rearing to adulthood; species of the Anopheles gambiae complex were distinguished using molecular techniques. The standard method for isolating and quantifying bacteria in water was used to quantify coliforms in water samples. The effect of density and interspecific cohabitation on the aquatic development of Culicidae was studied. The larvae were reared at different densities in a monospecific situation or in interspecific cohabitation. The number of individuals at each stage of development, the number of dead larvae, and the number of emergences were counted every 24 hours. The culicid fauna in Youpwé during the study period consisted of Culex quinquefasciatus (42.6%, n = 415), Anopheles coluzzii (39.8%, n = 388), and Aedes aegypti (17.6%, n = 171). Fecal coliforms were found at all breeding sites; the species identified were Enterobacter aerogenes (51.8%), Escherichia coli (6.5%), and Salmonella typhimurium (9.4%). A positive and significant correlation was observed between E. coli density and larval productivity of the genus Aedes (P-value = 0.002). In the laboratory, significantly slower pre-imaginal development was observed in a high-density interspecific cohabitation environment than in a monospecific environment. This was the case, for example, for the interspecific combination Anopheles gambiae/Culex quinquefasciatus, which had a significantly longer development time than Anopheles gambiae reared in a monospecific environment (P-value < 0.05). The same observation was made for the interspecific combination Anopheles culuzzii/Culex quinquefasciatus and Anopheles culuzzii in a monospecific environment (P-value < 0.05). In addition, a bias in favor of females in interspecific cohabitation was noted. It would therefore be appropriate, based on the results of more specific work on the mechanisms influencing fecal coliforms, to move towards integrated vector management. This requires the integration of sex ratio and cohabitation into entomological surveillance programs.

Keywords: Breeding Sites, Culicidae, Faecal Coliforms, Inter-Specific Cohabitation, Larval Productivity

I. INTRODUCTION

Blood-feeding Culicidae remain a major global health threat, as vectors of parasites and arboviruses that seriously impact human health. While malaria remains the deadliest parasitic disease, with approximately 282 million cases and 610 000 deaths, reported worldwide in 2024 (World Health Organization [WHO], 2025), the rapid spread of dengue fever shortly after the COVID-19 crisis is redefining priorities. In 2023, the world recorded a record 6.5 million cases, a figure already surpassed in August 2024 with more than 14.4 million cases reported worldwide (WHO, 2024 ; European Centre for Disease Prevention and Control [ECDC], 2026). In Cameroon, the circulation of dengue and chikungunya is now a documented epidemiological reality, requiring increased surveillance (Djeunang et al., 2022).

The effectiveness of conventional control tools, such as Long-Lasting Insecticided-treated Nets (LLITNs) and indoor residual spraying (IRS) with insecticides, is now compromised by the emergence of massive physiological resistance to pyrethroids in Anopheles gambiae s.l. (Akono et al., 2022 ; Koffi et al., 2023 ; Mbakop et al., 2022). At the same time, behavioural changes, such as exophagy, observed in these vectors allow them to circumvent domestic interventions (Akono et al., 2022; Djeunang et al., 2022). In this context, larval control using biolarvicides is emerging as a high-impact complementary strategy capable of reducing larval densities. In this context, larval control using biolarvicides is emerging as a high-impact complementary strategy capable of reducing larval densities by more than 60% in urban areas of Cameroon (Akono et al., 2022).

However, optimising this control in Cameroon requires a detailed understanding of larval ecology. Recent studies highlight that the productivity of breeding sites depends not only on physical, chemical and environmental factors (Antonio et al., 2009 ; Rejmankova et al., 2013), but also on complex interactions with bacterial microflora (Carnevale et al., 2009 ; Coon et al., 2016 ; Tawidian et al., 2021). Indeed, faecal coliforms and other environmental bacteria are not merely indicators of pollution, they also constitute an essential trophic resource that directly influences larval growth, survival and the vector capacity of emerging adults (Gimoneau et al., 2012; Tawidian et al., 2021). Despite these issues, the specific influence of bacterial load on Culicidae development remains insufficiently documented in Cameroon's aquatic ecosystems. This study aims to fill this gap by investigating the effect of bacteriological parameters and interspecific cohabitation on breeding site productivity in order to propose more targeted and sustainable vector control strategies.

1.1 Research Objectives

The main objective of this study, in an epidemiological context where vector control based on the use of Long-lasting insecticide-treated Nets (LLITNs) Indoor Residual Spraying (IRS) is losing its effectiveness, was to provide a complementary vector control strategy based on larval control. More specifically, the aim was to study the role of faecal coliforms in the larval development of culicidae and to assess the impact of multi-species cohabitation on larval productivity. In other words, the study sought to determine whether the simultaneous presence of several species influences larval survival, development time and sexing at emergence.

II. METHODOLOGY

2.1 Study Setting

This study was carried out in the city of Douala (03°40'- 04°11'N and 09°16'- 09°52'E), a wetland with numerous collections of water that are potential mosquito breeding grounds. The climate is hot and humid; the annual average temperature is 27°C and the average relative humidity 85%; abundant rainfall with an average of 3600mm of rain per year (World Meteorological Organisation [WMO], 2012) The air is constantly saturated with humidity, with an average of 99% relative humidity, in the rainy season from June to October, and 80% in the dry season (WMO, 2012). The hydrographic network consists of a main river, the Wouri, bordered by the Sanaga, the Dibamba, the Moungo and the Nyong. The vegetation is that of the low altitude rain forest. The relief consists of a set of wide valleys (Letouzey, 1985). Surveys were carried out in two localities with different degrees of urbanization: (i) Logbessou (04°05'N; 09° 46'E), a residential area in peril-urban area, straddling the districts of Douala 3 and Douala 5; which is established on a plateau, 3 meters above sea level. The primitive vegetation has gradually degraded under anthropogenic action. It is composed of formations of Grasses, Euphorbiaceae, Cesalpiniaceae, Sterculiaceae and Ulmaceae. Food crops visible in the rainy season are also components of this vegetation. The soil is ferralitic in nature, sandy-clay, yellowish and (ii) Youpwé (04°00'N; 09°42'E) a shanty town located in peri-urban area in the bank of Wouri, in the Douala 2 subdivision, at sea level. The district is bordered by mangroves and the soil is sandy.

2.2 Sampling, Isolation and Identification of Coliforms and Culicidae from Breeding Sites

The bacteria were sampled by taking 120ml of larval breeding water in previously sterilized and labeled glass vials. The surface spreading technique was used for culture by inoculating 20µl of water from the deposit into the "SCHERLAU Endo agar agar". The presumptive identification of bacteria which have developed after an incubation of 24 hours to 34 hours was done based on cultural characteristics (Guerin-Faublée et al., 1992). The colonies identified, were counted and the densities expressed in Colony-Forming Unit (CFU)/0.1ml of sample. The larval productivity was determined by counting the larvae collected, all stages combined, after a series of 5 200 ml releases for a total volume of one liter. The larvae collected were classified by breeding site and reared to adult stage according to the method of Desfontaine et al. (1991). Emerged adults were morphologically identified according to the keys of Holstein (1949), Gillies and De Meillon (1968), Gillies and Coetzee (1987) for the Anophelinae and Jupp (1996) for the Culicinae.

2.3 Laboratory Experimental Setup

The rearing device to examine the effect of densities and inter-specific coexistence of culicidae consisted of 24 transparent plastic tubes of 17cm long, 12cm wide and 5cm deep, containing 25cl of water each ; 12 tanks per study site and three replications for the larval densities of 30, 60, 90, and 120 mono-specific larvae or in cohabitation. For each larval density to be examined, there was a specific mono control tank. Every 24 hours, the number of individuals in each stage of development as well as the number of dead larvae was counted.

2.4 Statistical Analysis

The data were entered in an Excel sheet of Microsoft Office 2010 and the analyses were performed with SPSS software version 22.0 for Windows and PAST software version 4.0. Kaplan-Meier and Log Rank tests were used to analyse the mortality rate as a function of the number of pre-imaginal stages reared. Student's t-test for unpaired series and one-way ANOVA were used to compare the averages of development time, between two and more groups, respectively. The independence chi-square test was used to compare the sex ratio. Linear regression lines were obtained using PAST 4.0 software. SPSS 22.0 software was used to compare the means and determine Pearson correlations (r) between the productivity of each culicid species and the density of bacterial species. The significance threshold was set at the probability $p < 0.05$.

III. FINDINGS & DISCUSSION

3.1 Findings

3.1.1 Composition of Culicidian and Bacterial Fauna in Breeding Sites

A total of 959 mosquitoes were identified during the study. The genus *Culex* was the most common (42.6%; n=415), followed by the genus *Anopheles* (39.8%; n=388) and the genus *Aedes* (17.6%; n=171). The genus *Anopheles* was represented by *Anopheles coluzzii*, the genus *Culex* by *Culex quinquefasciatus* and the genus *Aedes* by *Aedes aegypti*. The bacteria in the faecal coliform group identified were *Enterobacter aerogenes*; the major species representing 51.8% of the samples collected, *Escherichia coli* 6.5% and *Salmonella typhimurium* 9.4% of the sample collected (Tab. 1).

Table 1

Frequence and Abundance of Bacterian Species in Culicidae Brinding Site in Youpwé

Bacterial species	Number of CFU/0.1ml	Frequence (%)
Escherichia coli	4635	6.5
Enterobacter aerogenes	37095	51.8
Salmonella typhimurium	6730	9.4
Shigella ; Pseudomonas ; Proteus	23160	32.3
Total	**71620**	**100**

CFU: Colony-Forming Unit

3.1.2 Effects of Bacteria on the Productivity and Composition of the Culicidian Fauna of Breeding Sites

A positive and significant correlation was observed between the density of *Escherichia coli* and the larval productivity of the genus *Aedes* (r = 0.510 ; P-value = 0.002). However, a negative but not significant correlation was observed between *Escherichia coli* density and the productivity of the *Anopheles* genus (r = - 0.181; P-value = 0.169) and *Culex* genus (r = - 0.148; P-value = 0.217) (Tab. 2).

Table 2

Influence of Bacteria on the Productivity of Culicidae (r= Spearman correlation; p=P-value)

Productivity	**Number of Colony-Forming Unit(CFU)/0.1ml**							
	Escherichia coli		***Enterobacter aerogenes***		***Salmonella typhimurium***		**Other bacteria**	
	r	P-value	r	P-value	r	P-value	r	P-value
Anopheles	-0.181	0.169	-0.141	0.228	0.159	0.201	-0.154	0.208
Culex	-0.148	0.217	0.161	0.198	0.138	0.234	0.057	0.382
Aedes	0.510	0.002	-0.144	0.224	-0.007	0.486	0.062	0.373

3.1.3 Effects of Density and Inter-Specific Cohabitation on the Sex Ratio, Development and Survival of Culicidae

The frequency of emergence of culicid females was less than 50% in the mono-specific culture tanks ; it was 47.1±7.3% in *A. gambiae* and 37.2±7.3% in *A. coluzzii*. However, in inter-specific cohabitation, emergence is in favor of females. It remained above 50% ; i.e. 72.2 ± 9.8% in *An. gambiae* and 65.9 ± 6.1% in *An. coluzzii* (tab. 3).

Table 3

Frequency of Emergence in Anopheles in Monospecific Cultures or in Cohabitation

Culicidae	**Number of individuals rearing**		**Number of individuals emerged (%)**		**Number of emerged female (%)**		**ddl**	**P-value**
	T	**C**	**T**	**C**	**T**	**C**		
An. gambiae	90	45	37(41,1)	21(46,6)	14(37,8)	17(80,9)	/	0.071§
	180	90	37(20,5)	34(37,7)	18(48,6)	25(73,5)	1	0.265
	270	135	54(20,0)	32(23,7)	29(53,7)	21(65,6)	/	0.717§
	360	180	54(15,0)	36(20,0)	24(44,4)	25(69,4)	1	0.232
An. coluzzii	90	45	53(58,8)	24(53,3)	23(43,4)	14(58,3)	/	0.673§
	180	90	53(29,4)	32(35,5)	15(28,3)	21(65,6)	1	0.057
	270	135	50(18,5)	37(27,4)	20(40,0)	27(72,9)	1	0.071
	360	180	52(14,4)	48(26,6)	18(34,6)	29(60,4)	1	0.118

T= mono-specific culture used as a control; C= inter-specific culture with 50% *An. gambiae* and 50% *An. Coluzii* ; Data are presented as numbers and percentages. The chi-square test of independence and Fisher's exact probability (§) were used to compare proportions. The significance level was set at P-value < 0.05. Pre-imaginal development was significantly slower in cohabitation than in mono-specific culture for *A. gambiae* (P-value<0.05) and *A. coluzzii* (P-value <0.05). However, in cohabitation, pre-imaginal development is significantly faster in *C. quinquefasciatus* than in *Anopheles* (P-value <0.05) (Tab. 4).

Table 4

Duration of Larval Transition from Stage 1 to adult in Culicidae Reared in Mono-Specific and Inter-Specific Cultures at Different Densities

Parameters	**Average Number of individuals reared**							
	30		**60**		**90**		**120**	
Specimen in culture	Ag	Ag/Cx	Ag	Ag/Cx	Ag	Ag/Cx	Ag	Ag/Cx
Duration to emergence (day)	14.8	15.5	14.7	15.6	15.8	16.3	15.3	16.7
P value	0.0084		0.0032		0.0876		0.0004	
Specimen in culture	Ag/Cx	Cx/Ag	Ag/Cx	Cx/Ag	Ag/Cx	Cx/Ag	Ag/Cx	Cx/Ag
Duration to emergence (day)	15.5	15.3	15.6	14.8	16.3	15.2	16.7	15.8
P value	0.0722		0.0001		0.0001		0.0001	
Specimen in culture	Ac	Ac/Cx	Ac	Ac/Cx	Ac	Ac/Cx	Ac	Ac/Cx
Duration to emergence (day)	10.3	10.7	12	12.5	12.2	15.2	13.7	15.8
P value	0.2193		0.0741		0.0001	0.0001	0.0001	
Specimen in culture	Ac/Cx	Cx/Ac	Ac/Cx	Cx/Ac	Ac/Cx	Cx/Ac	Ac/Cx	Cx/Ac
Duration to emergence (day)	10.7	11.2	12.5	11.8	15.2	14	15.8	15.3
P-value	0.0001		0.0001		0.0001		0.0001	

Ag=*Anopheles gambiae* control ; Cx=*Culex quinquefasciatus* ; Ag/Cx=cohabiting *Anopheles gambiae ;* Cx/Ag=cohabiting *Culex quinquefasciatus* ; Ac=control *Anopheles coluzzii ;* Ac/Cx=cohabiting *Anopheles coluzzii* ; Cx/Ac=cohabiting *Culex quinquefasciatus.*

Survival rates decreased non-significantly with increasing abundance of pre-imaginal stages in culture, regardless of culicid species (P-value >0.05) (Tab. 5).

Table 5

Survival Rates of Culicidae Reared in Mono-Specific and Inter-Specific at Different Densities

Number of individuals reared	**Jour 0**		**Jour 4**		**Jour 8**		**Jour 12**		**Jour 16**		**P-value**
	Ag (%)	Ag/Cx (%)	Ag (%)	Ag/Cx (%)	Ag (%)	Ag/Cx (%)	Ag (%)	Ag/Cx (%)	Ag (%)	Ag/Cx (%)	
30	100	100	93.3	84.3	77.6	75.4	66.5	64.4	40.9	46.4	0.670
60	100	100	89.3	8.2	62.7	64.3	28.7	40.9	21.6	31.1	0.087
90	100	100	78.4	73.2	63.5	50.9	22.6	33.1	16.4	25.0	0.553
120	100	100	74.2	76.0	46.4	57.6	30.5	32.8	17.8	22.2	0.396
	Ag/Cx (%)	Cx/Ag (%)	Ag/Cx (%)	Cx/Ag (%)	Ag/Cx (%)	Cx/Ag (%)	Ag/Cx (%)	Cx/Ag (%)	Ag/Cx (%)	Cx/Ag (%)	
30	100	100	84.3	91.1	75.4	73.3	64.4	44.2	46.4	37.8	0.715
60	100	100	83.2	79.9	64.3	49.9	40.9	27.8	31.1	0.0	0.584
90	100	100	73.2	78.4	50.9	39.4	33.1	33.1	25.0	28.9	0.634
120	100	100	76.0	68.9	57.7	52.0	32.8	31.1	22.2	22.7	0.512
	Ac (%)	Ac/Cx (%)	Ac (%)	Ac/Cx (%)	Ac (%)	Ac/Cx (%)	Ac (%)	Ac/Cx (%)	Ac (%)	Ac/Cx (%)	
30	100	100	75.5	84.4	60.0	71.1	56.6	55.5	0.0	0.0	0.670
60	100	100	77.8	76.7	70.0	64.4	52.8	48.8	0.0	35.5	0.523
90	100	100	69.6	77.0	55.9	69.6	49.6	53.3	28.5	25.9	0.300
120	100	100	77.5	76.1	65.8	71.6	50.2	43.9	23.3	26.1	0.041
	Ac/Cx (%)	Cx/Ac (%)	Ac/Cx (%)	Cx/Ac (%)	Ac/Cx (%)	Cx/Ac (%)	Ac/Cx (%)	Cx/Ac (%)	Ac/Cx (%)	Cx/Ac (%)	
30	100	100	84.4	93.3	71.1	70.5	55.5	28.9	0.0	0.0	0.256
60	100	100	76.7	93.3	64.4	71.1	48.9	55.5	35.5	32.2	0.432
90	100	100	77.0	97.0	69.6	79.2	53.3	64.4	25.9	32.6	0.814
120	100	100	76.1	75.5	71.6	62.2	43.9	48.9	26.1	28.9	0.522

Ag=*Anopheles gambiae* control ; Ag/Cx=*Anopheles gambiae* cohabiting with *Culex quinquefasciatus ;* Cx/Ag=*Culex quinquefasciatus* cohabiting with *Anopheles gambiae* ; Ac=*Anopheles coluzzii* control ; Ac/Cx= *Anopheles coluzzii* cohabiting with *Culex quinquefasciatus* ; Cx/Ac=*Culex quinquefasciatus* cohabiting with *Anopheles coluzzii.*

3.2 Discussion

A total of 974 mosquito larvae were collected in Youpwé, with a predominance of *C. quinquefasciatus* (42.6%; n=415), which is better adapted to the types of breeding sites found in urban areas (Hadji et al., 2013 ; Kbibch et al., 2009), characterized by pollution and the presence of household and industrial waste, as observed by Mbida *et al*., (2017) in the same locality. The predominance of the *Culex* genus is therefore a biological indicator of the urbanization of the environment (Darriet et al., 1986). The species *An. coluzzii* was the only of the *A. gambiae* complex present in these types of breeding sites ; this species is better adapted to the urban environment of coastal areas than the other twin species (Akono et al., 2022 ; Etang et al., 2016 ; Hoyochi et al., 2025 ; Mbida et al., 2017).

Bacteriological analysis of Culicidae breeding sites in Youpwé has shown that these areas are polluted with faecal peril bacteria due to anarchic urbanization and failure to observe hygiene rules. *E. aerogenes* (51.8%, n=37095UFC/0.1ml) is the predominant species; compared to other bacteria, it is able to multiply and live for a long period outside its host. *E. coli* and *S. typhimurium* are the least represented species with a cumulative frequency of 15.8% (n=11,365CFU/0.1ml). Their high sensitivity to pH and temperature is unfavourable to their survival in the outdoor environment (Bornet, 2000). Analysis of the effect of bacteria on the productivity of breeding sites has shown a positive and significant correlation between the larval productivity of the *Aedes* genus and the abundance of *E. coli*; the two species have a trophic relationship, bacteria being a food source for the *Aedes* genus (Harisson et al., 2023 , Merritt et al., 1992). Similarly, a negative and significant correlation was observed between *S. typhimurium* abundance and culicidian diversity; culicidian fauna would be sensitive to the toxins produced by this bacterium, which would have an impact on its development.

The sex ratio at emergence was in favour of females when the culicidae was raised in cohabitation. On the other hand, for mono-specific farms, the sex ratio was in favour of males. These results corroborate those of Kweka et al., (2012) who observed in Kenya more emergence of females compared to males when the species were reared in cohabitation and more emergence of males when the species were reared in mono-specific situations. Males appear to

be less able than females to cope with interspecific competition and the toxic substances emitted by different larvae, which would lengthen their development time in cohabitation. In a monospecific situation, males would emerge more rapidly than females, thus reducing their intraspecific competition time (Delatte et al., 2007); which tilts the sex ratio in their favour under natural conditions (Malaria Research and Reference Reagent Resource Center [MR4], 2010). However, females have a longer developmental time in the wild; their intra-specific competition for food resources and space is longer, which increases their mortality rate. Muturi et al. (2010) observed interspecific predation between *Culex* and *Anopheles* larvae when food sources are scarce; these researchers had identified DNA from each genus in the other's organism. The remains of carcasses observed in cohabitation ponds suggest this predation. Under natural conditions, interspecific cohabitation favours the emergence of female, hematophagous mosquitoes, thus increasing the risk of disease transmission to humans (Kweka et al., 2012). In addition, males that emerge in very small quantities would reduce the fertilization rate in females and influence the size of the population of the next generation. The productivity of the mono-specific roosts present, would supplement the small size of the male populations which would emerge from the roosts in sympatry, which would maintain, or even increase the population of Culicidae in the nature, in association with the effects of global warming which increase the temperatures (Duvalet, 2006 ; Muturi et al., 2010). A strategy to combat larvae against malaria would be to destroy the natural roosts, the roosts of choice for *Anopheles.* The consequence could be that gravid females are directed towards culicine breeding site to lay their eggs; the low density of male anophelines that go there will not be able to ensure optimal fertilization; this fact could reduce the anopheline population in subsequent generations.

Bioassays show that increasing the larval density of mosquitoes in culture media increases their mortality. This result corroborates the work of Gimnig et al., (2002) and Khandaker and Roitberg (2013), who showed that larval mortality is positively correlated with larval density and available space in the larval breeding site. Nekrasova (2003) mentioned that at high larval density, culicidal mortality increases and is multiplied by a factor varying from 1.5 to 2.2. Agnew *et al.*, (2002) noted that, specifically for insects, competition due to density is often associated with high mortality of larval stages, delayed maturity and a reduction in the emerging adult population. Indeed, higher densities encourage greater toxic production, which slows larval growth and causes high mortality (Roubaud & Tourmanoff, 1930). The development time of cohabiting Culicidae is significantly longer than in monospecific rearing, whatever the species. Competing larvae secrete a competitive substance that delays the development of other species. This substance had been called "Growth Retardant Factor (GRF)" by Moore & Fisher (1969), the higher the larval density, the higher its concentration in the environment (Moore & Wihtacre, 1972). Other studies have highlighted the existence of a factor which delays the development of culicidae in competition between *Aedes* and *Culex* (Sutcliffe & Benedict, 2012), between *Cx. quinquefasciatus* and *An. gambiae* (Kweka et al., 2012). In the wild, the delayed development of cohabiting larvae reduces their chances of emerging by increasing the risk of being eaten by predators, washed out, or having their breeding site destroyed. However, the development time of *An. coluzzii* remained shorter than that of *An. gambiae*, as also observed by Gimonneau et al., (2014), giving *An. coluzzii* a better chance of survival and disease transmission.

However, it should be noted that this work was carried out purely under laboratory conditions. Factors such as humidity, sunlight, and temperature could alter these results in either direction. Fieldwork will need to be carried out to compare the results and obtain a better assessment.

IV. CONCLUSION & RECOMMENDATIONS

4.1 Conclusion

For the first time in Cameroon, a study has been conducted to determine the impact of faecal bacteria on the development of the pre-imago stages of culicids, as well as the effect of the cohabitation of mosquito larvae of different species on their biological cycle. This preliminary study shows that the presence of these bacteria reduces the productivity of mosquito larvae. The cohabitation of different species of Culicidae prolongs the duration of development and increases the sex ratio in favour of females. This reveals a higher health risk because a Culicidae population with a higher proportion of females would lead to an increase in potential vectors and thus the risk of transmission of diseases such as malaria or dengue fever. Further research is therefore needed to determine the various mechanisms by which this faecal bacteria influence mosquito development and to limit as much as possible the proliferation of female insects due to their cohabitation. This will enable this aspect to be better addressed in order to improve epidemiological surveillance and risk management.

4.2 Recommendations

In light of the results obtained in this study, it is recommended that public authorities immediately incorporate sex ratio into health risk indicators for vector-borne diseases. A larval habitat that is “less productive” in terms of larval quantity but produces a very large number of females may be more risky than another that is highly productive but balanced. Therefore, targeting mixed breeding sites should be a priority in vector control campaigns in order to

break the prolonged development cycle that favours the selective survival of females. With regard to research, it would be wise to conduct further studies to determine the mechanisms of inhibition. That is, to identify whether the decline in larval productivity is due to bacterial toxins, acidification of the habitat, or competition for oxygen. Secondly, it is also recommendable to determine whether certain strains of specific faecal bacteria could be used as a natural control agent to limit larval density in urban areas.

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