Indoor Resting by Outdoor Biting Females of *Anopheles gambiae* Complex (Diptera: Culicidae) in the Sahel of Northern Senegal

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ABSTRACT Three villages in the Senegal River basin were selected to study the biting and resting behavior of *Anopheles gambiae* s.l. in relation to human habits, rainfalls, and rice culture irrigation. All inhabitants sleep outside throughout the year, mainly under poor quality bednets. Mosquitoes were collected host-seeking during the night on human bait outside and resting during the day inside and outside in pit shelters. *An. gambiae* s.s. and *An. arabiensis* fed mainly outside, the only place where hosts are available; fed and gravid females resting indoors fed outside. The proportions of *An. gambiae* s.s. and *An. arabiensis* in outdoor biting catches and in indoor spray catches were not significantly different, but they differed from year to year with the latter sampling method. *An. gambiae* s.s. predominated in 1990, a more wet and humid year, whereas *An. arabiensis* was more common in 1991, which was an arid year. Both species are highly endophilic in this arid area where outdoor-resting places are limited.

KEY WORDS *Anopheles gambiae* s.s., *Anopheles arabiensis*, biting, resting behavior, Senegal

TWO DAMS HAVE been built on the Senegal River, one upstream at Manantali (Mali) to create a reservoir and the other downstream at Diama (Senegal) to prevent the inflow of saltwater during the dry season. Irrigated development schemes have been established along the river and ecological changes are expected. The impact of development on human health, especially malaria, was studied previously in Senegal (Faye et al. 1993a, b).

Malaria is endemic in this area and transmission takes place during the rainy season and the beginning of the dry season. *Anopheles gambiae* s.s. and *An. arabiensis* are the primary vectors in this sahelian area where transmission is unstable. Considerable heterogeneity has been documented in their host selection and resting pattern in subSaharan Africa. *An. arabiensis* is generally more zoophilic and exophilic than *An. gambiae* (Mnzava et al. 1995), although both are considered endophilic and both bite humans indiscriminately indoors and outdoors. Resting habits have been linked to some extent to chromosomal inversion polymorphisms (Coluzzi et al. 1979); and in Nigeria, the genetic variants adapted to the arid climate were prone to bite and rest indoors (Coluzzi 1984).

In this study, which was carried out as a part of a longitudinal malaria survey in the Diomandou rice irrigation scheme, exophagic and endophilic females of *An. gambiae* s.l. are reported. This behavior is of practical importance because it makes them vulnerable to insecticide house spraying or impregnated materials.

Materials and Methods

Study Area. Field work was undertaken in 3 villages of the Podor District in the Senegal River basin. The climate of this area is sahelian, with rain falling on <20 d from late July to early October. Total annual rainfall was 158 mm in 1990 and 152 mm in 1994, but only 95 mm fell in 1991 at the Ndioum Station which is 10 km from the study area. Vegetation is sparse, with few trees in and around villages. The mean minimum and maximum temperatures are >20 and >30°C during the cool season, respectively, and >24 and >35°C during the warm season from March to October.

The 3 villages selected for study were Diomandou Walo, Diomandou Dieri, and Toulde Galle,

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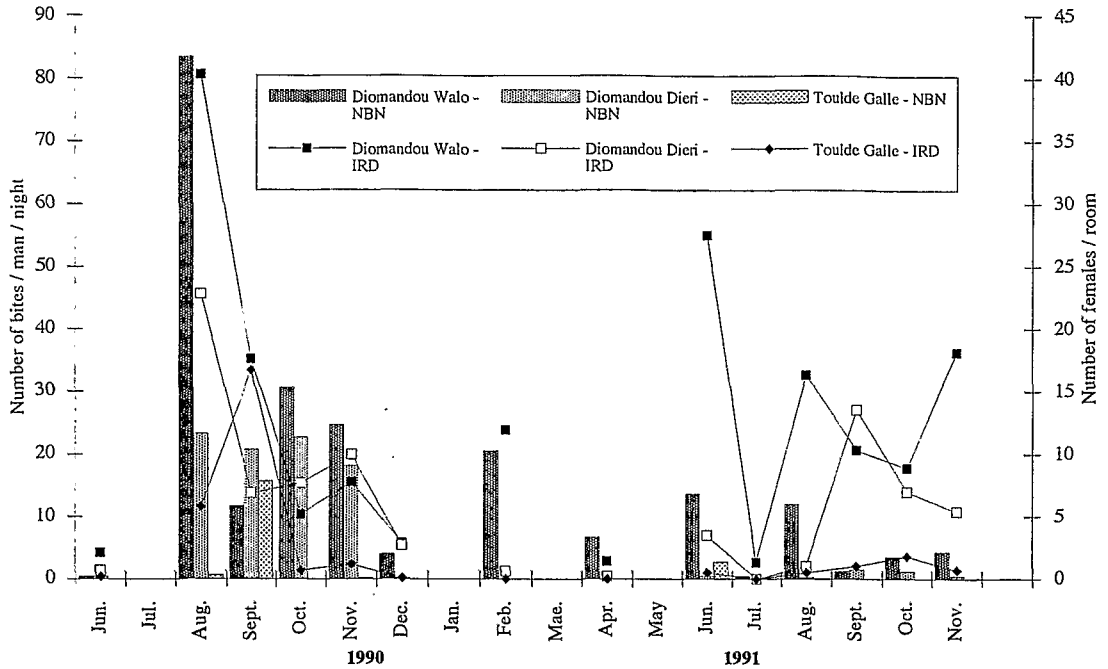

Fig. 1. Numbers of *An. gambiae s.l.* biting outdoors per human per night (NBN) and resting indoors (IRD) per room.

with 143, 809, and 600 inhabitants living in 42, 234, and 160 rooms, respectively. The first 2 villages are within the irrigated zone, whereas the 3rd village is 5 km outside the irrigated zone. The houses have mud walls with grass thatch or corrugated iron roofs. The number of rooms per house varies from 1 to 7, according to family size. Residents spend the night outside in the courtyard or under verandas throughout the year, even during the cold season. Most adults use bednets, which have limited protective effect because generally they are poorly maintained. Some children and adolescents do not use bednets. Domestic animals including cows, horses, donkeys, sheep, and goats usually are penned in open enclosures near rooms, but roofed cow sheds were present at a few houses.

Survey Methods. Anophelines were sampled using the following 3 methods: (1) All night catches outside on 4 human baits (protected by chemoprophylaxis) to estimate the number of anopheline bites per human per night. Two collections from 2100 to 0700 hours were made each month in each village during the rainy season. Few *An. gambiae* s.l. females can be collected before 2100 hours in this area. (2) Diurnal pyrethrum spray collections were made in the same 10 representative rooms in each village, twice per month to estimate the indoor resting density as the number of female anophelines resting per room. Catches were made in the morning from 0800 to 1000 hours after the overnight bait catches. (3) Diurnal outside resting collections were made in 2 Muirhead Thompson pit shelters in each village twice per month during the rainy season.

Species of the *An. gambiae* complex were identified by examining polytene chromosomes of the ovarian nurse cells of half-gravid resting females caught during 1990 and 1991 (Coluzzi et al. 1979). In 1994, the polymerase chain reaction method was used to identify biting as well as resting *An. gambiae* s.l. females caught in August, using thorax, abdomens, legs, or wings preserved dry (Paskewitz and Collins 1990).

Blood-engorged females caught resting indoors were tested by enzyme-linked immunosorbent assays to determine the source of bloodmeal (Beier et al. 1988).

Results and Discussion

Composition of the Anopheline Population. With the exception of a few *Anopheles pharoensis* Theobald and *An. rufipes* Gough, most anophelines (>95%) belonged to the *An. gambiae* complex Giles, represented by *An. gambiae* s.s. and *An. arabiensis* Patton. The relative proportions of members of the *An. gambiae* s.l. determined in each year were as follows: 1990, *An. gambiae* s.s. 67% (95% C L, : 53–81%) and *An. arabiensis* 33% (19–47%) of 42 identified females; 1991, 36% (23–49%) and 64% (51–77%) of 56 females; and 1994, 59% (51–67%), and 41% (33–49%) of 141 specimens. *An. gambiae* s.s. was more common than *An. arabiensis* in 1990 and 1994, but *An. arabiensis* was most prevalent in 1991. No significant differ-

Table 1. Number of mosquito blood meals (*n*) and human blood index (HBI) of *An. gambiae* s.l. females in the 3 villages

Month	Diomandou Walo		Diomandou Dieri		Toulde Galle	
	n	HBI	*n*	HBI	*n*	HBI
Aug. 1990	69	0.27	78	0.45	18	0.67
Sept. 1990	41	0.34	16	0.5	43	0.77
Total	110	0.30	94	0.46	61	0.74
Sept. 1991	29	0.93	44	0.77	3	1
Oct. 1991	19	0.15	13	0.77	9	0.78
Total	48	0.62	57	0.77	12	0.83

n, Number reacting.

ence was observed in *An. gambiae* s.s. frequencies between 1990 and 1994 ($\chi^2 = 0.84$, df = 1, $P > 0.30$). In 1994, *An. gambiae* s.s. frequencies in biting catches (52%, $n = 60$) and in resting catches (64%, $n = 81$) were not significantly differrent ($\chi^2 = 2.24$, df = 1, $P > 0.10$). These findings confirmed previous observations that *An. gambiae* s.s. was present in this part of the Senegal River Valley and that the proportion of *An. gambiae* s.s. increased with rainfall (Coz 1973, Petrarca et al. 1987).

Population Dynamics. Only 1 fed and 7 gravid *An. gambiae* s.l. females were collected in the pit shelters in 1991 and 1992, and these collections were discontinued in 1994.

Anopheles gambiae s.l. was collected only in the rainy season at Toulde Galle, but its collection extended into the dry season in the 2 villages within the irrigation zone (Fig. 1). Irrigation is an important environmental modification in dry savannas, because it extends the temporal availability of breeding places.

The average annual number of *An. gambiae* s.l. was higher from June 1990 to April 1991than from June 1991 to April 1992 (Fig. 1). The decrease was more marked for outdoor biting females (4.2-fold in Diomandou Walo, 16.8-fold in Diomandou Dieri, and 3.8-fold in Toulde Galle).

Feeding Habits. Table 1 shows the human blood index of indoor resting females of *An. gambiae* s.l. in August–September 1990 and in September–October 1991 for each village. The human blood index in Toulde Galle was significantly higher than those reported in the 2 villages within the irrigation zone in 1990 ($\chi^2 = 23.84$, df = 1, $P < 0.001$), but the proportion of *An. gambiae* s.l. females that fed on humans was comparable among the 3 villages in 1991 ($\chi^2 = 3.67$, df = 2, $P > 0.10$).

Among 8 *An. gambiae* s.s. and 9 *An. arabiensis* specifically identified of 73 blood-engorged females collected in 1994 collection, 5 and 4 had fed on humans, respectively, and 3 each on cattle; 1 *An. gambiae* s.s. fed on sheep, and 1 *An. arabiensis* on a horse.

In an urban area of Senegal (Pikine), where domestic animals are rare, *An. arabiensis* blood feeds almost exclusively on humans (Vercruysse and Jancloes 1981), in contrast to other areas of Senegal where the presence of domestic animals deviates ≥30% of *An. gambiae* s.l. to cattle (Vercruysse 1985; Konate et al. 1994; Faye et al. 1995a, b).

Data on human blood indices from indoor samples of resting *An. arabiensis* populations studied in different parts of Africa and reported by Sharp and Le Sueur (1991) indicated that the majority of specimens (>80%) had fed on humans. However, White et al. (1972) reported a lower overall human blood index for *An. arabiensis*, which showed an inverse relationship between the proportion of human blood meals and the number of cattle present.

Comparison of Outdoor Biting and Indoor Resting Anophelines. The human biting habit of *An. gambiae* s.l. depends on host availability. In the study area where blood meals were only available outdoors, all anophelines were exophagic. Therefore, it was tempting to try to determine what proportion rested indoors.

Fed and gravid *An. gambiae* s.l. females comprised a higher proportion of indoor resting collections in 1990 than in 1991 for both Diomandou Walo ($\chi^2 = 107.19$, df = 1, $P < 0.001$) and Diomandou Dieri ($\chi^2 = 7.58$, df = 1, $P < 0.01$). At Toulde Galle, relative proportions of fed and gravid females of *An. gambiae* s.l. were comparable in 1990 and 1991 ($\chi^2 = 0.004$, df = 1, $P > 0.90$). The fed to gravid ratio in the indoor-resting collections ranged from 0.9 to 1.9, indicating that most of the blood-fed females rested indoors long enough to become gravid (Table 2).

The lack of indoor resting unfed females indicated that after oviposition, most females refed the same night and then rested indoors. Although pit shelters used for sampling the outdoor resting population collected few *An. gambiae* s.l., these females were all blood fed or gravid, indicating that

Table 2. Percentage of *An. gambiae* s.l. females in different gonotrophic states in indoor-resting collections at the 3 villages

States	Diomandou Walo				Diomandou Dieri				Toulde Galle			
	1990		1991		1990		1991		1990		1991	
	n	%	*n*	%	*n*	%	*n*	%	*n*	%	*n*	%
Unfed	91	9	366	26	53	8	46	13	5	2	1	2
Fed (F)	584	59	479	34	294	46	158	47	130	52	28	59
Gravid (G)	310	31	556	39	287	45	126	38	115	46	18	38
F/G	1.9		0.9		1		1.2		1.1		1.5	

Table 3. Mean number of *An. gambiae* s.l. females biting humans outdoors and resting indoors

No. and indexes	Diomandou Walo		Diomandou Dieri		Toulde Galle	
	1990	1991	1990	1991	1990	1991
TCHB	731	137	343	15	67	11
NBN	21.5	5.1	10.1	0.6	1.9	0.5
TCIR	985	1,401	634	330	250	47
IRD	10.5	15.2	6.5	5.2	2.8	0.7
HBI	0.39	0.53	0.46	0.67	0.73	0.83
IRDH/NBN	0.2	1.6	0.3	5.7	0.9	1.2

TCHB, Total females caught at human baits; NBN, number of bites per human per night; TCIR, total females collected resting indoors; IRD, number of indoor resting females per room; HBI, human blood index; IRDH/NBN, females fed on humans outdoors and resting indoors ratio.

females must blood feed shortly after emergence or rest outdoors at sites near larval habitats.

Because *An. gambiae* s.l. females collected at human bait in 1990 and 1991 were not identified specifically, we could not assess the proportion of *An. gambiae* s.s. and *An. arabiensis* resting indoors using the number of bites per night and the indoor resting density estimates of abundance. Posted data were used to calculate the ratio of *An. gambiae* s.l. females that fed on humans outdoors (number of bites per night) and rested indoors (indoor resting density) (Table 3).

The ratio of human blood fed house resting to human biting densities is only indicative of the resting behavior of *An. gambiae* s.l. females. In the irrigation zone, this ratio was lower in 1990 than in 1991. A possible explanation for the differences in ratio in 1990, despite a higher number of total females caught at human bait, was that more females rested outdoors in 1990 (a wetter and more humid year which provided more outdoor resting places for mosquitoes).

The number of bites per night probably was overestimated because collectors captured both landing and biting mosquitoes, and because the children (30% of the population in the study area) generally received fewer bites than the adult mosquito catchers (Carnevale et al. 1987).

In the study area, *An. gambiae* s.s. and *An. arabiensis* were exophagic and strongly endophilic. Such behavior was reported for *An. rufipes* in Senegal (Faye et al. 1993a, 1995a, b; Konate et al. 1994) and for other *Anopheles* (e.g., *An. sergenti* Theobald, *An. claviger* Meigen, *An. pulcherrimus* Theobald, and *An. sacharovi* Favre) in dry areas (Farid 1956, Gramiccia 1956, Zahar 1973). This behavior, although suspect, has never been documented for *An. gambiae* s.l.

It has been recognized that malaria control by insecticide spraying of houses is more successful with endophilic than exophilic species. A method of vector control that offers an alternative to house spraying is the pyrethroid impregnation of bednets. The large use of bednets by householders and the effectiveness of impregnated bednets for *An. gambiae* s.l. (as observed in other countries and in a small trial in eastern Senegal, Faye 1996) suggest that this control measure also could be effective in northern Senegal.

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