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Impacts of rice production on the incidence of mosquitoes and malaria transmission in the district of Malanville, Northen Benin

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Abstract

To evaluate the impact of rice production on malaria transmission in the district of Malanville in northern Benin, an entomological study was carried out from January to December 2021. Therefore, human landing catches (HLC) activities were conducted over two consecutive nights in 6 random houses selected from each study site for adult mosquito collection monthly. Additionally, indoor pyrethrum spray catches (PSC) were done in 6 additional houses at each study site. This scheme of mosquito sampling was the same each month during the study period. Female mosquitoes collected by HLC particularly the Head-thoraces of these mosquitoes were tested for the presence of *circumsporozoite protein* (CSP). Mosquitoes collected by PSC were used for species identification based on the Polymerase chain reaction (PCR) technic. Results from this study showed a total of 63,012 female mosquitoes were caught from the two methods whereas 58,285 were by HLC. *Plasmodium falciparum* was mainly transmitted by *Anopheles gambiae s.s and Anopheles arabiensis where* malaria transmission was high from June to November during the rainy season and declined during the dry season (December-May). The average entomological inoculation rate (EIR) was significantly higher during the rainy season compared to the dry season (p < 0.05). These findings showed that rice production increased mosquito fauna but doesn't have a significant impact on malaria transmission. Therefore, communities living close to rice production areas will permanently be exposed to mosquito bites throughout the year.

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Introduction

In 2019, according to World Health Organization (WHO) estimation, 216 million cases of malaria occurred worldwide where most malaria cases were in the WHO African Region (90%), followed by the WHO South-East Asia Region (7%) and the WHO Eastern Mediterranean Region(2%) (WHO, 2016). At the same time, Africa's demography is rapidly changing with an increasing number of people moving to urban (Bloom et al., 2003; Yousif HM, 2005; Yousif HM, 2006). Based on the United Population Fund (UNFPA), Nations Africa's population is the fastest growing in the world (WHO, 2016). Population growth has resulted in increased demand for food supply. A solution to the problem has been the development of agriculture. This explosive rate of population growth requires a great increase in food production (Potts et al., 2013). The severe pressure to feed the growing population has led many African governments to look for better methods of expanding agriculture production, such as irrigation. For this reason, many countries in sub-Saharan Africa have developed irrigation projects to provide food security, improved diet and increased income for a rapidly growing population. Unfortunately, water-related projects may aggravate the problem of mosquito-borne disease by increasing the number of larval habitats and extending the duration of the transmission season.

In Benin, a country in West Africa, in order to meet food security and poverty alleviation, irrigation agriculture is rapidly becoming a major economic activity in the cities. In general, non-used spaces (marshland, road edges, beaches, etc) are turned into gardens where rice is cultivated. The production has been intensified by the development of large marshy areas, particularly the one located at Malanville, north of Benin to increase the yield. The activity is believed to be directly responsible for the creation of almost 1,000 jobs thereby addressing urban demand for food and unemployment.

The advantages of rice cultivation are considerable. They contribute to the improvement of living conditions by supplying food, income and employment to urban populations.

The economic and social impact of rice production in this area is however limited by the presence of mosquito breeding sites (Yadouleton *et al.*, 2009). In fact, agronomic practices in rice production areas create numerous trenches that retain rain and irrigation water. These stagnant bodies of water provide suitable breeding grounds for mosquitoes, particularly *Anopheles gambiae*, the main vector of malaria in Africa. The farmers also dig wells that are used for water irrigation.

The wells are favorable breeding habitats places for An. gambiae. Furthermore, used containers at the sites gather bodies of water and become ideal sites for egg-laying by the malaria-transmitting mosquitoes, and this certainly will increase the incidence of malaria in this area which represents a serious issue for public health. One concern is that irrigating agriculture, particularly rice production, promoted to increase food security and alleviate poverty might increase the urban malaria risk by creating breeding sites for the Anopheles vector with potentially higher epidemiological risk of malaria in urban than rural areas (Matthys et al., 2006; Klinkenberg et al., 2008) In Benin, there are few data available that investigate the association between malaria transmission and rice production. The present study was conducted in the biggest site of rice production at Malanville in northern Benin with the aim of investigating the entomological aspects of malaria transmission in relation to seasonal variation of malaria vector populations in this area of Benin.

Materials and methods

Study area

The study was conducted at Malanville, in Benin, from January to December 2021. The rice production farm is located in the far north of Benin near the Niger River at11°52N and 3°23E in a highly populated zone (Fig. 1). The farm is 100 hectares in size and shared between 10 local cooperatives of approximately 1,000 farmers.



Fig. 1. Map of Benin showing the study site.

This northern part (Malanville) of Benin is characterized by a Sudanian climate (semiarid) with only one rainy season per year (main annual rainfall = 900mm). Agricultural practices in this farm create numerous trenches that retain rain and water from irrigation systems which represented suitable breeding sites for mosquitoes, particularly *Anopheles gambiae*, the main malaria vector in the areas.

Mosquitoes collection

From January to December 2021, mosquito catches were carried out monthly for two consecutive nights per month in houses located close to the farm following the scheme below:

(1) Human Landing Catches (HLC) Indoor and outdoor in the 4 selected houses and performed monthly over two consecutive nights (9:00 PM to 5:00 AM); (2) Indoor Pyrethrum Spray Catches (PSC) done in 4 other selected compounds. For better results interpretation, the same compounds in each sampling method were maintained throughout the study. Moreover, for HLC activities, some people called " catchers" were trained to catch landing mosquitoes prior to blood feeding using mouth aspirators prior in order to minimize the risk of malaria transmission. They gave prior informed consent and received anti-malaria prophylaxis and yellow fever immunization. The sporozoite infection rate was evaluated by using the *Anopheles gambiae* s.l populations collected by HLC. The total mosquitoes collected by HLC and PSC were kept -20°C for further analysis.

Mosquito identification

A random sample of 100 females of *An. gambiae* collected by PSC were used for species identification based on Polymerase chain reaction (PCR) technic.

Laboratory processing of mosquitoes: Circumsporozoite enzyme-linked immunosorbent assay

The head-thoraces of these females captured from the human landing catches were homogenized in 250 μ l of grinding buffer using a glass pestle. *Circumsporozoite* (CS) protein micro-plate ELISA using 50 μ l/well of the homogenate was done in 96-well microtitre plates coated with anti- *P. falciparum* monoclonal antibodies at 22–25°C for 30 min (Wirtz *et al.,* 1987) monoclonal antibody (MoAb) horseradish peroxidase conjugate incubated for 1h was used to detect Captured CS antigen.

The addition of ABTS [2,2-azino-di-(3ethylbenzthiazoline- 6-sulphonate)] substrate gave a green colour reaction for positive results which were read by visual assessment of the colour reactions, and OD was measured within 30 min using a spectrophotometer (Multiskan Ascent, Model 354; ThermoLabsystems, Finland) at 414 nm. Sample positivity was determined by titration of PfCSPpositive control antigen using cut-off OD values equivalent to 12pg of PfCSP or 50 sporozoites(Wirtz *et al.*, 1987).

Entomological parameters

Three entomological indicators of malaria parasite transmission intensity at the sites were calculated to evaluate the impacts of rice cultivation on malaria transmission malaria: the human biting rate (HBR), which represents the number of mosquitoes biting a person during a given time (bites/p/t) (time being night, month, or year) (Yadouleton *et al.*, 2010); the CSP rate is the proportion of mosquitoes found with Plasmodium falciparum CSP over the total number of mosquitoes tested (Yadouleton *et al.*, 2010); the Entomological Inoculation Rate (EIR) was calculated as the product of the HBR by the CSP rate (Yadouleton *et al.*, 2010).

Data analysis

An analysis of variance (ANOVA) was performed to compare the entomological estimates (HBRs, EIRs) between the seasons in this district.

Ethical considerations

Ethical authorization was given by the Ethical Committee of the Ministry of Health in Benin. However, the consent form was signed by the head of each household for the spray catches HLC activities.

Results

Species diversity and density

Mosquito fauna composition

A total number of 63,012 mosquitoes were collected from January to December 2021.

Among these populations of mosquitoes, 58,285 were collected by HLC and the remaining from PSC where *An. gambiae* (15.68 %) and *Culex quinquefasciatus* (82.1 3%) (Table 1) represented the most abundant species caught.

The proportion (15.73 %) of malaria vectors (*Anopheles gambiae* and *Anopheles funestus*) contributed certainly to malaria transmission.

Species	HLC	PSC	Total	%
Aedes aegypti	75	112	187	0.3
Aedes sp	25	36	61	0.1
Anopheles funestus	21	12	33	0.05
Anopheles gambiae	9,154	713	9,867	15.68
Anopheles pharoensis	112	86	198	0.31
Anopheles ziemanni	345	212	557	0.9
Culex quinquefasciatus	48,342	3,412	51,754	82.13
Culex tigripes	103	59	162	0.25
Culex Sp	108	85	193	0.31
Total	58,285	4,727	63,012	100

Table 1. Mosquito species caught from January to December 2021 in the study area.

Species of Anopheles gambiae.

Mosquitoes from PSC were analysed by PCR for identification of sibling species among *An. gambiae s.l.* complex. The PCR anaylisis of the species revealed the presence of two sub-species of *An. gambiae: An. gambiae s.s.*, and *Anopheles arabiensis* with a predominance of *An. gambiae s.s.* (95%).

Seasonal abundance and biting rates

Results from this study show that the highest bites of *An. gambiae* s.l. was found in July during the rainy seasons (196.5 bites/p/n) and the lowest in May (0.81 bites/p/n) during the dry season (Fig. 2). The average of bites/person/night during the rainy season

(108.138 bites/p/n) was 24 times significantly higher than those obtained during the dry season (4.491 bites/p/n) (p < 0.05).

Vectors infection to CSP and malaria transmission risk

9,154 mosquitoes caught from the HLC, were assayed									
by ELISA.	Resul	ts from this an	alysis show that	a few					
(1.8%)	of	Anopheles	(163/9,154)	were					

circumsporozoite protein positive for *Plasmodium* spp at the study site. In this part of the north of the country, transmission occurred during the rainy season (June to November). The average of EIRs was 10.58 times significantly higher in the rainy season (1.8 infective bites /person/night) than what was obtained during the dry season (0.17 infective bite /person/night). Data from the study showed that the annual EIRs is 10.18 (Table 2).

Table 2. Seasonal variation of EIR from January to December 2021 at Malanville.

Entomological parameters	Jan	Feb	Mar	Apr	May	June	Jul	Aug	Sept	Oct	Nov	Dec	Total
Total of Mosquitoes Collected by HLC	25	22	19	15	13	1815	3144	1719	1139	834	321	88	9,154
Number of bites/person/night	1.56	1.38	1.19	0.94	0.82	113.44	196.5	107.44	71.19	52.13	20.06	5.5	572.125
positive for <i>P. falciparum</i>	3	2	2	1	1	34	65	21	15	9	6	4	163
circumsporozoite proteins													
Proportion of circumsporozoite	0.120	0.090	0.105	0.670	0.077	0.188	0.020	0.012	0.013	0.011	0.019	0.045	0.6
Entomologic inoculation rate.	0.188	0.125	0.125	0.063	0.063	2.212	4.062	1.312	0.937	0.562	0.375	0.250	10.19

Discussion

The findings from the present study showed clear evidence of the dynamics of malaria transmission in rice production areas. The abundance and fluctuations in larval and adult densities of samples collected were inherent to the environmental and ecological characteristics related to rice cultivation (Jacob BG et al., 2005). Our work has shown with regard to the mosquito species identification in the district of Malanville that Anopheles coulluzzi is the main specie found and is responsible for malaria transmission. This result confirms several reports on malaria transmission in West Africa on the involvement of An. gambiae coullizi as the main vector (Yadouleton et al., 2010, Romoli ., Gendrin M. 2018.). On other hand, the presence of An. Arabiensis among the species of anopheles identified seems that this vector plays a part in malaria transmission in this district. This was confirmed by the reports of malaria transmission in many African countries (Mwangangi et al., 2013; Degefa et al. 2017). However, the predominance of Anopheles coulluzzi (95%) in the study area is consistent with its distribution throughout West Africa as the main vector of malaria transmission (Yadouleton et al., 2010; Gnanguenon V et al., 2014). The presence of higher biting rates and sporozoite infective Anopheles

(Table 1) in households from January to December showed that malaria parasite transmission was permanent during the year and was reinforced by the presence of breeding sites, permanent pools and puddles maintained during watering of rice crops. Reports from many scientists on rice cultivation in sub-Saharan Africa confirmed that the activity is exposed to greater malaria risk as well as more

mosquitoes (Yadouleton et al., 2010).

In fact, malaria epidemics were reported very important in many areas where there is rice production (Dolo *et al.*, 2014) . In Mali and Côte d'ivoire, the vectorial capacity of *Anopheles gambiae* and *arabiensis* Patton was shown to be 150 times higher in the rice field area compared to the cottongrowing area (Briët O *et al.*, 2003; Ijumba, Lindsay; 2001). Consequently, it has been postulated that the impact of establishing new rice irrigation schemes on malaria might be less problematic in areas of high and stable malaria transmission.

The economic advantage of rice production and its contribution to malaria transmission are two major challenges in sub-Saharan Africa for its development, there is an urgent need to improve methods for growing rice without producing mosquitoes.



Fig. 2. Seasonal abundance and biting rates of An. gambiae from January-December 2021.

The social and economic development, often induced by increased rice production might be, at least partially, invested in antimalarial measures (e.g., purchase of insecticide-treated bed-nets), and hence contribute to a decline in malaria prevalence⁵.

However, the creation of new rice fields in areas where malaria transmission is low may lead to epidemics and pose a great toll on primary health care systems, as observed, for instance, in a newly developed swamp.

Moreover, this study showed an increased level of the entomological inoculation rate (EIR) during the rainy season. The increased level of EIR in malariaendemic countries is not synonymous with a higher incidence of malaria due to the infected bites' saturation. Indeed, a similar study conducted in Côte d'Ivoire has shown that the rainy season increases the density of mosquito fauna without influencing the annual incidence of malaria⁶. These authors conclude that the increased number of mosquitoes associated with irrigation does not necessarily lead to an increased level of malaria transmission.

These findings showed that people living close to rice production area farms are more exposed to malaria whereas the risk is less for those living far from this activity.

Conclusion

These findings showed that rice production increased mosquito fauna but doesn't have a significant impact on malaria transmission. Therefore, people living close to rice production areas are more exposed to mosquito bites throughout the year. It is, therefore, crucial to rethink rice cultivation which would minimize the development of mosquito breeding sites and consequently malaria transmission.

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