Impact of indoor residual spraying on malaria transmission in the Moïssala health district, Chad

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Keywords: Malaria, Impact of transmission, Indoor spraying, Moïssala Health District, Sudano-Guinean zone, Chad

Posted Date: December 31st, 2019

DOI: <https://doi.org/10.21203/rs.2.19781/v1>

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Abstract

Background

Malaria is a major public health and development problem in Africa. In Chad in 2016, with 720 765 confirmed cases and 1 686 deaths, malaria is the main cause of consultations, hospitalizations and deaths in health facilities. A longitudinal entomological study was carried out from 07 to 24 December 2016 in the Moïssala health district. The objective of the study was to assess the impact of malaria transmission one year after two cycles of indoor residual spraying of 80% bendiocarb wettable powder (Ficam WP 80 W).

Methods

Two areas were defined for the study: the town of Dembo, located in the sprayed area, was chosen as the test town and Moïssala, located in the untreated area, was chosen as the control town. Two sampling methods were used: collection of resting endophilic fauna and direct capture of aggressive mosquitoes from human subjects.

Results

A total of 16 sessions of catches on human subjects totalling 32 man-nights were conducted and 160 rooms were sprayed per site. For 160 sprayed chambers per site, two anopheles were captured in Dembo compared to 547 in Moïssala. Three anophelian species have been morphologically identified. In Moïssala, *An gambiae* sl (*An colluzzii*: 96%, *An gambiae*: 4%), *An funestus* and *An rufipes* were captured at rest in the rooms and on human bait. In Dembo, the two anopheles, *An colluzzii* and *An funestus* were captured at rest in the rooms. On human bait as in endophilic fauna, *An gambiae* sp was the most common species in Moïssala. The aggressive anophelian density was zero in Dembo while it is 9 bites per man per night in Moïssala. Only *Anopheles gambiae* sp was found infected in aggressive wildlife. The sporozoite index of the aggressive fauna of *An gambiae* sp was 7.45%. Malaria transmission was estimated at 0.67 infected bites per man per night, or 244.55 infected bites per man per year. In endophilic fauna, *An gambiae* sp and *An rufipes* were the two species found infected in Moïssala with sporozoite indices of 6.70% (23/343) and 20% (2/10) respectively. However, in Dembo, neither of the two captured individuals was found infected.

Conclusions

The indoor residual spraying campaign in the eastern zone of the Moïssala health district has led to the collapse of the density and aggressiveness of malaria vectors. However, its evaluation over a short period of time is not sufficient to assess the impact of malaria transmission in this stable and highly endemic malaria zone.

Introduction

Malaria is a major public health and development problem in Africa. In 2016, 216 million cases of malaria were reported in 91 countries, an increase of 5 million from the previous year and the number of deaths reached 445 000. Africa still accounts for 90% of the associated cases and deaths [1].

However, since its creation, the World Health Organization has always had malaria eradication on its agenda [2]. Efforts have been made to eradicate this scourge in many parts of the world [3]. However, in Africa, eradication is being delayed not only for economic reasons, but also because of insufficient knowledge of how the disease is transmitted in the continent's different ecological areas.

In Chad in 2016, with 720 765 confirmed cases and 1 686 deaths, malaria is the main cause of consultations (34%), hospitalizations (30%) and deaths (35%) in health facilities [4]. In addition to the loss of human lives, malaria is costly in terms of public health expenditures [5].

Divided into three geo-climatic zones that determine three epidemiological facies of malaria, northern Chad is desert with no local transmission [6]. The Sahelian climate center corresponds to an area with unstable malaria due to short seasonal transmission. The south with a Sudanian to Sudano-Guinean climate is characterized by stable malaria, with long seasonal transmission.

Most of the entomological studies conducted in Chad have concerned the Sudanian and Sahelian zone where malaria transmission is attributed to *An gambiae sl*, *An funestus*, *An pharoensis* and *An ziemanni* [7, 8]. Yet, to date, no survey has assessed the dynamics of malaria transmission in the transition zone between the Sudanese and Guinean zones. However, the epidemiology of malaria depends largely on the biotope.

Our survey focuses on malaria transmission. It was carried out at the request of the non-governmental organization "Médecins Sans Frontière-France (MSF-France)" to evaluate the effectiveness of the indoor spraying campaign carried out in the eastern zone of the Moïssala health district. Since 2010, MSF-France has been working in the Moïssala health district in support of the Ministry of Public Health and the National Malaria Control Program. It has developed preventive (awareness, seasonal malaria chemoprophylaxis) and curative (seasonal malaria hospital management unit) activities.

In an effort to strengthen protection against mosquito bites in addition to long-lasting insecticide-treated nets distributed by the Ministry of Public Health, MSF-France considered using Fendona® (a pyrethroid insecticide) for an indoor residual spraying campaign in the eastern zone of the Moïssala health district. The recommendations of the NMCP following a study carried out in 2015 on mosquito susceptibility in the said health district (data presented in another article) finally led to the use of Ficam® (insecticides of the Carbamates class).

The objective of the study is to evaluate the impact of this spraying campaign on the dynamics of malaria transmission in two villages, one (Dembo) in the sprayed area and the other (Moïssala) in the control area. More specifically, this involves determining the aggressive density of mosquito populations and assessing the rate of plasmodial infection of vectors.

Materials And Methods

Study area and population

To assess the impact of this campaign, two sites were selected. These are Dembo, located in the sprayed area and Moïssala, which was not treated at all, served as a control.

Moïssala (8°20'16"N 17°46'05"E) is the chief-town of the department of Barh Sara, Mandoul Region. It is located 75 km from Koumra, the capital of the region and more than 700 km south of N'Djamena, the capital of Chad. It has a population of approximately 33 098. The city of Moïssala is located on the banks of a permanent river, the Barh Sara, a tributary of the Chari River that originates in the Central African Republic. The river can be crossed with a ferry or a canoe. However, during the rainy season and depending on the level of the floods, the crossing with the ferry is not always guaranteed. The alternative is the road for a two-day trip to Dembo.

Dembo is the chief-town of the Dembo Sub-Prefecture in the department of Barh Sara. It is located 25 km east of Moïssala and about 27 km north of the border with the Central African Republic. It has a population of approximately 19 013. The city is bordered by a pond (Goumoud) which is watered in the rainy season and dried up in the dry season. During wintering, swamps form between Dembo and Moïssala, making traffic extremely difficult. Sometimes, when the rains are very heavy, the road is impassable throughout the season but this difficulty disappears more or less completely towards the end of the dry season.

The two cities have geographical and cultural similarities. The climate is tropical of the Sudano-Guinean type, characterized by the alternation of a long rainy season, from May to November, and a dry season, from December to April. The average annual rainfall exceeds 900 mm per year with a maximum of rainfall in August. The average annual temperatures range from 17°C in December/January to 40°C in March/April. Soils are generally sandy or clayey with a high humus content. In some places, soil impermeability causes swampy areas during the rainy season. The once heavily forested region has undergone intense clearing due to high population pressure.

The forest, which consists of very large trees (fig trees, "caïlcédrats", tamarind trees, Karités, "Nérés", etc.), vines and epiphytic plants, has suffered particularly from human activity, which has largely cleared it to replace it with food crops, but now only remains in the form of highly degraded galleries along watercourses.

The main crops are: sorghum, groundnuts, rice, cassava, cotton, beans, sesame and market gardening. The population is composed of Mbaye, Nar, Gor, Gor, Ngama, Daye and Peulhs ethnic groups. The activities practiced by the population are: traditional rainfed agriculture, fruit growing, animal husbandry, trade and fishing. Large and small livestock are parked at night in enclosures in the vicinity of houses.

Most of the houses are of traditional, rectangular and circular types, with dry clay or brick walls. Their roofs are made of metal sheet or thatch. The population is supplied with drinking water through

traditional wells and boreholes equipped with human-powered pumps.

The main diseases of the inhabitants of Moïssala and Dembo are in order of importance from the point of view of the number of cases reported by health facilities: malaria, mainly *Plasmodium falciparum*, is endemic, malnutrition, measles, meningitis, etc.

Entomological data from the region are non-existent. Both cities use the same vector control methods based on the use of long-lasting insecticide-treated nets widely distributed to all households during the 2014 mass campaign.

The study took place from 07th to 24th December 2016 in four of the eight districts of the city of Moïssala and in four of the seven districts that make up the city of Dembo (Figure 1).

Methodology

Sampling techniques

Mosquito population sampling was carried out by two methods of capture:

1. Direct capture on human bait at night (indoor and outdoor);
2. Intra-domestic spraying of pyrethrum-based insecticide (Red Can®) during the day.

Capture on human

Aggressive fauna on humans has been studied using the Le Goff et al. method (9). For this method, 3 parameters were determined:

1. The aggressiveness rate (**ma**) which is the number of mosquito bites per person per night;
2. The sporozoite index (**SI**) of the captured mosquitoes;
3. The daily inoculation rate (**h = ma x SI**).

The catches were made by volunteers, all young men recruited in each city. These volunteers were previously trained after obtaining their consent. "Men-night" are used both as bait and as captors. They were protected from malaria by Sulfadoxine-Pyrimethamine chemoprophylaxis. Catches were made for four consecutive nights in four neighbourhoods of each site, with one hut per neighbourhood. This corresponds to 16 catch sessions per site, or 32 "men-night". The neighbourhoods were selected according to a peripheral-intermediate-central transect. For each capture point, the consent of the head of the household was obtained for the access of the catchers to their household. For each compound, two catch points are retained. One inside the hut (used as a bedroom) and the other outside. The captures are organized from 6pm to 6am by two teams of two capturers each. The first team operates from 6pm to midnight and the second from midnight to 6am.

Volunteers are rotated at each capture session (between teams, capture points and houses) to minimize biases due to their individual ability and attractiveness. Each capturer is equipped with a torch, a watch, hemolysis tubes and bags labelled by time slot and bearing the identification of the capture point. In a practical way, each volunteer, seated on a bench/stool/chair, captures with a hemolysis tube the mosquitoes placed on his bare legs up to his knees. The hemolysis tubes containing the mosquitoes are then placed in bags corresponding to catch time slots.

Capture of endophilic fauna after indoor residual spraying with insecticides

This method was used for the collection of female endophilic mosquitoes to assess the density of resting mosquitoes in bedrooms. It consists of spraying a pyrethrum solution into bedrooms to collect mosquitoes at rest. The insecticides used are commercial products based on ORO brand Pyrethroids (permethrin: 0.25%; tetramethrin: 0.20%; d-ferothrin: 0.01%; piperonyl butoxide: 0.34%).

Before each spray, white bed sheets were spread to cover the entire floor of the room and the furniture, then an agent protected by a mask and glasses, after closing the door and windows, sprays the room for about fifteen seconds before leaving. After about ten minutes of waiting, the sheets are carefully removed and the lightning-fallen mosquitoes are recovered with the help of pliers in petri dishes bearing indications on the site, the district, the compound and room number. These spraying sessions take place between 6am and 11am.

Treatment of captured mosquitoes

Mosquitoes captured by the above methods are identified using a binocular magnifying glass according to the morphological genus and species criteria [10]. Females of anopheles are then counted and classified according to the location, time and method of capture, repletion state of their abdomen (unfed, fed, half-gravid, gravid) and individually deposited in microtubes containing a desiccator (silicagel) for transport and storage. The microtubes are assembled in bags and stored at minus 20°C for further analysis at the laboratory of "Laboratoire du Centre de Recherche Entomologique de Cotonou (Benin)". The determination of the circumsporozoite antigen of malaria parasites in mosquitoes was done by the Elisa technique [11, 12]. Molecular identification of *An gambiae* complex species was performed by PCR SINE 200 according to the protocol of Santolamazza et al [13].

Statistical analysis

All data recorded on the survey sheets were entered into Microsoft Excel software and transferred to version 20 of the IBM SPSS statistical analysis software. The daily aggressiveness rate (**ma**) was calculated from the number of bites received per man per night. **m** represents the number of mosquitoes per man and **a**, the daily frequency of bites performed by a female anopheles on a man. The sporozoite index corresponds to the presence in females of anopheles of the circumsporozoite antigen established by ELISA CSP research. This index is estimated in number of infected female anopheles out of the total number of anopheles analysed times one hundred. The daily entomological inoculation rate (**TIE = mas**)

was calculated from the product of the daily aggressive density (**ma**) and the proportion of females with CSP antigen (s). The daily entomological inoculation rate allows the monthly and annual inoculation rate to be deducted. Anophelian density is the product of endophilic anophelian fauna and the number of rooms sprayed.

Results

Anopheles species present

Three anopheles species have been identified in Moïssala in the control area. These are *An gambiae sl*, *An funestus* and *An rufipes*, whereas in Dembo in the treated area, *An gambiae sl* and *An funestus* were collected (Table 1).

Molecular forms within the *An. gambiae* complex

Of the 386 individuals in *An gambiae* complex randomly selected and subjected to PCR molecular forms, *An colluzzii* is 96% (371/385) in majority compared to *An gambiae* 4% (14/385) in Moïssala. In Dembo, the only individual caught belongs to *An coluzzii* species.

Aggressive fauna

Time rhythm and aggressiveness rate

In Moïssala, the aggressiveness of mosquitoes starts from 6pm and continues throughout the night both indoor and outdoor. There is a high rate of biting between 10 p.m. and 4 a.m., no matter where you are. Inside, the bite peak occurs around 3 a.m. while outside, a high rate is reached earlier, between 10 p.m. and 11 p.m., and a second one between midnight and 1 a.m.

During the four successive night captures, 303 anopheles were captured in Moïssala while in Dembo, the aggressiveness rate was zero for the same number of capture sessions. The 303 anopheles were captured during 16 capture sessions totalling 32 Men-night. The average aggressive anophelian density in Moïssala was 9 bites/man/night (Figure 2).

Anopheles gambiae sl bites both inside and outside houses. However, 55% of the females were caught indoors, indicating the tendency of this species to endophagy. On the other hand, *Anopheles funestus* is exclusively caught outdoors. The distribution of the number of anopheles caught on humans is summarized in Table 2 for each species (Table 2).

Sporozoite indices of aggressive fauna

The sporozoite antigen indices tested in *An funestus* and *An rufipes* were negative. On the other hand, out of a total of 295 *Anopheles gambiae sl* randomly selected and observed at Elisa for infection, 22 were found to carry Plasmodium sporozoites, representing a sporozoite index of 7.45% (Table 3).

The entomological inoculation rate

The average entomological inoculation rate of *Anopheles gambiae* sl in Moïssala during the study period was 0.67 infected bites per man per night, or 244.73 infected bites per man per year.

Time distribution of malaria transmission in Moïssala

The time aggressiveness of Moïssala's mosquitoes starts from 6pm and continues throughout the night. However, the schedules of infected bites are observed from 8pm and persist throughout the rest of the night. However, transmission is null between 6pm and 8pm as well as in the early morning. The high transmission rate occurs during aggressiveness peaks, reaching an average of a maximum of 5 infested bites per person per night during the survey period

Endophilic fauna

Density of anopheles per room

During the capture of the morning residual fauna, 547 anopheles were captured in Moïssala against 2 in Dembo for a total of 160 sprayed rooms per locality, i.e. an average density of 3.41 anopheles per room in Moïssala against 0.01 in Dembo. *An gambiae* sp is by far the most frequent anopheles at rest during the day in houses in Moïssala, while in Dembo the frequency is equal to *An funestus*. In Moïssala, *An funestus* and *An rufipes* are present in small numbers in the houses but in Dembo, the results of the surveys show the absence of *An rufipes*.

Sporozoite indices of endophilic fauna

Considering the small number of *An funestus* and *An rufipes* captured, all individuals were subjected to the search for plasmodial infection. On the other hand, of the 547 *An gambiae* sp collected, 343 were submitted to Elisa for infection detection and 23 were carriers of Plasmodium sporozoite antigens, representing a sporozoite index of 6.70%. Two out of 10 *An rufipes* were infected with sporozoites, representing a sporozoite index of 20%. None of the 13 individuals in *An funestus* were found infected.

Discussion

Effective malaria control is based on an integrated approach of several strategies, [14, 15, 16]. For vector control, prevention of malaria transmission through the widespread use of insecticide-treated nets and indoor residual spraying is recommended [17]. But first, knowledge of the sensitivity of malaria vector anopheles to insecticides and their biting behaviour is required before measures can be deployed and post-deployment evaluation of these measures [18, 19]. The objective of this study was to assess the impact of the indoor spraying campaign on malaria transmission in the eastern zone of the Moïssala health district after two cycles of bendiocarb spraying. This evaluation was carried out by measuring certain entomological parameters of malaria transmission using methods adapted to the study of vectors.

To better evaluate this campaign, these parameters should have been studied for a year. This would take into account the seasonality of transmission and vector density. For example, the results of our study did not allow us to conclude on the degree of effectiveness of IRS in the short, medium and long term. Moreover, our results also did not allow us to conclude whether the difference in the specific composition between 2015 and 2016 is related to IRS. However, the results obtained could be used as reliable scientific databases for future vector control planning. The IRS coverage rate was over 85%, which reflects a high level of support from the local population. According to WHO, for IRS to have a positive impact on transmission, a coverage rate of 80% must be achieved. In 2015, five species of *Anopheles* were identified in the Moïssala health district (*An gambiae sl*, *An rufipes*, *An nili*, *An pharoensis* and *An ziemanni*), while the 2016 results reported only three species: *An gambiae sl*, *An rufipes* and *An funestus* with a prevalence of *An gambiae sl*. These results are comparable to those obtained by Kerah et al, [7] in Goulmoun, Diarra et al, [8] in Douguia in Chad and Saotoing et al, [20] in Maroua in Cameroon where *An gambiae* predominates with 84.5%, 87.5% and 81.79% respectively during night captures on human bait and in residual morning intradomiciliary fauna. These results could be explained by the similarity of these areas in geoclimatic and ecological terms. The aggressive anophelian density is 9 bites/man/night in Moïssala and zero in Dembo. Moïssala's results are similar to those obtained by Himeidan et al [21] in Sudan, Dossou yovo et al [22] in Bouaké, Ivory coast, who reported aggressiveness of 8.82 bite/man/year in Um Salala and 8.9 bite/man/night respectively.) Dembo's results (0 bites/man/night) would be related to the spray effect. On the other hand, Kerah et al [7] in Goulmoun (Bongor) in Chad reported a significantly higher Anophelian aggressiveness of 67bite/man/night. The density of endophilic fauna is 3.41 anopheles per room in Moïssala and 0.01 anopheles in Dembo. Similar results were reported by Keïta et al, [23] along the Niger River in Mali where densities were low in the sprayed huts and higher in the control huts. The difference in densities between Dembo and Moïssala is believed to be linked to the widespread use of insecticide-treated mosquito nets and, for Dembo, coupled with indoor residual spraying. The entomological comparison of the treated area with the untreated area in this study confirms the results that indoor spraying leads to a decrease in vector density and aggressiveness, with a significant decrease in malaria transmission [24, 25]. This statement, even if it is not shared by some authors [26, 27], is confirmed during this survey and seems relevant for malaria prevention even in areas of permanent transmission. Concerning this parameter, transmission in the control area is about 0.67 infective bites per person per night. However, it is zero in the treated area. The aggressiveness rate of *Anopheles* indoors (53%) is higher than outdoors (47%). This study corroborates those of Kerah and al [57], Doannio et al, [28] in Ivory coast and Coz and Brengues, [29] in Burkina Faso, who observed a rate of aggression of 55.8% respectively, significantly higher inside than outside the home.

In Moïssala, Anophelian aggressiveness starts from 6pm and continues throughout the night both inside and outside the houses. In both cases, a high rate of bite is observed between 8 p.m. and 5 a.m. However, the maximum is obtained inside between 02 a.m and 04 a.m and only decreases significantly after 4 hours. Outside the houses, a high rate was reached earlier, between 10p.m and 11p.m and the second between midnight and 1a.m. This intense activity of anopheles between 8 p.m. and 4 a.m. had already been reported previously in some studies (28, 30) while others place it after the second half of the night

[31]. These results confirm that the aggression cycle has not been altered by the mere use of nets in absence of spraying. The biting rate decreases from 02a.m outside compared to inside. This decrease in aggressiveness would be due to the drop in temperature in December.

Malaria transmission in the Moïssala health district is due to 2 vectors: *An gambiae s/* which has been found infected on human bait inside (7.40%) and outside (7.09%) as well as in morning fauna (6.70%) and *An rufipes*. Out of 10 *An rufipes* captured in the morning fauna and analysed by Elisa CSP, 2 were positive, i.e. 20% of infected animals. This result does not make *An rufipes* a secondary vector [10, 32] but an excellent one to consider in the transmission of malaria to Moïssala. Our results according to which *An rufipes* is captured at rest in the rooms and on human bait are similar to those observed in Dielmo in Senegal [33] and Dori in Burkina Faso [34]. However, investigations incriminating this species in transmission are rare [35]. Although *An funestus* is, with *An arabiensis*, a major vector of malaria in Chad, this species plays no role in the transmission of malaria to Moïssala.

Conclusion

The indoor residual spraying campaign in the eastern zone of the Moïssala health district has led to the collapse of the density and aggressiveness of malaria vectors. However, its evaluation over a short period of time is not sufficient to assess the impact of malaria transmission in this stable and highly endemic malaria zone. The long-term effectiveness of the indoor residual spraying campaign on malaria vectors and transmission will only be assessed after several years of transmission. In addition, the use of mosquito nets and other control measures should be taken into account in this assessment. Although, this intervention was appreciated and won the support of the target population. However, the high cost of the campaign could undermine the sustainability of the strategy, especially since the population's financial income is suffering as the country goes through a period of economic crisis.

Declarations

Consent for publication

Not applicable

Disclosure Statement

No competing financial interests exist

Source of support

This study was carried out with the financial support of the Non-Governmental Organization Médecins Sans Frontière France. We would like to thank the organization very much.

Conflict interest declaration

The authors declare no competing interest

Author's contributions

KHC conceived, analyzed the data and coordinated the study. DKI conceived, conducted the study, analyzed the data and drafted the manuscript. DDA, AMO, MT, ANF, YKE, BAA, DB, MP conducted the study and reviewed the manuscript. All authors read and approved the final version of the manuscript.

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Tables

Table 1. Results of aggressive and residual morning fauna collection

Study sites	Species	PSC %		CAH %		Total
Moïssala	<i>An gambiae sl</i>	547	95.96	295	97.36	842
	<i>An funestus</i>	13	2.28	4	1.32	17
	<i>An rufipes</i>	10	1.75	4	1.32	14
	Total	570		303		873
Dembo	<i>An gambiae sl</i>	1	50	0	0	1
	<i>An funestus</i>	1	50	0	0	1
	Total	2		0		2

PSC = pyrethrum spray catch, CAH = human bait capture

Table 2. Number of species per collection station

		Collection station		Total
		Outdoor	Indoor	
Species	<i>An funestus</i>	4	0	4
	<i>An gambiae sp</i>	134	161	295
	<i>An rufipes</i>	3	1	4
Total		141	162	303

Table 3. Sporozoite indexes inside and outside the rooms

		Elisa CSP		Sporozoite indexes
		Negative	Positive	(%)
Collection station	outdoor	124	10	7.5
	Indoor	149	12	7.5
Total		273	22	

Figures

Figure 1

Location of study sites.

Figure 2

Cycle of time aggressiveness inside and outside the rooms in Moïssala

Figure 3

Hourly infection rates