

Mini Review

Productivity of Malaria Vectors from Drainage Ditches in Niamey, Republic of Niger

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Abstract

Malaria is a public health priority in Niger. We carried out an entomological study in 10 neighborhoods of the city of Niamey. We collected mosquitoes every three days, from each site, for 15 days at the end of the dry season and 15 days during the mid-rainy season in 2011, 2013, and 2014. A single collection of larvae was carried out at a drainage ditch and a pond during the rainy season of 2016. We sought pre-imaginal stages at 16 breeding sites, 38% of which were open-air ditches for the drainage of domestic waste water and rainwater. *Anopheles* larvae were raised until adult emergence. In addition to harvesting larvae, adult mosquitoes were captured using light traps and knock-down insecticide sprays. We harvested a total of 10,551 *Culicidae* larvae, composed of *Anopheles* sp., *Culex* sp. and *Mansonia* sp. Twelve percent (12%) of the larvae collected were *Anopheles* sp. *Anopheles* larvae were found at four of the six surveyed open ditches, accounting for 12.7% of collected larvae. The search for adults yielded 20,274 *Culicidae* females (35.2% *Anopheles* sp.). This survey reveals that the open ditches of Niamey, despite being severely polluted are productive breeding grounds for *Anopheles gambiae* sensu lato (s.l.).

INTRODUCTION

Mosquitoes, like all Diptera, are insects that undergo complete metamorphosis, i.e. larvae and adults have a very different appearance and lifestyle. Immature stages live in an aquatic environment and adults in the open air. Mosquitoes have colonized diverse natural environments and adapt to many lifestyles due to their morphological variety and ecological plasticity. They exploit almost all types of aquatic habitats for breeding. Female mosquitoes lay eggs either on the surface of the water or on a wet substrate that may subsequently be flooded; the eggs hatch as soon as the embryo is fully developed.

Malaria is the deadliest of the parasitic diseases transmitted by mosquitoes. According to the WHO (2015), the number of malaria cases worldwide declined from 262 million in 2000 (range: 205-316 million) to 214 million in 2015 (range: 149-303 million), a reduction of 18%. The number of deaths due to malaria has dropped from approximately 839,000 in 2000 (range: 653,000-1.1 million) to 438,000 in 2015 (range: 236,000-635,000), a reduction of 48%. Most cases and deaths in 2015 were in Africa (88%).

The prevalence of malaria parasites in cities is generally lower than that observed in neighboring villages [1,2]. (Studies on the effects of urbanization on malaria transmission in sub-

Saharan Africa showed different transmission rates in urban, peri-urban, and rural areas [3,4]. Urban malaria transmission rates vary and are affected by a number of factors, such as location (altitude, proximity to the sea, rivers, etc.), climate, land use, socio-economic factors, the local vector species, waste management, and more. Factors associated with urban landscapes and the behavior of urban dwellers strongly influence the fate of anopheline populations and affect the reservoir of the parasite. Overall, the process of urbanization tends to reduce anopheline breeding sites [2,5] and is therefore expected to reduce malaria transmission. The inventory of urban breeding sites by [6], indicated that most are artificial, including urban agriculture, tires, and ditches; note however, that urbanization is a process and the vectors continuously adapt.

Malaria is transmitted by *An. gambiae* sensu stricto (s.s.) and/or *An. arabiensis* in most large modern African cities [5,7]. The breeding sites for these two species of the *An. gambiae* s.l. complex are small pools of fresh, shallow, calm, clear, sunny water. However, studies in both rural and urban areas in Africa have revealed the presence of *Anopheles* larvae, vectors of malaria, in waste water [8,9].

Malaria is a public health priority in Niger. It accounted for 28.3% of all morbidity recorded in 2014. Specific mortality due to malaria was 52.1%, and is higher in children under 5 years of

age with a rate of 62.3% (Directory of Health Statistics of Niger, 2015) [10]. The urban community of Niamey alone accounts for 10% of all malaria cases reported in Niger and the incidence has increased steadily in recent years. In the 1970s, there were few mosquito breeding sites in Niamey [11,12], and the transmission of malaria was higher in the neighborhoods near the river [13], than in other neighborhoods [14], except for unsanitary areas with standing water in pools and ponds. There are few entomological data for Niamey, and they are outdated, and from single surveys. This lack of knowledge is further exacerbated by the climatic changes of recent years, in particular the substantial decline in rainfall (20-30%) over the last 40 years and successive droughts [15]. Although attenuated by interventions of the National Program for the Fight against Malaria (PNLP), invasions of *Anopheles* throughout Niamey have not diminished. The speed and scale of population growth of Niamey has prevented the public authorities from providing adequate infrastructure, including an effective system of sanitation and rainwater drainage.

We report work to update knowledge about the inventory and geographical distribution of mosquitoes, and to evaluate the contribution of poorly maintained open ditches that surround the city of Niamey in the development of *Anopheles* larvae.

MATERIALS AND METHODS

Study site

Niger is a landlocked Sahelian country with an area of 1,267,000 km². It has environmental problems due to an imbalance between the human population, available resources, and economic development. In 1956, at the end of the colonial era, Niger was little urbanized: the urban population was 100,000 (3.6% of the total population), and Niamey had only 23,000 inhabitants [16]. Niamey, has been the capital of Niger from its independence to the present day, and was chosen because it was a healthy site, as well as for political reasons. It has witnessed spectacular population growth and expansion, leading to problems of urban development in the growing agglomerations because of insufficient technical and financial resources. The city is located in the south-west of the country between 13° 28' and 13°35' north latitude and meridians 02° 03' and 02° 12' longitude and covers an area of approximately 239 km² [17]. The Niger River flows through the city in a north-west-south-east direction and has a bed width of 300-750 m [18]. Niamey is also crossed by temporary tributaries (koris) of the river, which flow only during the rainy season (from June to September). The largest of these is the Gounti Yéna, which cuts across and drains the plateau of the left bank.

The climate is Sahelo-Sudanian with a rainy season of four months (June to September). Total rainfall averages 550 mm per season, with 76% of the precipitation between July and August. The average annual temperature is 29°C and maximum daytime temperatures 45°C in the shade, at the end of the dry season in April. Parallel to the increase in population, roughly organized agricultural activities have developed in urban and peri-urban areas (market gardening, rice cultivation). These activities ensure the supply of fresh produce to the capital, but also create favorable conditions for the formation of persistent pools and puddles, in which mosquitoes can breed, in addition to

the banks of the Niger River [19]. The amount of both solid and liquid waste in the city has grown, and there are only 150 km of sewers for an urbanized area of more than 11,000 ha with more than one million inhabitants. This network is poorly distributed and mostly serves the city center and neighboring districts; some districts only have large, East-West oriented open drainage ditches. This network is ineffective because of poor design and maintenance. The urban community of Niamey is divided into 99 administrative districts (Law n° 2002-012 of 11 June 2002) and five public health districts with 234 health facilities [20].

Choice of sites

Entomological surveys were carried out in 10 districts of Niamey, selected to represent or conform to the following criteria (Figure 1)

Homogeneous distribution to cover the entire urban area, and the five public health districts of Niamey;

Central or peripheral location of the neighborhood;

Development of organized or informal agricultural activities in the neighborhood;

Presence of breeding sites for vectors (ponds, river, drainage channels, etc.);

Various distances of the neighborhood from the river;

Health status of the neighborhood around the Integrated Health Centers (ISC);



Figure 1 Distribution of the study sites in 10 districts of Niamey where mosquitoes were collected.

Population density of the neighborhood

Capture of mosquitoes

We collected mosquito at each site, every three days for 15 days, at the end of the dry season (June) and for 15 days in the middle of the rainy season (September) of 2011, 2013, and 2014. We followed a total of 16 breeding sites in the 10 surveyed districts, including six open ditches, six ponds, one garden pit, and three sites at the river. A single collection of larvae was carried out at one of the open ditches for two consecutive days during the rainy season of 2016. Adult mosquitoes were identified with determination keys [21]. After morphological identification, the specimens were stored between -20° and -30°C until further analysis.

Aquatic stages collection and specimen processing: A semi-standardized protocol was used to search for larvae and pupae. The technique consisted of manually collecting aquatic stages of the mosquitoes by dipping a ladle into the water during the daytime [22]. The volume of the ladle was 350 mL (Bioquip, Gardena, CA, USA) as described by « Clarke Mosquito Control Products, Roselle, IL ». Ten (10) “scoops” were made per site and per pass and fifteen (15) “scoops” to the last passage of the rainy seasons. Mosquito larvae and pupae were immediately stored in a suitable jar containing water from the site and transported to the laboratory within three hours of collection. The larvae were morphologically identified and cultured, and the pupae identified up to the imaginal stage for confirmation.

Biological parameters of the early generations of Anopheles: We carried out a comparative study of pupal development, larval production, and adult production on larvae and pupae collected at an open ditch and those collected at a permanent pond during the 2016 rainy season (August). These pre-imaginal stages were followed under near-natural conditions at the insectarium of the CERMES (in a room without air conditioning, resulting in a temperature of the insectarium essentially identical to that outside). We evaluated three parameters:

Pupal development: the pupation rate was calculated as the ratio of the number of pupae obtained relative to the number of stage-IV larvae collected;

Larval production: the larval survival rate was calculated, relative to the number of larvae reaching stage IV, for each stage of larval development;

Attaining adulthood: The rate of emergence was calculated as the ratio of the number of adults to the number of pupae collected.

Adult mosquito collection: Two sampling methods for adult mosquitoes were used for each neighborhood and at each pass:

Capture of mosquitoes at rest after spraying with a knock-down insecticide (Spray Catch technique). Two rooms used as bedrooms were sampled in the early morning. Closed rooms were sprayed using commercial (Mobil®) aerosols containing pyrethroids to harvest residual fauna (endophilic fauna).

Mosquito capture using light traps. Two light traps (CDC mini

trap light, Model 512, John W. Hock Co., Gainesville, FL, USA) were used from 7:00 pm to 7:00 am outside bedrooms.

Geographic information system

All mosquito capture sites were geo-referenced using a Hand-held Global Positioning System (GPS). Characteristics, including the type of neighborhood and breeding site, the presence of vegetation, the presence of landfill, the nature of the water, the distance of the breeding site from the river, and the type of nearby crop were recorded. The number of people who spent the night in the room, the presence of insecticide-treated mosquito nets in the huts, and the presence of animals in the yard were recorded for adult mosquito sample collections.

Physicochemical quality analysis of surface water

Physicochemical measures and the presence or absence of aquatic organisms and micro-organisms were determined as approximate indicators of surface water quality.

Larval breeding sites were characterized using the following criteria for each site: the presence of predators, water acidity (pH), salinity, electrical conductivity (EC) and turbidity, and whether the location was anthropogenic or natural. A portable pH meter (HI 98130 HANNA instruments; USA) was used to measure pH, salinity, and EC.

RESULTS AND DISCUSSION

Abundance of Culicidae and Anopheles sp. in the neighborhoods of Niamey

We collected 10,551 *Culicidae* larvae during the monitoring of the breeding sites. Of these, 12% (1,255) were *Anopheles* sp. The *Culicidae* larvae were collected in 842 positive ladles cops of the 4,577 carried out. *Anopheles* sp. larvae accounted for 53% of *Culicidae* harvested in the central districts, but only 12 and 7% in peripheral and intermediate neighborhoods, respectively. During the same period, we captured 20,274 *Culicidae* adults, of which 35.2% (7,141) were *Anopheles* sp. Unlike the larvae, the proportion *Culicidae* adults that were *Anopheles* sp. was very low (5%) in the most urbanized central neighborhoods but high in peripheral (46%) and intermediate (28%) neighborhoods (Table 1). The *Anopheles* species found were *An. gambiae* sl. (94.8%), *An. pharoensis* (2.8%), *An. rufipes* (1.5%), *An. funestus* (0.5%), *An. ziemanni* (0.3%) and *An. nili* (0.1%). The other *Culicidae* included *Culex* sp. (59.8%), *Mansonia* sp. (2.5%) and *Aedes* sp. (2.5%).

Physicochemical characteristics of the water

We tested water at four types of breeding sites: permanent, semi-permanent, or temporary pools (or ponds); the river; a garden pit; and open ditches.

The maximum pH (8.2) was recorded in the river; the other water collection sites were less alkaline. The open ditches were well oxygenated (5.86 mg/L), whereas oxygen content was very low in the river (1.81 mg/L) and garden pit (1.57 mg/L). The EC of the water was high in the open ditches (1179.1µs/cm) and low in the garden pit (320.3µs/cm). The mean water temperature of the study sites ranged from 25.9°C (temporary pools) to 28.5°C (open ditches) (Table 2).

Table 1: Density of *Culicidae* and *Anopheles sp.* in neighborhoods of Niamey.

Neighborhood location	Neighborhoods	Immature			Adult		
		<i>Culicidae sp.</i>	<i>Anopheles sp.</i>		<i>Culicidae sp.</i>	<i>Anopheles sp.</i>	
		Total	Total	%	Total	Total	%
Periphery	Bangabana	4078	375	9%	3507	485	14%
	Banigoungou	1763	152	9%	5248	3669	70%
	Tondibia	62	53	85%	2581	1363	53%
	Foulan kouara	302	186	62%	953	91	10%
	Total 1	6205	766	12%	12289	5608	46%
Intermediate	Gamkalé	60	52	87%	1308	160	12%
	Karadjé	1192	219	18%	1194	359	30%
	Lamordé	1	1	100%	1923	818	43%
	Madina	2717	17	1%	505	50	10%
	Total 2	3970	289	7%	4930	1387	28%
Center	Boukoki	367	197	54%	1136	31	3%
	Wadata	9	3	33%	1919	115	6%
	Total 3	376	200	53%	3055	146	5%
	TOTAL	10551	1255	12%	20274	7141	35%

Table 2: Principal physicochemical measures of the water of the breeding sites.

Measure	Larval breeding sites					
	River	Pools/Ponds			Open Ditches	Garden pit
		Permanent	Semi-permanent	Temporary		
pH	8.2	7.9	7.8	7.9	7.6	7.8
Dissolved oxygen (mg/L)	1.81	3.98	3.23	2.58	5.86	1.57
Conductivity (µs/cm)	378.2	743.8	667.8	645.5	1179.1	320.3
Water temperature (°C)	27.5	28.1	28.4	25.9	28.5	26.6

Abundance of *Anopheles sp.* at the larval breeding sites

During the survey, a total of 4,607 ladle scoops were made, resulting in the collection of 1,255 malaria vector larvae. The surveys of the breeding sites showed that the diverse environments tested were all exploited by both *Culex sp.* and *Anopheles sp.* The mean number of *Anopheles sp.* larvae per ladle scoop was 0.27, (range 0.01 in the temporary pools to 0.72 in the permanent pools). Larval abundance varied widely in time and space. During the dry season, the abundance of *Anopheles sp.* larvae was very low at all surveyed sites, except permanent ponds; indeed, more than half (52%) of the larvae collected during the study were from such ponds. Only three *Anopheles sp.* larvae were collected from temporary pools during the survey and thus these pools appear to play a negligible role in the transmission of malaria (Table 3).

Abundance of *Anopheles sp.* in ditches

Culicidae larvae were present in 387 of the 1,724 ladle scoops from ditches (Table 4). We found *Anopheles sp.* larvae in four of the six open ditches surveyed, and they made up 2.3% of the *Culicidae* larvae collected. There were, on average, 0.41 *Anopheles sp.* larvae per positive ladle scoop. *Anopheles sp.* larvae were most abundant in the ditches of the Foulan Kouara district and were not observed in the ditches in the districts Boukoki and Gamkalé.

Biological characteristics of the early generations of *Anopheles sp.*

Anopheles sp. larvae collected from an open ditch and a permanent pond were reared and the first generation studied. We collected 2,311 larvae from a pond and 79 from a ditch during two collections during the rainy season of 2016. We followed the pre-imaginal development cycle in the laboratory for 10 days. Pupation and larval survival rates were higher for larvae collected from the ditches (31.4 and 44.3%, respectively) than for those collected from the pond (27.3% and 37.9%, respectively). In contrast, the emergence rate was higher for pupae from the ponds (40.6%) than those from the ditches (36.4%) (Table 5).

CONCLUSION

We report an inventory of the mosquitoes in the capital of Niger (Niamey): we describe their geographical distribution, abundance, seasonal variation and relationship with the environment and in particular the involvement of open ditches in the life cycle of *Anopheles gambiae sl*, the principal malaria vector. The two genera of *Culicidae* collected in the largest numbers in Niamey were *Culex sp.* and *Anopheles sp.* The biological ecology of malaria vectors in urban areas in Niamey is characterized by low Anophelian density in the most highly urbanized areas and high heterogeneity. These features are consequences of the diversity of local ecology in each neighborhood in Niamey, as we had already observed in 2002 and 2008 [19]. In 1997, Julvez et al.

Table 3: Larval breeding sites and seasonal variation of malaria vectors.

Year	Season	Number of ladle scoops	Number of <i>Anopheles sp.</i> larvae						Total	Per ladle dip
			Larval breeding sites							
			River	Pools/ponds			Open ditches	Garden pit		
	Permanent	Semi-permanent	Temporary							
2011	Rainy	1387	74	174	94	0	98	8	448	0.32
	Dry	515	0	26	0	0	0	0	26	0.05
2013	Rainy	770	27	243	2	3	60	95	430	0.56
	Dry	640	0	131	0	0	0	0	131	0.20
2014	Rainy	825	5	81	83	0	2	49	220	0.27
	Dry	470	0	0	0	0	0	0	0	0
	Total	4607	106	655	179	3	160	152	1255	0.27
	Percentage		8%	52%	14%	0%	13%	12%		
Total ladle scoops			807	1052	247	527	1724	250	4607	
Larvae per ladle scoop			0.13	0.62	0.72	0.01	0.09	0.61	0.27	

Table 4: Density of *Culicidae* and *Anopheles sp.* in the ditches.

Neighborhood	Number of ladle scoops	Number of ladle scoops positive	Number of <i>Culicidae</i>	<i>Anopheles sp.</i>	
				Total	Per positive ladle scoop
Bangabana	315	158	3183	2	0.01
Boukoki	336	0	0	0	0
Foulan Kouara	167	49	195	101	2.06
Gamkalé	276	0	0	0	0
Karadjé	350	74	836	40	0.54
Madina	280	106	2691	17	0.16
Total	1724	387	6905	160	0.41

Table 5: Pre-imaginal development of *Anopheles sp.* in the laboratory.

Origin of larvae	Larval survival rate (%)	Rate of pupation (%)	Rate of emergence (%)
Channel	44.3 (35/79)	31.4 (11/35)	36.4 (4/11)
Pool	37.9 (876/2311)	27.3 (239/876)	40.6 (97/239)

[14], observed that malaria transmission in Niamey was higher in neighborhoods near the river than in other neighborhoods. Labbo et al., 2016 [19], observed that the hydrological situation in Niamey has not changed much over the last decade. But the author noted the change in malaria transmission. The main reason is that the data of Julvez et al., are old (1992-1995). In addition, climate change in recent years has seen a dramatic decline in precipitation (20-30%) and subsequent drought. [23]

During the survey, we monitored 16 breeding sites. Although very different, these environments are exploited by both *Culex sp.* and *Anopheles sp.* We found most *Anopheles sp.* larvae in permanent pools, consistent with our observations in 2002 [19]. Fluctuations in mosquito densities were very marked during the periods of heavy rain, whereas their numbers were relatively constant before this period. The emergence of adult vectors is abrupt and relatively asynchronous between sites, probably due to the pressure of human and environmental factors specific to each site. The consequence is heterogeneous

malaria transmission in Niamey, each neighborhood probably constituting a particular case. This notion of heterogeneous transmission within a relatively small-urbanized area (250 km²) is consistent with the clinical data, showing high heterogeneity in the number of cases reported annually in the 60 ICS of Niamey (MSP, PAA2017).

The open ditches in Niamey are productive breeding sites for *Anopheles*: larvae from these breeding sites were productive and pre imaginal development was observed in the laboratory. We found no species of *Anopheles*, other than *Anopheles gambiae sl.* (the principal vector of malaria), in these ditches. The physicochemical measures of the water in which we found *Anopheles* were similar to those found by [24], in the polluted waters of Lagos. Anopheline larvae were present in two thirds (2/3) of the open channels surveyed. Entomological studies of the ditches in Niamey have not been performed for 40 years [11,12]. Anopheline larvae generally develop in relatively clean water, unlike the larvae of *Culex quinquefasciatus*, which can grow in water, contaminated with organic matter and are readily found in urban areas where hygiene is poor [25]. However, larvae of *An. gambiae sl.* are found in various sources of polluted water in urban environments in sub-Saharan Africa [9,26,27], although rarely. Nevertheless, some studies [4,28,29] have concluded that water pollution in cities is the main factor limiting the development of anophelines.

Our study updates knowledge about mosquitoes in general and, in particular, the ecology of the principal vectors of malaria

in Niamey. The presence of *An. gambiae* sl. larvae in open ditches has serious implications for the epidemiology of urban malaria. Changes in the behavior of anopheles and their opportunism in the face of environmental changes must be considered before planning, executing, and evaluating vector control measures in general, and in particular those implemented by the Malaria Control Program in Niger [30].

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