PRESIDENT'S MALARIA INITIATIVE

Development of a Pilot Dry Season Vector Control Strategy—Final Report

Integrated Vector Management (IVM) Task Order 2

Contract GHA-I-02-04-00007-00

Prepared for:

United States Agency for International Development

Prepared by:

RTI International
3040 Cornwallis Road

Post Office Box 12194
Research Triangle Park, NC 27709-2194

May 2010

UNIVERSITY OF BAMAKO

FACULTY OF MEDICINE, PHARMACY AND ODONTO-STOMATOLOGY

BP: 1805, TEL: (223) 66 98 58 87, FAX: (223) 20 22 47 89.

INTERNATIONAL CENTER FOR EXCELLENCE IN RESEARCH (ICER)

MALARIA RESEARCH AND TRAINING CENTER ENTOMOLOGY (MRTC-E)

PROJECT

DEVELOPMENT OF A PILOT DRY SEASON VECTOR CONTROL STRATEGY

Final Report

May 2010

Contact: Nafomon SOGOBA MSc., PhD

E-mail: nafomon@icermali.org

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ACRONYMS AND ABBREVIATIONS

An.: *Anopheles*

CDC: Center for Disease Control and Prevention

CSP: Circumsporozoite Protein

ELISA: Enzyme Linked Immuno-Sorbent Assay

GM: Geometric Mean

ICER: International Center for Excellence in Research

IRS: Indoor Residual Spray

KD: Knockdown

M: M molecular form

MBR: Man biting rate

MRTC: Malaria Research and Training Center

MS: MS molecular form

NIH: National Institutes of Health

NMCP: National Malaria Control Program

PCR: Polymerase Chain Reaction

PMI: President's Malaria Initiative

PSC: Pyrethrum Spray Catch

RTI: Research Triangle Institute

S: S molecular form

USAID: United-States Agency for International Development

Vs: Versus

WHO: World Health Organization

SUMMARY

In the Sudan savanna of Mali there is marked seasonality in malaria transmission, a consequence of very low densities of mosquito vectors during the dry season, and so control efforts are generally focused on the rainy season. However, there are parts of that environment, such as the riparian parts of the Niger River, where there is perennial breeding of *Anopheles gambiae s.l.* together with dry season malaria transmission. Thus control measures targeting adult mosquitoes during the dry season in these areas are of interest, and may decrease the size of mosquito population and transmission in the subsequent rainy season. This study aims to explore the effectiveness and potential impact of a dry season IRS in hamlets along the Niger River where mosquitoes continue breeding in the dry season and describe ways in which such an approach might limit malaria transmission during the rainy season. Entomological parameters of malaria transmission were monitored using PSC before and after the IRS in 3 sets of hamlet-inland villages in 2008 and 2009, respectively. Mosquito infection rates were assessed by ELISA and the molecular composition by PCR based techniques. Mosquito density and entomological inoculation rate (EIR) in 2009 (after IRS) were lower than that of 2008 (before IRS) in the 3 hamlets under study (Bozokin, Fourda and Somonosso). The geometric mean number of mosquitoes per house during the rainy season showed a reduction of 40.0% in Bozokin [1.5 (0.6—2.4) vs 0.9 (0.3—1.5)]; 8.3% in Fourda [1.2 (0.5—1.8) vs 1.1 (0.6—1.6)], and 33.3% in Somonosso [1.8 (1.2—2.5) vs 1.2 (0.5—1.8)]. The same pattern was observed in EIR, measured as the number of infective bites per person per season, with a reduction rate of 79.4% (0.34 vs 0.07), 36.4% (1.87 vs 1.19) and 42.9% (0.28 vs 0.16) respectively in Bozokin, Fourda and Somonosso. Mosquito density and EIR decreased between 2008 and 2009 in hamlets where the IRS was performed. However excepting in Bozokin, this reduction was < 50%. Additional dry season IRS intervention may be required to observe any significant reduction in malaria transmission in subsequent rainy season.

1. BACKGROUND/JUSTIFICATION

Vector control is one of the major elements of the World Health Organization (WHO) global malaria control strategy. Indoor residual spraying (IRS) is one of the primary vector control interventions for reducing and interrupting malaria transmission. Recent data re-confirms the efficacy and effectiveness of IRS in malaria control in countries where it was implemented well. The primary effects of IRS towards curtailing malaria transmission are: i) to reduce the life span of vector mosquitoes so that they can no longer transmit malaria parasites from one person to another, and ii) to reduce the density of the vector mosquitoes. In some situations, IRS can lead to the elimination of locally important malaria vectors when based on the knowledge of their ecology.

In the Sudan savanna of Mali there is marked seasonality in malaria transmission, a consequence of very low densities of mosquito vectors during the dry season, and so control efforts are generally focused on the rainy season. However, there are parts of that environment, such as the riparian parts of the Niger River, where there is perennial breeding of *Anopheles gambiae s.l.* together with dry season malaria transmission. Thus control measures targeting adult mosquitoes during the dry season in these areas are of interest, and may decrease the size of mosquito population and transmission in the subsequent rainy season in remote inland villages. The aim of this project was to explore the effectiveness and potential impact of a dry season IRS in hamlets along the Niger River where mosquitoes continue breeding in the dry season and describe ways in which such an approach might limit malaria transmission during the rainy season. The development and implementation of this dry season malaria control strategy was jointly initiated by Malaria Research and Training Center (MRTC), the National Institute of Health (NIH) and the Center for Disease Control (CDC), Atlanta with financial support of the Agency for International Development (USAID) under the President's malaria initiative (PMI) and Research Triangle Institute (RTI). This is the final report of the first year of this pilot dry season vector control study.

2. ACTIVITIES/OBJECTIVES

The main activities/objectives accomplished during this period were:

i) Characterize and monitor malaria transmission parameters to provide baseline information on entomological parameters of malaria transmission;

ii) Perform selective IRS during the dry season in hamlets spanning the Niger River between Kirina and Kenieroba,

iii) Assess the impact of IRS on malaria transmission by monitoring entomological parameters of malaria transmission, after performing the selective IRS, in threes set of hamlets and inland villages.

3. METHODS

3.1. STUDY AREA AND PERIOD

The study was conducted in the rural community of Bancoumana, district of Kati, region of Koulikoro from April to December 2009. The study area lies between Kirina and Kenieroba along the Niger River at both sides. The map below shows the study area (**Figure 1**). The IRS was implemented during the dry season (April 2009) and malaria transmission parameters were monitored from May to December 2009 in three sets of hamlet and inland villages:

- Kenieroba and its fihermen's hamlet Fourda at the left side of the Niger River
- Bancoumana and its fishermen hamlet Bozokin at the left side of the Niger River
- Dangassa its fihermen's hamlet Somonosso at the right side of the Niger River

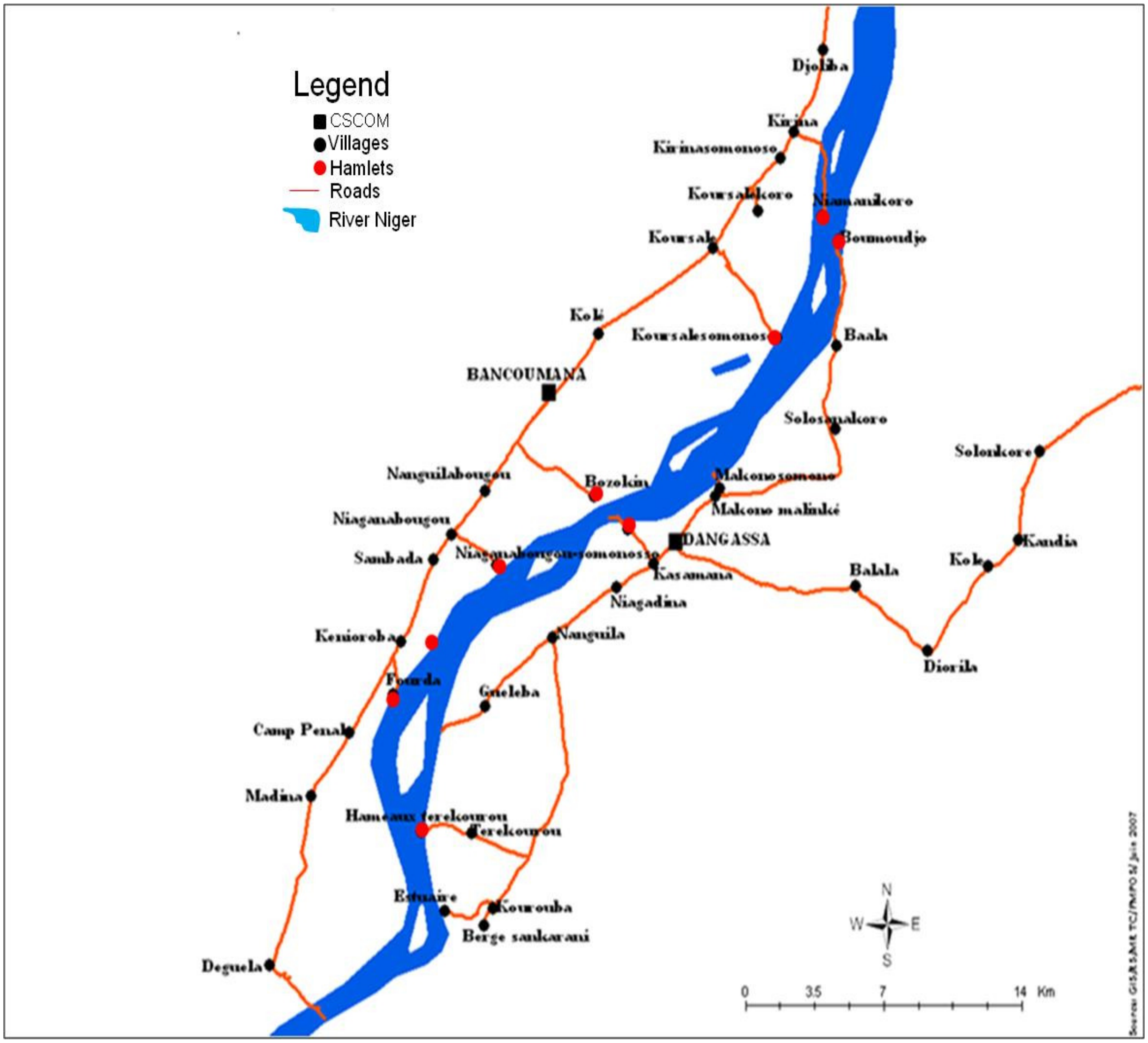

FIGURE 1: Map of the study area showing the Niger river (blue), the fishermen' hamlets (red dots) and the big inland villages (dark dots).

3.2. Population information, mobilization and dwellings census

A field trip was organized by the research team to meet with the population of each locality. When the team arrived in a locality, it first explained to the community leaders (chief and his advisers) the objectives of the study. The meeting was then extended to the families' chiefs and the whole population. After their approval 2-4 younger persons were selected in the population to serve as guides for the research team to get into the different families. The study objectives were again explained to all of the family members present and particularly to ladies who take care to the rooms. The number of all sleeping houses and stores were recorded onto an appropriate sheet.

3.3. Susceptibility tests to insecticide

The Mali National Malaria Control Program (NMCP) with the support of USAID under the PMI is implementing an indoor spraying in two health districts of Mali: Bla and Koulikoro. Lambda-cyhalothrin (25 mg/m2), a pyrethroid, is being used in this IRS campaign. Therefore it was decided to use the same insecticide in this study. Although this insecticide was selected for this campaign after a susceptibility test, it was good to repeat this in our study area before deciding to use it in our IRS. Thus, one locality at both sides of the Niger river in the study area was selected for this test. The test was performed according to the World Health Organization (WHO) standard procedure as follows:

- Wild female mosquitoes were collected from three Fourda and Bozokin at the right side and Dangassa somonosso at the left side of the Niger River and transported to the insectary of MRTC in Bamako for oviposition.
- The F1 female mosquitoes reared from the eggs of the wild female were kept and sugar-fed for 2-5 day-old.
- A maximum of 25 mosquitoes per WHO test tube were exposed to insecticide treated papers. A minimum of three replicates (exposed) and one control (unexposed) using mosquitoes from the same batch were carried out.
- Mosquitoes were exposed to the insecticides for 1 hour and kept in holding containers on 10% sugar solution for 24 hours before total mortality is recorded.
- Mosquitoes were taken to the field to perform the test. The the relative humidity were recorded during the test

3.4. Indoors Residual Spraying

3.4.1. Soak pit and washing area construction

The soak pit and the washing area were constructed in respect to the norms of those constructed by RTI for the current IRS campaign of the PMI project.

3.4.2. Implementation of the IRS

The IRS process was done as follow:

- At least one day before, the population was informed and asked to take out of the houses meals, liquids and all removable materials.
- In the village the spraying team was joined by two village helpers who were introducing the sprayers to each family chief,
- Before spraying the house, permission was obtained from the family chief and the house owner,
- After having obtained the permission the supervisors got into the houses to check if all required materials were removed,
- The spray was then done
- Advices were given to the house owners by the supervisors regarding the time before opening the house after spraying and before cleaning as well as where to put the rubbish.

3.5. Efficacy testing of the insecticide

The lambdacyhalothrin (25 mg/m2), a pyrethroid sold under the label ICON 10CS is the insecticide used for this study. The same insecticide has been used by RTI in their IRS campaign in two districts of Mali. The WHO standard cone test was used to assess the insecticide's biological efficacy. A colony of adult female mosquito of Kisumu strain (2-5 days old) known to be susceptible to the insecticide was used. They were reared in the lab and transported to the field in paper cups held in a container and covered with a wet towel. Mosquitoes were kept to rest 24 hours before starting the test.

Five rooms were randomly selected in each locality for the test. A cone was fixed on each of the four sides of the selected room for the test. Ten mosquitoes were gently introduced in each of these cones so that they are in contact with the wall surface for 30 mn (**Figure 2: a, b, c, d**). A control cone was fixed on a stack of paper on the wall and mosquitoes were exposed to the unsprayed surface for the same time period. During the assay the temperature and the relative humidity in the house were recorded. After the 30 mn of exposition, the mosquitoes were gently extracted from the cones and put back in the cups and the knock-down rate was determined. Mosquitoes were then fed with 5% sugar solution and kept for observation. The mortality rate was established after 24 hours of observation. When the mortality in the control group was between 5-20%, the formula of Abbott was used to correct the mortality rate in the test group. The test was invalid when the mortality rate in the control group was more than 20%.

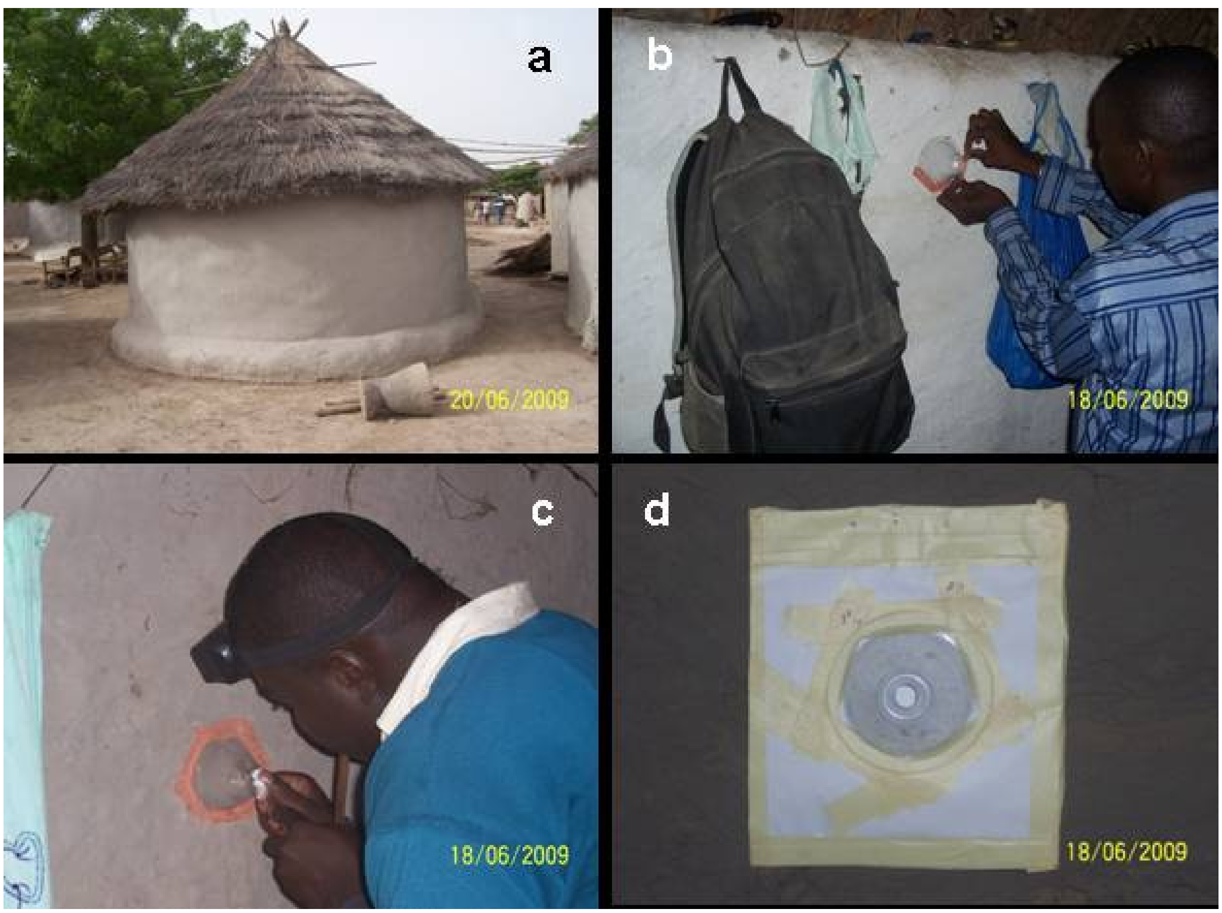

FIGURE 2: Process of the World Health Organization (WHO) standard cone test technique.

3.6. Monitoring of malaria transmission parameters.

To monitor entomological parameters, mosquitoes were collected using pyrethrum spray catches performed at day time in human sleeping houses. A list of all compounds and houses of each locality was established by the research team. The sampling was constrained to respect the proportion of house-roof type (thatch roof vs. metal roof) and the size of the locality. Thus, from this list, 60 houses (one per compound) in each inland village and 10 in each hamlet were randomly selected for mosquito sampling. Collected mosquitoes were morphologically identified, counted by species and recorded onto an appropriate sheet. At the same time, the number of person who slept in the house the previous night and house roof type, were also recorded. Mosquitoes were sorted under a binocular by abdomen stage in unfed, fed, half-gravid and gravid. The total number of mosquito collected in a locality was divided by the number of houses prospected to estimate the mean density of mosquito per house in the given month. The sum of fed and half-gravid was divided by the total number of sleepers multiply by 30 to estimate the man biting rate (MBR) which is the mean number of mosquito bite per person per night in this locality during this month. This was multiply by 30 to have the monthly MBR.

The head-thorax portion of collected mosquito was tested for the presence/absence of malaria parasite by Enzyme Linked Immuno-sorbent Assay (ELISA) method. The infection rate (IR) is the proportion of mosquito having parasite. The entomological inoculation rate was estimated by multiplying the proportion of infected mosquito by the MBR. This represents for a person, the risk of getting infected in a locality at a given time period. The remaining buffer solution of ELISA was used to determine the molecular composition of mosquito population by polymerase chain reaction (PCR) technique.

4. Results

4.1. Population information, mobilization and dwellings census

The population of all the fishing hamlets located at both sides of the Niger River between Kirina (Commune rurale du Mandé) and Kenieroba (Commune rurale de Bancoumana), expressed their willingness and approval to participate in the study. The number of houses obtained during the dwellings census is shown in **Table 1.** We can see from this table that it was in Fourda, Bozokin and Dangassa Somonosso, where the entomological parameters are being monitored, that we had the highest number of houses.

TABLE 1: Number of houses recorded per village.

Localities	**Number of houses in place**	**Number of houses in construction**	**TOTAL**
Fourda	130	30	160
Belenidankan	38	2	40
Bozokin	167	2	169
Niaganabougou -hamlet	47	2	49
Koursalé-somonosso	32	0	32
Niamanicoro	13	0	13
Dangassa-somonosso	156	11	167
Boumoudjou	167	7	174
Terekoudou	17	0	17
TOTAL	**767**	**54**	**821**

4.2. SUSCEPTIBILITY TESTS

Figures 3 and **4** present the knockdown and mortality rate of 3 days old F1 generation of mosquito collected in the different localities before the performance of the IRS.

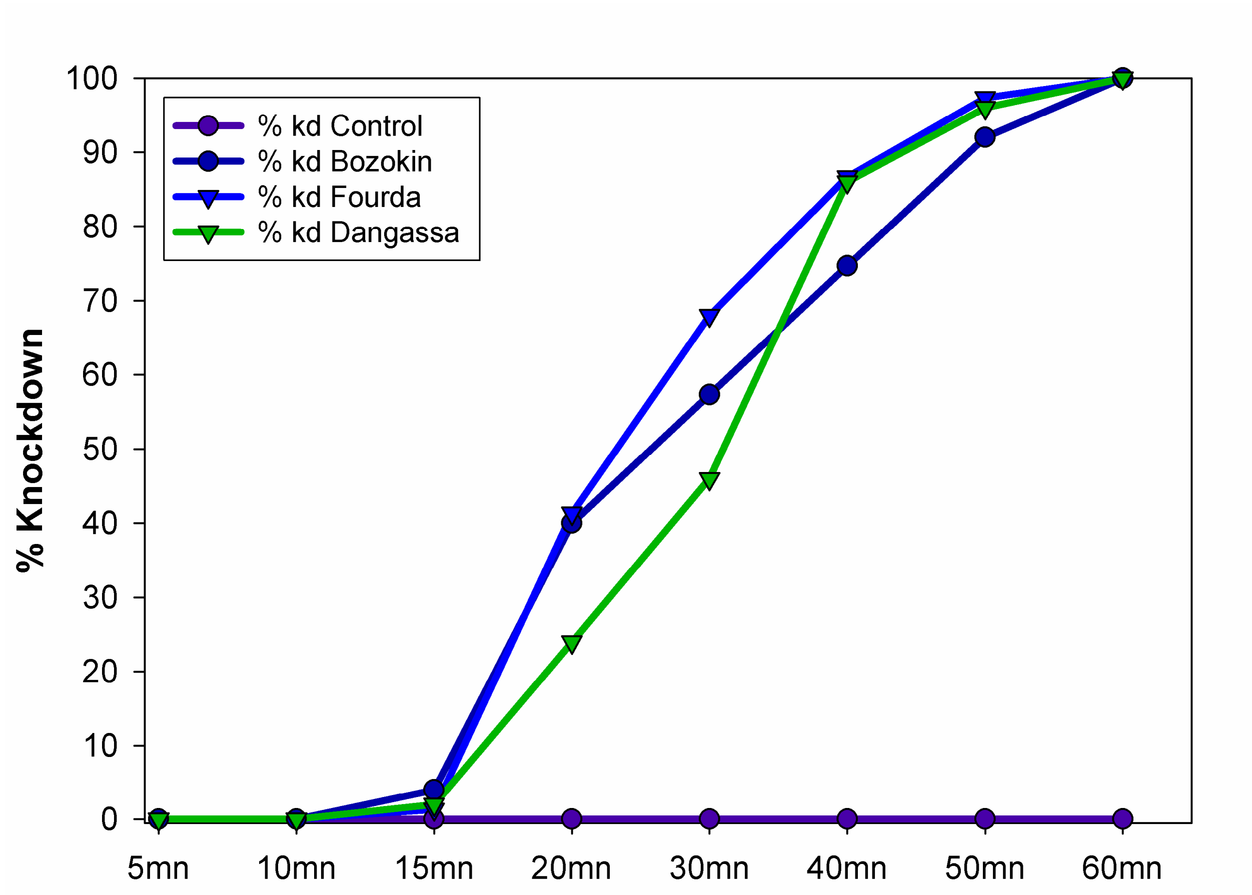

FIGURE 3: Knock-down (kd) time period for *An. gambiae s.l.* to lamdacyhalothrine in Bozokin, Fourda and Dangassa in April 2009 before performing the IRS

The 50% of mosquitoes were knocked down before 25 mn post exposition in Fourda, just after 25 mn and 30 mn respectively in Bozokin and Dangassa Somonosso. 95% of the exposed mosquitoes were knocked down after 45 mn in Danagassa and around 60 mn in Bozokin and Fourda.

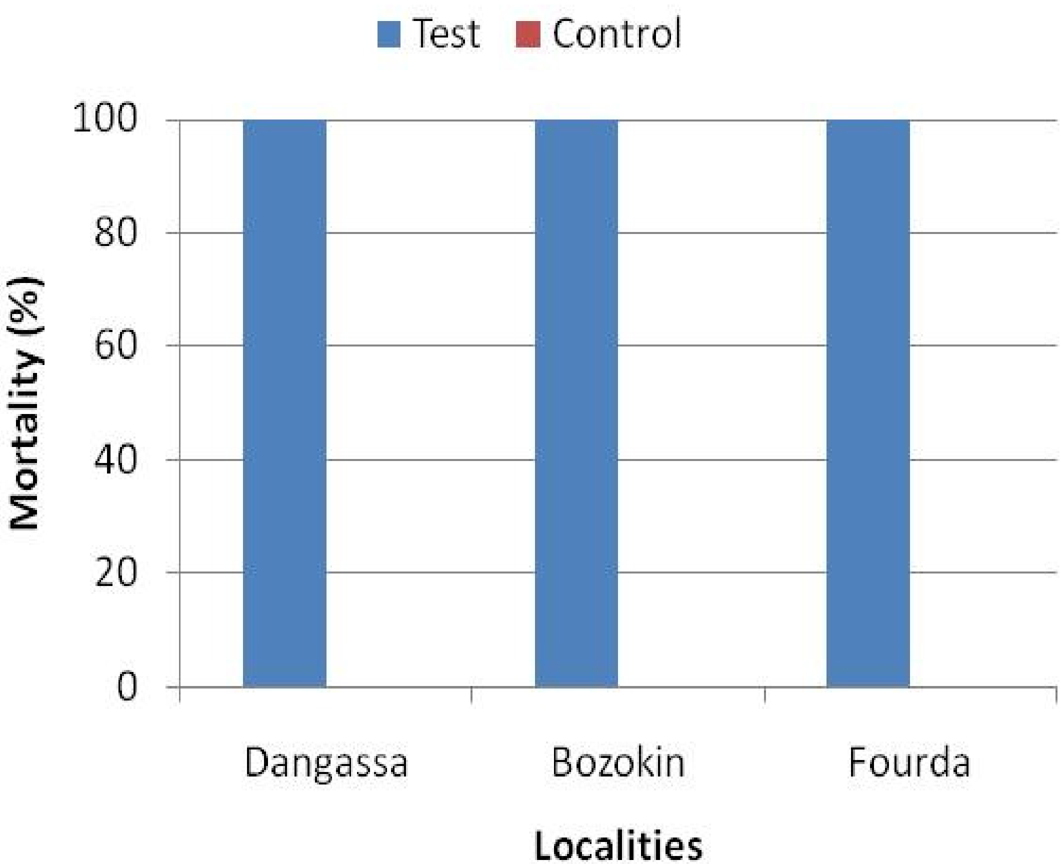

FIGURE 4: Overall mortality rate of *An. gambiae s.l.* 24 hours after exposition to the insecticide in Dangassa, Bozokin and Fourda

At 24 hours after the exposition of mosquitoes to the insecticide, the mortality rate was 100% in all localities attesting that the population of *An. gambiae s.l.* was susceptible to the lamdacyhalotrine in this area according to WHO standard procedure for insecticide resistance.

4.3. Performing the IRS

4.3.1. Soak pit and washing area construction

The spraying process started with the construction of the soak pit and the washing area (**Figure 5**). It is located in the courtyard of the CSCOM of Bancoumana. Thus it is secured from children and animals access.

FIGURE 5: The soak pit and the washing area located in the courtyard of the community health center (CSCOM) of Bancoumana.

4.3.1. The IRS implementation

To perform the IRS, 10 sprayers trained by RTI from Koulikoro district were hired. They were supervised by members of the research team. The IRS was performed as describe in section 3.4.2. **Tables 1** and **2** present the percentage of houses and the number and type of population covered in each hamlet.

TABLE 2: Coverage rate of the IRS in the different localities, April 2009.

Localities	Number of compounds	Number of houses sprayed	Number of houses not sprayed	Coverage rate
Fourda	14	114	4	96.61
Belenidankan	4	25	3	89.28
Bozokin	18	119	21	85.00
Niaganabougou -hamlets	5	37	12	75.51
Koursalé-somonosso	1	22	2	91.66
Niamanicoro	8	21	0	100.00
Dangassa-somonosso	10	140	22	86.41
Boumoudjou	21	122	30	80.26
Terekoudou	1	15	0	100.00
TOTAL	82	615	94	86.74

The overall coverage rate of the houses was 86.74% with a maximum of 100% in Terekoudou (15/15) and Niamanicoro (21/21), and a minimum of 75.51% (37/49) in Niaganabougou hamlet.

TABLE 3: Structure of the covered population in the different localities.

Localities	**Other inhabitants**	**Number of children < 5 years**	**Number of pregnant women**
Fourda	237	54	6
Belenidankan	60	12	2
Bozokin	311	74	8
Niaganabougou hamlet	112	26	4
Koursale somonosso	30	2	2
Niamanicoro	53	14	2
Dangassa Somonosso	281	75	11
Boumoudjou	289	62	9
Terekoudou	35	5	1
TOTAL	**1408**	**324**	**45**

***NB**: The indirect coverage will be at least 15,000 inhabitants when we consider the inland big villages like Koursale, Bancoumana, Kenieroba and Dangassa.*

Table 3 presents the number of children < 5 years old, pregnant women and other inhabitants recorded in the sprayed houses per village. The spraying directly covered around 2,000 inhabitants among which we have 324 children less than 5 years old and 45 pregnant women.

4.4. EFFICACY TESTING OF THE INSECTICIDE

The knockdown (after 60 mn) and mortality (after 24 hours) rates of the Kisumu Anopheles strain (known susceptible) by cone test just after the IRS for each locality are shown in **Tables 4** and **5.**

TABLE 4: The knockdown and mortality rate of the Kisumu Anopheles strain after exposition to the sprayed wall one month after the IRS in selected houses of the different localities.

Localities	Rates (%)	Rooms					TOTAL
		1	**2**	**3**	**4**	**5**	
Bozokin	Mortality	90.0	70.0	90.0	100.0	100.0	90.0
	Knockdown	67.5	26.7	92.5	50.0	100.0	67.3
Fourda	Mortality	100.0	90.1	100.0	95.0	100.0	97.0
	Knockdown	97.5	100.0	100.0	97.5	92.5	97.5
Dangassa Somonosso	Mortality	72.5	77.5	100.0	90.0	96.7	87.3
	Knockdown	82.5	65.0	100.0	96.7	86.7	86.2

These results show that an overall mortality rate > 80% was observed in all localities indicating that the spraying was well done and the treated surface is killing mosquitoes. Seven month after the IRS, the efficacy test of the insecticide was replicated by the same team in the same selected houses in the different localities. The results of this test are shown in Table xxx.

TABLE 5: The knockdown and mortality rate of the Kisumu Anopheles strain after exposition to the sprayed wall seven months after the IRS in selected houses of the different localities.

Localities	Rates (%)	Rooms					TOTAL
		1	**2**	**3**	**4**	**5**	
Bozokin	Mortality	37.5	16.7	55.0	23.3	36.7	33.8
	Knockdown	25.0	6.7	60.0	30.0	33.3	31.0
Fourda	Mortality	20.0	27.5	23.3	20.0	0.0	18.2
	Knockdown	2.5	30.0	66.7	32.5	0.0	26.3
Dangassa Somonosso	Mortality	2.5	0.0	10.0	10.0	10.0	6.0
	Knockdown	0.0	0.0	20.0	6.7	0.0	5.3

There was a drastic reduction in the insecticide efficacy in both the knock down and mortality rates. This reduction was 53.9% (67.3% vs 31.0%) in Bozokin, 73.0% (97.5% vs 26.3%) in Fourda, 93.8% (86.2% vs 5.3%) in Dangassa Somonosso for the knockdown and 62.4% (90.0% vs 33.8%) in Bozokin, 81.2% (97.0% vs 18.2%) in Fourda and 93.2% (87.3% vs 6.0%) in Dangassa Somonosso.

4.3. MALARIA TRANSMISSION PARAMETER

4.3.1. VECTOR POPULATION COMPOSITION

Over the study period, about 99.7% of collected mosquitoes were *An. gambiae s.l.* and only 0.3% were *An. funestus* (n = 16093).

4.3.2. MOLECULAR STRUCTURE OF AN. GAMBIAE S.L.

Figure 6 shows variation in the molecular composition of *An. gambiae s.l.* population during the rainy and dry seasons

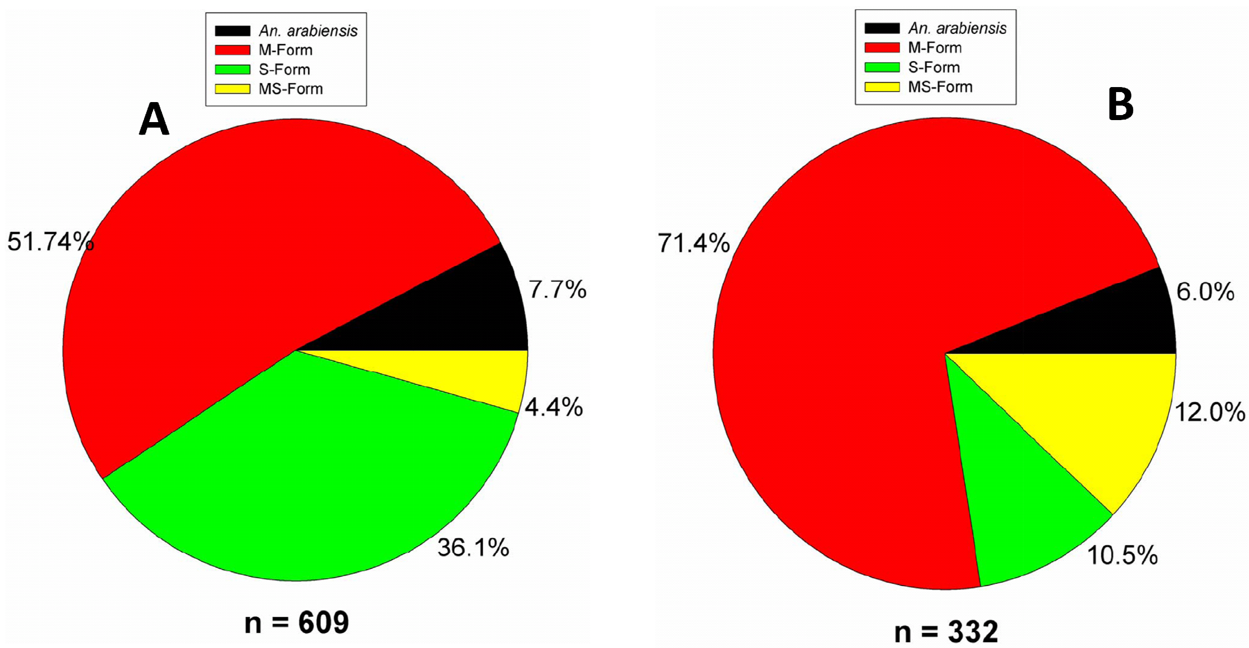

FIGURE 6: Molecular composition of *An. gambiae s.l.* population during the rainy (**A**) and dry (**B**) seasons in the study area.

The S molecular form frequency was high in the rainy season compared to the dry season (36.1% vs 10.5%). In the opposite, M and MS molecular forms frequencies increased during the dry season (71.4% vs 51.7% and 12.0% vs 4.4%, respectively). *An. arabiensis* species was presents in all seasons at a stable and low frequency (7.7% vs 6.0%).

4.3.3. MOSQUITO DENSITY, INFECTION RATE AND ENTOMOLOGICAL INOCULATION RATE

Figure 7, 8 and **9** showed the temporal variation in mosquito density per house and the entomological inoculation rate (EIR) in fishermen 'hamlets (IRS performed) and their respective inland villages (no IRS) in 2008 (before IRS) and 2009 (after IRS).

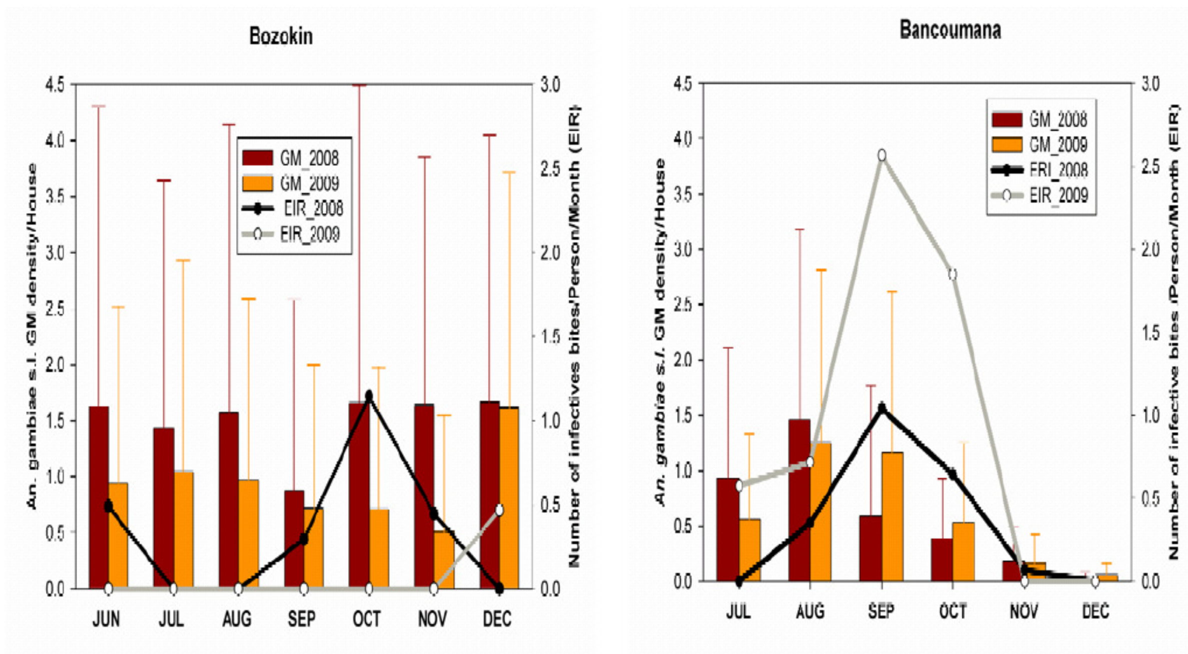

FIGURE 7: Variations in the geometric mean number of A*n. gambiae s.l.* per house per month (bars, error bars = 95% ci) and the mean number of infective bites per person per month (lines with 1 se) in Bozokin and Bancoumana during the rainy seasons of 2008 (before IRS) and 2009 (after IRS)

The geometric mean number of mosquitoes per house during the rainy season of 2009 (after IRS) showed a reduction of 40.0% compared to 2008 (before IRS) in Bozokin [1.5 (0.6—2.4) vs 0.9 (0.3—1.5)]. The EIR measured as the number of infective bites per person per season showed the same pattern with a reduction of 79.4% in 2008 compared to 2009 (0.34 vs 0.07), 36.4% (1.87 vs 1.19). In it remote big village of Bancoumana, the GM number of mosquito per house was lower at the beginning of the rainy season of 2009 compared to 2008. However the EIR per month was consistently higher in 2009 than in 2008.

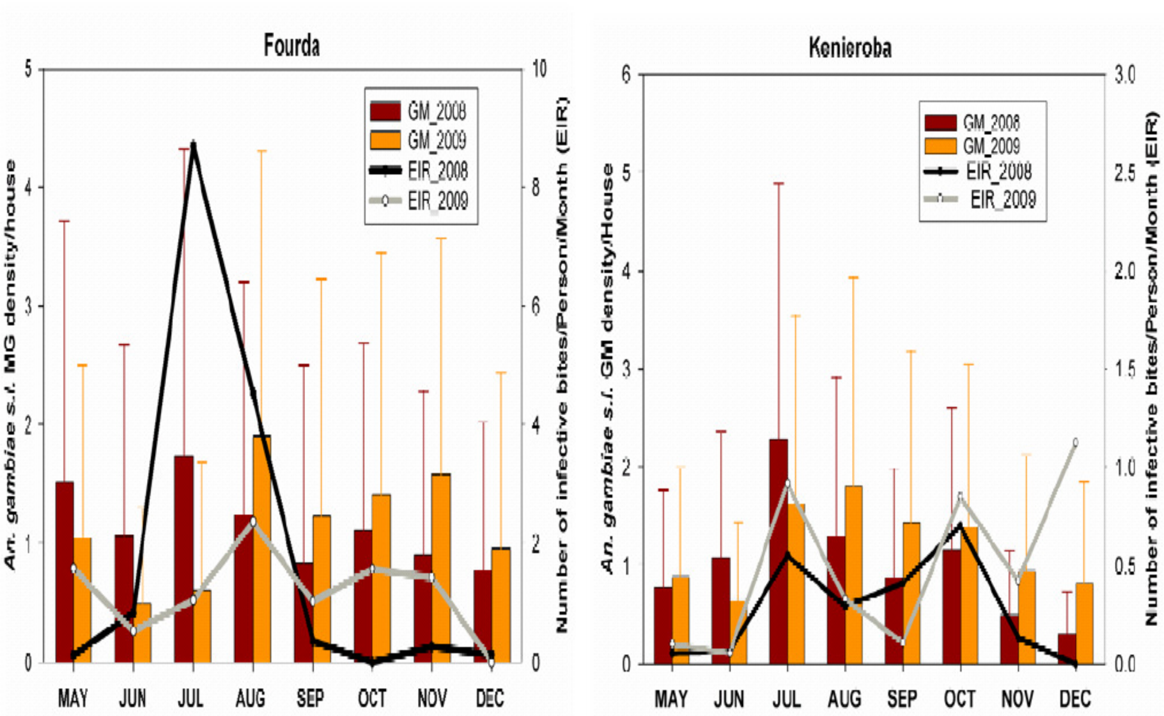

FIGURE 8: Variations in the geometric mean number of *An. gambiae s.l.* per house per month (bars, error bars = 95% ci) and the mean number of infective bites per person per month (lines with 1 se) in Fourda and Kenieroba during the rainy seasons of 2008 (before IRS) and 2009 (after IRS).

Mosquito density and entomological inoculation rate (EIR) in 2009 (after IRS) were lower than that of 2008 (before IRS) in Fourda with a reduction rate of 8.3% in the geometric number of mosquito per house in 2009 compared to 2008 [1.2 (0.5—1.8) vs 1.1 (0.6—1.6)], and 36.4% in the number of infective bites per person per season (1.87 vs 1.19). Any pattern of reduction was observed in it remote big village of Kenieroba, where the GM number of mosquito per house per month were even higher, from August to December, in 2009 than in 2008.

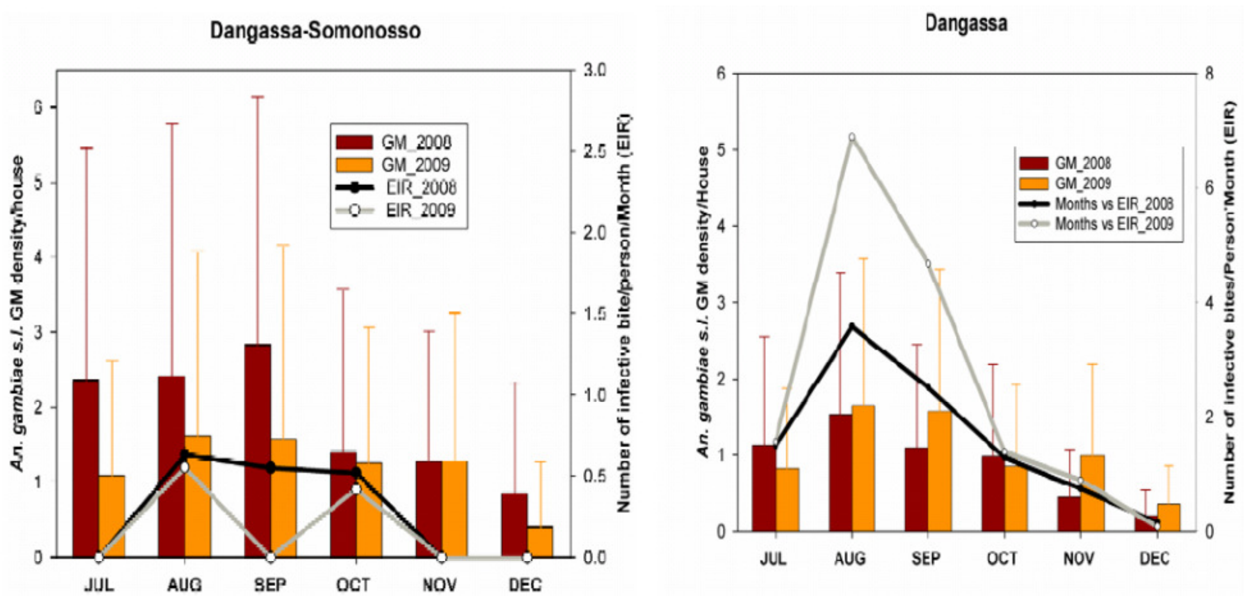

FIGURE 9: Variations in the geometric mean number of *An. gambiae s.l.* densities (bars, error bars = 95% ci) and entomological inoculation rate (lines with 1 se) in Somonosso and Dangassa before (year 2008) and after (year 2009) performing an IRS during the dry season in Somonosso.

In the fishermen hamlet of Dangassa (Somonosso), the GM number of mosquito per house as well as in the mean number of infective bites person per rainy season was lower in 2009 than 2008. The reduction rate 33.3% in 2009 compared to 2008 [1.8 (1.2—2.5) vs 1.2 (0.5—1.8)] in the GM density and 42.9% in the mean number of infective bites (0.28 vs 0.16). In Dangassa, its remote big village, no trend of reduction was observed either in the GM number of mosquito and the mean number of infective bites.

5. Support to MRTC research effort

This project financially contributed to the research efforts of MRTC by supporting the field activities of the implementation of the IRS and mosquito density monitoring as well as salaries of two field and one lab technicians for one year.

6. Discussion/comments

The objective of this pilot study was to explore the effectiveness and potential impact of a dry season IRS in hamlets along the Niger River where mosquitoes continue breeding in the dry season and describe ways in which such an approach might limit malaria transmission during the rainy season in remote big villages. The IRS was performed in respect to all the environmental requirements of RTI and PMI. An overall coverage rate of 86.7% was observed attesting the acceptance and participation of the population. This coverage rate is above of what is recommended by WHO (xxxxx). *An. gambiae s.l.* was the most prevalent malaria vector (99.7%) in the study area. It was composed of 7.1% of *An. arabiensis*, 51.7% of M molecular from, 27.1% of S form and 7.1 of MS form (n = 939). The insecticide susceptibility test performed before the IRS showed that they were fully susceptible to the lamdacyhalotrine which was used in the IRS. Insecticide efficacy test using WHO standard cone test one month after the IRS showed that it was well performed. However, six months after performing the IRS, the insecticide efficacy test showed a drastic decrease in the bio-efficacy of the insecticide (62.4% to 93.2%). Is it due to the high temperature observed at the time we performed the IRS (April)? Or to the very wet situation along the river when it is flooded? This situation needs to be investigated.

No significant reduction in mosquito densities and EIR was observed in any of the sprayed hamlet and no clear pattern in term of reduction was observed in the remote big villages. The non significant decrease in the sprayed hamlets may be due to the drastic decrease in the bio-efficacy of the insecticide mentioned above. The same phenomenon, in addition to the fact that the IRS has not been performed before the mango rain, can explain the situation observed in the remote big village because mosquitoes may have had time to migrate before the performing of the IRS in the hamlets. It is also important to note that no one may expect a significant reduction in malaria transmission after only one round of spraying. Therefore two to three consecutive IRS at the appropriate time is required for a significant impact the hamlets, hence in the remote big villages.

7. Future perspectives

For the future we recommend

- to replicate the study two more times with the IRS performed on the required time,
- to make a new design of the study with a control instead of pre and post test assessment of the impact of the IRS. This will avoid the effect of confounding factors such the year to year variation in malaria transmission, subsequent to the year to year variations in the rainfall in the Soudan savanna areas.
- to follow the insecticide bio-efficacy test at basis of every two months with a particular attention to the environmental factors.