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Influence of Ecological Setting on Occurrence of Artificial Container-breeding Vector Mosquito Species (Diptera: Culicidae) and Oviposition Attraction to Mineral Salts in Larval Habitats, in Minna, North-central Nigeria

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ABSTRACT: The peculiar challenges associated with mosquito vector control in developing urban areas, occasioned by the complex heterogeneity in vector behaviour and population dynamics, informed this study to elucidate the influence of eco-type on Mosquito species occurrence and responses to presence of mineral salts in oviposition sites. Two distinct ecotypes namely, semi-mineral and Urban, were selected as ovitrap stations, in Minna, Nigeria. In addition to the control experiment, adequately replicated ovitraps enriched with individual and combinations of nitrates, sulphates and phosphates were setup in the two ecotype sites and monitored for mosquito oviposition activities. The results indicated significantly ($P<0.05$) lower number of species and aggregate Mosquito densities in the urban (Mean=17.14±4.52 larvae/ovitrap) than semi-rural ecotype (21.76±3.93 larvae/ovitrap). While, three mosquito species: *Aedes aegypti*, *Culex p. pipiens* and *Cx. restuans,* were common to both ecotypes, additional three species namely; *Anopheles quadrimaculatus, Cx. Tarasalis* and *Cx. quinquefasciatus* were encountered in the semi-rural ecotype, though in low densities. Significantly higher larval densities were recorded in ovitraps enriched with mineral salts (range= 19.75±4.57 to 24.25±4.62 larvae/ovitrap) than the control (14.50±4.86 larvae/ovitrap). While, Nitrate ovitraps yielded the highest number of larvae (23.50±7.39 larvae/ovitrap) among the individual mineral salt-enriched ovitraps, and closely rivalled those containing combination of all three salts. The individual mosquito species demonstrated significant preferences for certain lone/single or combined salts-ovitraps. These findings revealed significant spatial heterogeneity in cosmopolitan mosquito population behaviour; and calls for the incorporation of diverse species-and ecotype-sensitive tools in mosquito vector control programs in such human communities.

KEYWORDS: *Aedes, Anopheles, Culex,* Ecotype, Mosquito distribution, Ovitrap

1 BACKGROUND

Mosquito-borne diseases including, malaria, Filariasis, encephalitis, yellow fever, dengue fever, etc, constitute the principal bane of public health challenges, particularly, in the Tropics [1]. Recent WHO statistics revealed that mosquito borne diseases are collectively responsible for several millions of human deaths and more than a billion cases of morbidity worldwide every year [2], [3]. Though, most of the mosquito-mediated diseases are extensively distributed on a global scale [4]; their local occurrence and intensity of transmission are closely related to those of the Culicid vectors [5], [6]. More importantly, however, the population ecology of vector mosquito species in a locality is largely determined by the suitability pf prevailing ecologic setting, for immature development and survival, as well as, dispersal of the adult stage [7]. While, mosquitoes breed profusely in naturally occurring water bodies [8], [9], [10], many of the vector species have evolved preferences for man-made water receptacles in peri-domestic environments [11], [12]. As a result of anthropogenic degradation of artificial mosquito breeding sites, the physico-chemical properties of the water bodies of artificial mosquito larval habitats have been significantly tilted in favour of the proliferation of certain species. This development have resulted in alteration of the otherwise mosquito species composition and relative abundance balance that often characterise natural ecological settings.

To this end, mosquito vector species composition and relative abundance, and hence transmitted diseases, have often demonstrated spatial heterogeneity in large human communities, especially where different ecological settings, dictated by degree of urbanisation, are contiguous. Thus, resulting in complex epidemiological patterns of mosquito-borne diseases; and making effective vector control difficult to attain. Most sub-Saharan urban settlements are typical conglomerations of diverse eco-types ranging from a cosmopolitan core to semi-rural outskirts [11]. Yet, the same anti-mosquito control strategies are implemented in all segments of such heterogeneous ecotypes [13], [14], irrespective of differences in vector behaviour and ecology, occasioned by spatial disparities in species occurrence and relative abundance. Therefore, in order to have a better understanding of the influence of ecotypes on mosquito population and oviposition behaviours in heterogeneous human communities, a necessary/vital information for effective control; this study was carried out to determine variabilities in species composition, relative abundance and oviposition behaviour of mosquito vectors in the representative ecotypes of Minna, North-central Nigeria.

2 MATERIALS AND METHODS

2.1 STUDY AREA

Located in North-central Nigeria, the study area, Minna (Long. 6^{o} 33'E and Lat. 9^{o} 37'N) is the capital city of Niger state. The area, covering an estimated 88 Km^{2} land mass, is inhabited by more than 1 million people. The climate of Minna is tropical, with two characteristic seasons namely, rainy (from May through October) and dry (December - March). Transitional seasonal periods occur in April and November. The area is relatively warm and dry, with mean annual temperature of about $30^{o}C$ and 60% relative humidity. Though, the total annual rainfall (1,334 mm) is effectively within the range for the Sudan savannah, the area is prone to water-logging during the middle of the rainy season, due to its poor drainage, occasioned by the characteristic pre-dominant low land *Fadama* edaphic and topographic features.

2.2 ECOTYPES

The Urban Setting: The urban ecotype selected for placement of ovitraps was located in Tunga area of Minna metropolis. The site is characterized by dense human population and modern housing structures. The drainage system is fairly well-planned. The vegetation is very scanty, consisting only of distantly distributed shade-providing trees and land-scaped horticultural species. Reasonable spaces around many of the houses are concreted/cemented and sometimes contiguous among a length of houses.

Semi-rural Ecotype: This eco-type was selected in the agrarian outskirt of Chanchaga sector of Minna, about 16 kilometres from the selected urban ecotype. The semi-rural setting was densely vegetated, with widely-spaced low standard houses thus, making room for inter-house expanse of open vegetated earth. Drainage system was practically non-existent in the area, resulting in proliferation of natural water receptacles in the rainy season. There were low anthropogenic pressures on the natural ecological attributes occasioned by low human population density.

2.3 EXPERIMENTAL DESIGN AND IDENTIFICATION OF MOSQUITO LARVAE

In each ecotype selected, four replicates of ovitraps consisting of a clay pot (3 litres capacity) holding 2 litres of rainwater were set up per treatment. The treatments consisted of the following: 9.50, 0.50 and 0.90 mg/Litre, respectively, of Nitrate (N), Phosphate (P) and Sulphate (S) salts individually dissolved in a set of four replicate ovitraps; as well as combinations of salt concentrations given above of Nitrate and Phosphate, N+P, Nitrate and Sulphate, N+S, Phosphate and Sulphate, P+S, and Nitrate, Phosphates and Sulphates, N+P+S. The salt concentrations used in this study were arrived at, been the average concentrations of such mineral salts in mosquito larval habitats in the study area [15], [16]. Also, a control experiment was set up, consisting of same number of ovitrap replicates as the test experiments, but with no mineral salt whatsoever dissolved in the water. The experimental setup was replicated at two stations in the field for seven days before mosquito larvae were recovered from them and taken to the Laboratory for identification. Seven days was considered long enough to attract ovipositing-mosquitoes, but not for adult emergence, as mosquito larval development takes between 9 – 15 days in the area [17], [18]. Mosquito identification was done to species level, using standard taxonomic keys [19], [20], [21]. The whole experiment was repeated within one week of termination of the first exercise.

2.4 DATA ANALYSIS

Data collected on mosquito larval density were processed as Mean±SD, using Microsoft Excel. Differences in mean values, among mosquito species, were compared for statistical significance using the Chi-square test; while the students' t-test was used for those between urban and semi-rural ecotypes. All statistical analyses were done at P=0.05 level of significance.

3 RESULTS

The differential occurrence and distribution of mosquito species in urban and semi-rural ecotype areas in Minna, North-central Nigeria, are highlighted in Table 1. Three mosquito species namely *Aedes aegypti, Culex pipiens pipiens* and *Cx. restuans,* were encountered in both eco-types but, in addition, three other species, i.e., *Anopheles quadrimaculatus, Cx, tarsalis* and *Cx. quinquefasciatus* were collected from the semi-rural eco-type. However, the additional mosquito species encountered in the semi-rural area occurred in significantly (P<0.05) low densities (range= 0.36±0.35 to 1.57±1.50 larvae/ovitrap). Significantly, *Cx. pipiens pipiens* was the most common species in both ecotypes (range= 9.29±2.78 to 11.47±3.35larvae/ovitrap). On the other hand, while *Cx. restuans* was the second most abundant mosquito in the urban setting (mean= 5.57±5.57 larvae/ovitrap), the species more-or-less shared the position with *Ae. Aegypti* in the semi-rural area (3.36±1.30 and 3.57±1.40 larvae/ovitrap respectively). Except for *Ae. Aegypti,* with respect to both individual species and aggregate mosquito collection, significantly higher densities were encountered in the semi-rural than urban eco-type.

Table 2 shows details of oviposition responses of the mosquito species to the presence of salt nutrients in the ovitraps. Significantly (P<0.05) lower aggregate larval density was recorded in the control ovitraps (mean=14.50±4.86 larvae/ovitrap) than any of the mineral salt-enriched ovitraps (range=19.75±4.57 to 24.25±4.62 larvae/ovitrap, in N+ S and N + P +S media, respectively). Individual attraction of the mineral salts for mosquito oviposition was significantly highest in the Nitrate (N) ovitraps (23.50±7.39 larvae/ovitrap), than the other salts, i.e. Sulphate (S) and Phosphate (P) which were almost equally attractive. Attractiveness of dual combinations of the mineral salts to mosquitoes were not significantly different (P>0.05) (range=19.75±4.57 to 21.25±3.94 larvae/ovitrap). However, combination of all three salts was significantly productive.

In term of number of species, the control and Sulphate (S) ovitraps were the least productive, as only three (3) species were encountered in them. While, only *An. quadrimaculatus* was absent in Nitrate ovitraps, those of Phosphates harboured all six (6) mosquito species encountered in this study. For ovitraps of salt combinations, only those of N + P were deficient in species representation, as *An. quadrimaculatus* and *Cx. tarsalis* were absent from their collections. Except for *Ae. Aegypti,* whose density distribution among the ovitraps varied within narrow limits (range=1.75±0.75 to 3.75±1.25 larvae/ovitrap), the mosquito species demonstrated oviposition preferences for individual or certain combinations of mineral salts. *An. quadrimaculatus* and *Cx. restuans* showed significant preference for P + S and N + P ovitraps, respectively. The *Cx. p. pipiens* mosquito had significant affinity for Sulphate, whether alone or combined with other salts. On the other hand, *Cx. tarsalis* and *Cx. quinquefasciatus* were preferentially attracted to ovitraps containing all the three mineral salts, although *Cx. tarsalis* showed a similar preference for Phosphate-enriched ovitraps.

4 DISCUSSION

The intensity of anthropogenic activities as reflected by differential ecotypes in Minna, North-central Nigeria, significantly influenced species occurrence and relative abundance of mosquitoes in the area. The higher number of mosquito species encountered in the semi-rural than urban setting recorded in this study, may be due to the greater diversity of larval breeding sites as well as adult ecological niches, characteristic of the former, i.e., rural ecotype, as earlier observed elsewhere in the tropics [22]. The requirements of urban development often results in clearance of natural climax vegetation and modifications of micro-climatic conditions. Thus, resulting in the simplification and/or elimination of natural larval breeding sites and adult resting sites in which many mosquito species have evolved to survive [23]. Usually, in urban areas, only few cosmopolitan mosquito species capable of utilizing the limited and often degraded larval habitats may occur [24]. However, the few mosquito species adapted to occurring in urban areas tend to have large densities, as a result of reduced competition and abundant supply of blood meal source. This phenomenon, however, contradicts the results of this study, as significantly higher aggregate mosquito density was recorded in the semi-rural than urban ecotype. The lower mosquito density recorded in urban ecotype may be due to the better anti-mosquito awareness and aggressive vector control activities in Nigerian urban centres [25]. Such anti-mosquito campaigns are directed primarily at the vectors of malaria, through the

WHO-supported wide-spread deployment of insecticide-treated bednets and elimination of larval habitats. This, probably, explains the absence of anopheles species from collections made in the urban ecotype.

Three mosquito species namely, *Ae. Aegypti, Cx. p. pipiens* and *Cx. restuans*, were collected in relatively high densities. Particularly, the first two (2) species are more serious vectors and nuisance mosquitoes of human peri-domestic environments, where they pose serious threats to public health. Thus, mosquito control strategies in the area must incorporate tools targeted at specific behaviour and ecology of the culicine mosquitoes as well; as such mosquitoes may be as important, epidemiologically, as the anophelines. The paucity of the three species restricted to the semi-rural area, i.e. *An. quadrimaculatus, Cx. tarsalis* and *Cx. quinquefasciatus* may be due to their ability to compete favourably with the more abundant species.

Significantly, higher Larval densities were recorded in ovitraps enriched with mineral salts than those containing only water (i.e., Control), even when such salts were present alone. This finding underscores the importance of mineral salts in mature development of mosquitoes. Mineral salts may affect mosquito larval development directly by moderating water physico-chemical parameters necessary for maintaining osmotic balance between the immature stages and the immediate environment or may affect them indirectly by enhancing the proliferation of phytoplankton [26], the major source of larval diet. More often than not, unguided peri-domestic anthropogenic activities result in the pollution of standing water bodies with mineral salts including those tested in this study, i.e., nitrates, phosphates and sulphates. Therefore, there is need to discourage the introduction of mineral salt-laden wastes into potential mosquito breeding sites, especially, where such habitats cannot be immediately eliminated or treated with larvicides.

Certain individual (e.g., Nitrate) and combination (e.g. N + P + S) of mineral salts were found to be significantly attractive to the mosquitoes while this may be due to biological requirements of the mosquitoes, it further confirms the ability of mosquitoes to discriminate among qualities of available oviposition [27] for optimum immature development. According to [28] the right choice of oviposition sites by gravid female mosquitoes, promote immature survival and vectorial fitness of emerging adults. To this end, chemical oviposition attractants of mosquitoes have been elucidated. In terms of number of species, the Sulphate (S) and Nitrate and Phosphate (N+P) ovitraps were the, respective, least productive individual and combined mineral salts media. This result does not show clearly, the relative importance of the mineral salts tested, for mosquito oviposition. It is, therefore, likely that other exogenous factors secondarily influenced by the mineral salts were involved in the attraction of the ovitraps to mosquito species. While. *Ae. aegypti* showed the least discrimination among the mineral salt-enriched ovitraps, relative to the Control, *Cx. p. pipiens* mosquitoes were particularly associated with sulphate ovitraps, whether alone or in combination. These observations are consistent with reported breeding ecology of the two mosquito species. *Ae. aegypti,* for example, is a foremost domestic container-breeding mosquito; and such containers are often found indoor and hardly receive nutrient salt inputs. This, probably, explains the reduced sensitivity of the *Ae. aegypti* mosquitoes to presence of mineral salts in the ovitraps. On the other hand, *Cx. p. pipiens* breeds intensely in highly polluted sites, such as septic tanks [29], [30], where sulphates constitute a major product of the decomposition of sewage.

Table 1. Mosquito species occurrence and relative abundance in urban and semi-rural ecological settings in Minna, North-central Nigeria.

Mosquito Species	**Ecological Setting**	
	Urban	Semi-Rural
Ae. Aegypti	2.29±0.59[a*]	3.57±1.40[a]
An. Quadrimaculatus	0.00±0.00[a]	0.36±0.35[b]
Cx. pipiens	9.29±2.78[a]	11.47±3.35[b]
Cx. restuans	5.57±5.75[b]	3.36±1.30[a]
Cx. tarsalis	0.00±0.00[a]	1.43±1.15[b]
Cx. quinquefasciatus	0.00±0.00[a]	1.57±1.50[b]
Aggregate	17.14±4.52[a]	21.76±3.93[b]

[*] Values followed by similar alphabets, in a row, are not significantly different at $P = 0.05$.

Table 2. Oviposition responses of mosquito species to presence of mineral salts in Ovitraps, in Minna, North-central Nigeria.

Mosquito Species	Control	Mineral Elements						
		NO_3	SO_4	PO_4	NO_3&SO_4	NO_3&PO_4	PO_4&SO_4	NO_3,PO_4&SO_4
Ae. Aegypti	2.50 ±1.13[ab]	3.75±1.25[b]	3.50±1.50[b]	2.75 ±0.25[ab]	1.75±0.75[a]	2.00±1.00[a]	3.00±0.00[b]	3.75±1.25[b]
An. quadrimaculatus	0.00±0.00[a]	0.00±0.00[a]	0.00±0.00[a]	0.50±0.32[b]	0.50±0.17[b]	0.00±0.00[a]	1.00±0.33[b]	0.50±0.15[b]
Cx. pipiens	9.50±3.29b[c*]	12.25±0.28[c]	13.25 ±1.75[d]	10.75±1.75[c]	13.50±2.50[d]	6.15±0.15[a]	8.25±2.75[b]	8.50±0.50[b]
Cx. restuans	2.50 ±0.44[a]	4.50±3.00[b]	3.50±1.50[a]	2.75±0.25[a]	2.00±0.50[a]	11.50±7.50[c]	3.50±0.50[a]	3.50±1.00[a]
Cx. tarsalis	0.00 ±0.00[a]	1.50±0.28[b]	0.00±0.00[a]	3.00±1.20[c]	1.00 ±0.26[b]	0.00±0.00[a]	1.50±0.42[b]	3.00±0.94[c]
Cx. quinquefasciatus	0.00 ±0.00[a]	1.50±1.11[b]	0.00±0.00[a]	1.50±0.17[b]	1.00±0.39[b]	0.50±0.43[b]	1.50±0.18[b]	5.00±0.78[c]
Aggregate	14.50±8.86[a]	23.50±7.39[c]	20.25±4.75[b]	21.25±3.94[b]	19.75±4.57[b]	20.15±9.08[b]	18.75±4.18[b]	24.25±4.62[c]

NO_3 = Nitrate; SO_4 = Sulphate; PO_4 = Phosphate

[*] Values followed by similar alphabets, in a row, are not significantly different at P = 0.05.

5 CONCLUSION

Ecological setting variation significantly influenced mosquito species occurrence and relative abundance in Minna Metropolis of North-central Nigeria. However, the encountered species may be those that are not vulnerable to the ongoing anti-anopheline campaign in the area. The mosquito species were significantly attracted to ovitraps enriched with mineral salts in whatever constitution. This portends serious threat to public health, as unavoidable mosquito larval breeding sites in the peri-domestic environments are often laden with anthropogenic wastes that decompose to release mineral nutrients that have served as oviposition attractants for mosquitoes in this study. Thus, mosquito control strategies in human communities with diverse eco-types must equally incorporate varied tools for effective and species-encompassing vector population reduction.

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