Contribution of two sibling species of *Anopheles gambiae* complex living in sympatry, *An. coluzzii* and *An. gambiae*, to malaria transmission in Alibori and Donga region, Northern Benin, West Africa

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INTRODUCTION

Anopheles gambiae s.l. is a complex of 8 species of mosquitoes, some of which highly contribute to malaria transmission whereas others play a minor role. Among these species, *An. gambiae s.s.* is becoming an extremely effective malaria vector, especially in tropical Africa. Based on molecular evidence, *An. gambiae s.s*. is comprised of 2 sibling species, *An coluzzii* and *An. gambiae*. The main goal of this study was to assess the contribution of each of the two sibling species present and living in sympatry in a region selected for Indoor Residual Spraying (IRS) on malaria transmission. The goal of this study was to measure and compare IRS-related entomological measures of transmission for each species prior to the implementation of IRS.

METHODS AND MATERIALS

Human landing capture (HLC) organized at night from 09 PM to 06 AM and pyrethrum spray capture (PSC) from 06 to 09 AM were used inside and outside of households to sample vector populations during the rainy (June-October) and dry (November-May) seasons in 6 districts targeted for eventual IRS implementation. Captured *An. gambiae s.l.* mosquitoes were:

- dissected to assess parity;
- tested to estimate sporozoite antigen positivity; and
- identified to species by molecular methods (PCR).
- used to estimate the human blood meal index, which represents the proportion of blood meals derived from humans by mosquito vectors used to estimate human biting habit;

In addition, PCR was used to:

- estimate the frequency of the Knock-down resistance (Kdr) L1014F mutation in the sodium channel, which is associated with resistance to pyrethroid insecticides; and
- estimate the frequency of the Ace-1R mutation, which is associated with carbamate and organophosphate insecticides

In the study, malaria transmission in a region is expressed in terms of Entomological Inoculation Rate (EIR) (ma x s) due to both two species. Then, the contribution of each species is the EIR calculated for the species divided by those of the 2 species x 100.

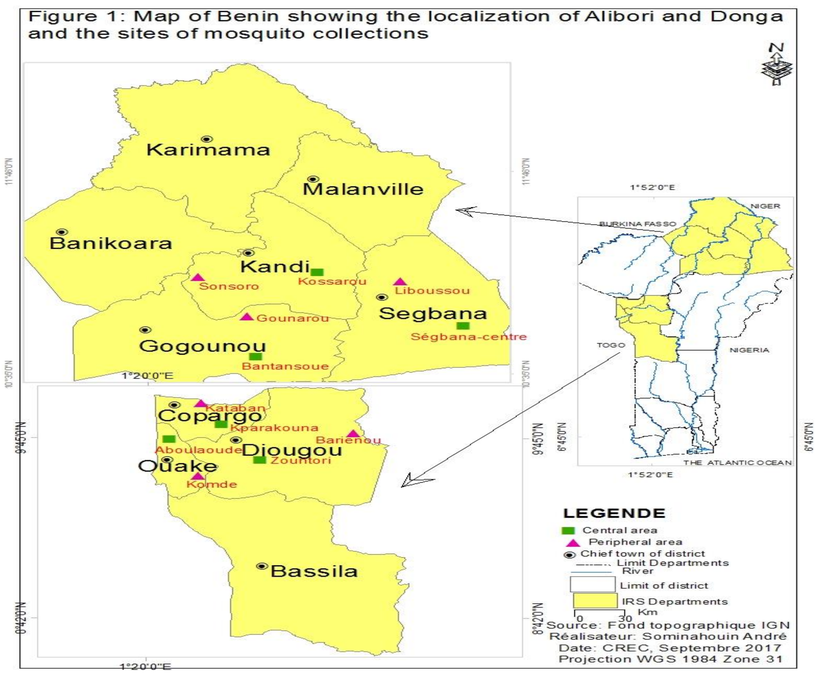


Figure 1: Map showing the study sites in Alibori and Donga regions of Benin where mosquito capture occurred.

RESULTS

Figure 2: Distribution of the frequency of captured *Anopheles gambiae and Anopheles coluzzii* during the dry and rainy seasons in Alibori and Donga regions in Northern Benin

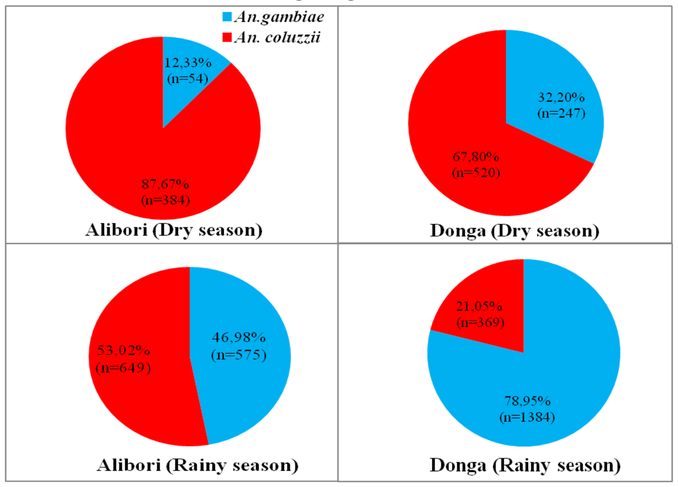
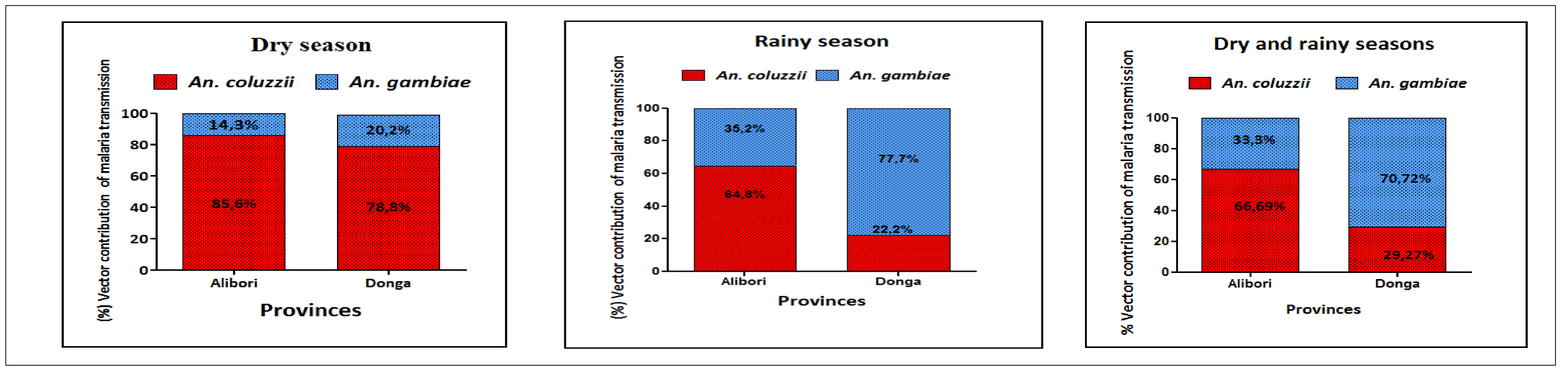


Table 1: Distribution of Knock-down resistance (Kdr) and Ace-1R frequencies in *Anopheles gambiae and Anopheles coluzzi* in Alibori and Donga regions in Northern Benin

Location	Species	Number tested	RR*	RS*	SS*	F*(Kdr)	P-value	RR*	RS*	SS*	F*(Ace 1)	P-value
Alibori	*An.gambiae*	170	123	34	13	0.82	.002	0	10	160	0.03	.138
	An. coluzzii	271	143	90	38	0.69		0	7	264	0.01	
Donga	An.gambiae	387	272	93	22	0.82	.153	0	20	367	0.03	.298
	An. coluzzii	152	102	34	16	0.78		0	4	148	0.01	
Total	An.gambiae	557	395	127	35	0.82	.003	0	30	527	0.03	.048
	An. coluzzii	423	245	124	54	0.73		0	11	412	0.01	

*Kdr homozygote resistant RR=X, Kdr heterozygote resistant RS=Y, Homozygote sensible SS=Z, F=XX

Figure 3: Percent contribution of *Anopheles gambiae and Anopheles coluzzii* to malaria transmission measured by EIR by season in Alibori and Donga regions in Northern Benin



DISCUSSION

From 1,302 *An. gambiae* collected by HLC in Alibori and Donga, 57.7% (751/1,302) were collected indoor against 42.3% (551/1,302) outdoor. The same trend was registered for *An. coluzzii*: 51.6% indoor (575/1114) against 48.4% outdoor (539/1114).

Same and high blood feeding rates were observed for the two species collected in bedrooms by PSC: 97.96 % (627/640) and 97.36 % (407/418) respectively for *An. gambiae* and *An. coluzzii* showing a high anthropophilic behavior of the two species.

However, *An. coluzzii* contributes to 85.6% and 78.8% of EIR in Alibori and Donga, respectively, during the dry season compared to *An. gambiae* that contributes to only 14.3% and 20.2%, respectively. This proportion was inversed in Donga during the rainy season *(An. gambiae* contributed to 77.7% compared to 22.2% for *An. coluzzii*). *An. coluzzii* in Alibori had a lower frequency of Kdr L1014F, which suggests that IRS will be more efficacious at killing *An. coluzzii* than *An. gambiae*.

CONCLUSION

Aopheles coluzzii and *An. gambiae* are the two vectors involved in malaria transmission in Alibori and Donga with a high abundance of *An. coluzzii* during the dry season, particularly in the Alibori region, due to a higher dryness associated with permanent Anopheles larvae breeding places created by the presence of rice areas and permanent rivers.

Despite the same rate of anthropophily, the same blood meal index, similar indoor and outdoor biting rates, and the same sporozoite rates, *An. coluzzii* contributes more to malaria transmission in our study areas. The lower frequency of Kdr L1014F in *An. coluzzii* could favor the impact of Long Lasting Insecticidal nets (LLIN) and result in better control of malaria in Alibori compared to Donga. But, concerning IRS, the use of pirimiphos methyl (organophosphate) like the previous years in Benin, we should expect the same impact in the two regions because of the very low level (0.01-0.03) of *Ace-1* mutation.

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