

Similar feeding preferences of *Anopheles gambiae* and *A. arabiensis* in Senegal

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Abstract

This study in Senegal compared the feeding preferences of *Anopheles gambiae* and *A. arabiensis* while controlling for equal accessibility to hosts located outdoors under bed net traps. All fed *A. gambiae* complex females were identified with the aid of the polymerase chain reaction and their blood meal sources were identified by enzyme-linked immunosorbent assay. A total of 605 anophelines, including 281 *A. gambiae* and 301 *A. arabiensis*, were captured, 32.2% in the human-baited traps and 67.8% in bovine-baited traps. 30.3% of *A. gambiae* fed in the former and 69.7% fed in the latter; the corresponding figures for *A. arabiensis* were 29.6% and 70.4%. Thus, when the hosts were located outdoors and made equally available, the feeding preferences of *A. gambiae* and *A. arabiensis* were similar ($P=0.81$). These results suggest that biases existed in previous studies, most of which suggested that *A. arabiensis* was more zoophilic than *A. gambiae*. Alternatively, the feeding behaviour of these 2 species may differ in various parts of Africa.

Keywords: malaria, *Anopheles gambiae*, *Anopheles arabiensis*, blood meals, feeding preferences, Senegal

Introduction

Critical evaluation of the intensity of human-vector contact is essential for the effective control of malaria. The vectorial capacity of a vector population depends on this contact and in particular on the human blood index (HBI), the percentage of mosquitoes biting humans (GARRETT-JONES, 1964). Accurate assessment of the HBI is difficult as it depends not only on the feeding preferences of each species but also on the mosquito sampling techniques and accessibility of the different potential hosts (KAY *et al.*, 1979). HBI calculation is generally based only on the indoor resting mosquito captures, excluding possible exophilic mosquitoes feeding on humans indoors.

Many studies of the feeding preferences of *Anopheles gambiae* and *A. arabiensis*, the main malaria vectors in Africa, have shown that their preferential hosts were humans or cattle. More rarely, females bite horses, donkeys, sheep, goats and sometimes even birds (WHITE *et al.*, 1972; GITHEKO *et al.*, 1994). Most of the studies published suggested that *A. gambiae* is more anthropophilic than *A. arabiensis* (see COZ, 1973; WHITE & ROSEN, 1973; MOLINEAUX & GRAMICCIA, 1980; COLUZZI, 1984; GILLIES & COETZEE, 1987; PETRARCA *et al.*, 1991; COSTANTINI *et al.*, 1996).

In contrast, our previous studies of captured indoor resting mosquitoes in Senegal suggested a similar HBI for both *A. gambiae* and *A. arabiensis* (see FONTENILLE *et al.*, 1997b; LEMASSON *et al.*, 1997). Such observations could be biased if *A. gambiae* and *A. arabiensis* are not equally endophilic. The objective of the present study, therefore, was to compare the feeding preferences of *A. gambiae* with those of *A. arabiensis*, when the hosts, humans or cattle, are equally accessible and in outdoor locations.

Material and Methods

Study site

The study was conducted in Dielmo (13°45'N, 16°25'W), a village in the region of Saloum, Senegal. The village location close to a river permits the persistence of anopheline larvae throughout the year. The rainy season lasts from June to October and an average of 700 mm of rain per year has been recorded in the last 20 years. The domestic animals found at night within the village are cattle, goats, sheep, horses, donkeys, dogs, cats and poultry. Studies of malaria transmission have been carried out in this village since 1990 (FONTENILLE *et al.*, 1997a).

Mosquito collection

From July to September 1997, collections of anopheline vectors were carried out during 2 or 3 consecutive nights every 2 weeks. Mosquitoes were captured using 3 identical rectangular bed net traps (length 2.25 m, width 1.70 m, height 1.70 m). These 3 traps were set up outdoors, positioned so that each trap formed the corner of an equilateral triangle with 5 m sides. The base of each bed net was raised 20 cm above the ground to allow mosquitoes to fly in when searching for blood meals. One bed net contained a 73 kg bovine as bait, in a small enclosure. A 67 kg human volunteer (male) slept in the second bed net, while the third served as a control and was without bait. Bait were rotated among the 3 traps every night. Each session started at 21:00 and ended at 06:00 the next morning. Female mosquitoes entering the traps were collected at 01:00, 03:00 and 06:00 with an aspirator. Anopheline mosquitoes were separated from other Culicidae and morphologically identified. Specimens belonging to the *A. gambiae* complex were further identified with the aid of the polymerase chain reaction technique described by SCOTT *et al.* (1993).

The blood meal of each identified mosquito was smeared on to a Whatman filter paper, and its source was identified using the enzyme-linked immunosorbent assay described by BEIER *et al.* (1988). This method allowed the identification of blood meals taken from humans, cattle, sheep, horses, donkeys or chickens.

Results

During 16 nights, 605 anopheline malaria vectors were captured in the baited traps, 32.2% in the human-baited traps and 67.8% in those with bovine bait (Table). The anopheline vector species caught were *A. funestus*, *A. gambiae*, *A. arabiensis* and *A. melas*.

Of the 281 *A. gambiae* captured, 31.0% were found in the human-baited traps and 69.0% in the bovine-baited traps; 31.9% of the 301 *A. arabiensis* were captured in the human-baited traps and 68.1% in those with bovine bait. These proportions are not significantly different ($\chi^2=0.06$, $P=0.81$).

The source of all blood meals was identified (Table). All were either human or bovine. There were some mixed blood meals, primarily human-bovine, but also 2 bovine-sheep and one bovine-horse (which are classified as 'bovine' in the Table). The proportions of mixed meals were 2.1% (6/281) for *A. gambiae* and 6.3% (19/301) for *A. arabiensis* (Table). This difference was significant ($\chi^2=6.17$, $P=0.013$).

In the human-baited traps, 4 *A. gambiae* and 8 *A. arabiensis* had only bovine blood meals, indicating that the meal had been taken before the mosquito was trapped. Similarly, 4 *A. gambiae* and 9 *A. arabiensis* captured in

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Table. Numbers of *Anopheles* which had fed on human or bovine hosts in bed net traps in Dielmo, Senegal

<i>Anopheles</i> spp.	Total no. of mosquitoes	No. fed	Trap with human host Blood meal source			Estimated no. ^b	No. fed	Trap with bovine host Blood meal source			Estimated no. ^b	Control trap No. fed
			Human ^a	Bovine	Mixed			Human	Bovine ^c	Mixed		
<i>A. funestus</i>	15	11	11(0)	0	0	11	4	0	4(0)	0	4	0
<i>A. gambiae</i>	281	87	82(4)	4	1	79(30.3%)	194	4	186 ^d (4)	4	186(69.7%)	0
<i>A. arabiensis</i>	301	96	86(9)	8	2	79(29.6%)	205	9	181 ^e (8)	15	188(70.4%)	0
<i>A. melas</i>	8	1	1(0)	0	0	1	7	0	7(0)	0	7	0
Total	605	195	180(13)	12	3	—	410	13	378(12)	19	—	0

^aNumber of mosquitoes estimated to have fed on humans outside the trap in parentheses.

^bNumber of mosquitoes estimated to have fed on the host in the trap (see text); figures in parentheses are the estimated proportions feeding in each trap.

^cNumber of mosquitoes estimated to have fed on bovines outside the trap in parentheses.

^dIncluding one mixed bovine and sheep blood meal.

^eIncluding one mixed bovine and horse blood meal and one mixed bovine and sheep blood meal.

bovine-baited traps had taken blood meals only from humans. These mosquitoes had clearly fed elsewhere than under the bed nets in which they were captured. If one considers that these fed vectors had entered the net seeking a resting site, one can assume that an equal number of vectors had fed on bovines other than the bovine bait, and had similarly entered the bovine bed net searching for a resting site. A similar assumption can be made concerning the mosquitoes which had fed on humans. One can, therefore, estimate the probable number of each species which had fed on the bait in each bed net trap (Table).

The numbers of the 2 *Anopheles* species caught in the 2 baited traps were not significantly different ($\chi^2=0$, $P=0.96$).

Discussion

A. gambiae and *A. arabiensis* were the dominant species of *Anopheles* caught in each of the 2 baited traps. Contrary to the usual technique of capturing indoor resting mosquitoes, our method gave *A. gambiae* and *A. arabiensis* an equal choice between 2 hosts: human and bovine. The comparison between these 2 species can therefore be made in a prospective way, and not retrospectively using a sample of endophilic mosquitoes.

When presented with equally available outdoor hosts, *A. gambiae* and *A. arabiensis* did not differ in their feeding preference. These observations confirmed our previous finding, using indoor spraying in the same village, Dielmo, that 73.7% of *A. gambiae* and 70.8% of *A. arabiensis* had fed on humans ($\chi^2=0.21$, $P=0.65$). No anopheline was captured in 2 pit shelters dug in the village, in holes in walls, or under tree roots, suggesting a high degree of endophily (D. Fontenille, unpublished observations). Similar results, indicating similar trophic preferences, have been obtained in other villages in Senegal (FONTENILLE *et al.*, 1997b; LEMASSON *et al.*, 1997).

Our results do not agree with most of the other studies comparing the feeding preferences of these 2 species. However, WHITE *et al.* (1972) noted in Tanzania that, if the HBI of *A. gambiae* is superior to that of *A. arabiensis* among indoor resting mosquitoes, the opposite was true among the outdoor resting population. Similarly, SERVICE *et al.* (1978) observed no significant difference in HBI between *A. gambiae* and *A. arabiensis* captured indoors in the region of Kisumu in Kenya. Such differences between studies can be explained by variations in sampling techniques. A valid comparison of feeding preferences can be made only if the 2 species are captured in the same place, at the same time and using the same sampling method. Additionally, at least 2 different hosts should be present. All of these essential conditions were present in our studies.

A. arabiensis may appear to be more zoophilic than *A. gambiae* only because it is more abundant in drier areas of Africa (sahelian and sudanian bioclimatic regions),

where cattle are also much more abundant. Another bias can be introduced if only half-gravid females are compared. These have been used in the past to determine the species through the study of polytene chromosomes. The use of PCR, as in our study, allowed the identification of all mosquitoes captured.

A. gambiae and *A. arabiensis* are highly polymorphic, with a genetic structure and behaviour which are adaptable to different environments. COLUZZI (1984) and PETRARCA & BEIER (1992) showed that their HBIs varied according to the degree of chromosomal inversion. *A. gambiae* and *A. arabiensis*, therefore, are not likely to exhibit the same behaviour throughout Africa. Comparison of various studies carried out in Africa shows that there are considerable differences between East and West Africa, particularly with *A. arabiensis*.

Several authors have shown that the sporozoite rate of *A. gambiae* is often higher than that of *A. arabiensis*. This higher rate is generally partly attributed to the higher degree of anthropophily exhibited by *A. gambiae*. In Dielmo, during a 3 years' follow-up study, we showed that the sporozoite rate of *A. gambiae* was higher than that of *A. arabiensis* (1.46% and 0.60%, respectively) (FONTENILLE *et al.*, 1997a). The present results indicate that several additional factors, other than the HBI, should be sought to explain differences in the sporozoite rate. These may include greater longevity of *A. gambiae* or other genetic factors related to vectorial competence.

In our experimental conditions, the calf attracted more *A. gambiae* and *A. arabiensis* than did the human, despite their similar weights. These results and previous work (HADIS *et al.*, 1997) suggest that the zoophily of *A. gambiae* and *A. arabiensis* may have been underestimated. This may have resulted from the fact that the feeding behaviour had usually been determined from mosquitoes resting in sleeping quarters (COSTANTINI *et al.*, 1998).

This work has demonstrated, once more, the difficulty of calculating the exact HBI for malaria vectors (GARETT-JONES, 1964; KAY *et al.*, 1979). The use of different trapping methods (bedroom spraying, granary spraying, outdoor bed net traps, pit shelters) for different species of *Anopheles* may permit a more accurate approximation and comparison of one vector species with another.

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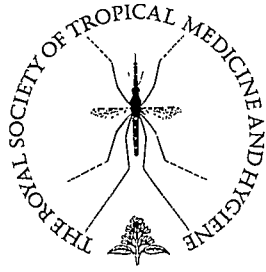
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Announcement

ROYAL SOCIETY OF TROPICAL MEDICINE AND HYGIENE Garnham Fellowship Fund Appeal

The appeal for funds to establish fellowships in memory of the late Professor P. C. C. Garnham, FRS, is progressing well. A Garnham Fellowship will enable a young physician or scientist to carry out a short term field project in parasitology or medical entomology in a tropical country of their choice and will be a fitting memorial to Cyril Garnham, who believed passionately in the importance of field work. The appeal has already received generous sponsorship from the Garnham family and the London School of Hygiene and Tropical Medicine. Glaxo Wellcome plc has made a generous donation on the understanding that the Society raises an equivalent amount. Fellows who have not yet contributed but would like to do so are asked to send a donation by cheque (in pounds sterling or Canadian or US dollars) or credit card (stating the number and expiry date) to the Honorary Treasurer, Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W1N 4EY, UK; fax +44 (0)171 436 1389, e-mail mail@rstmh.org



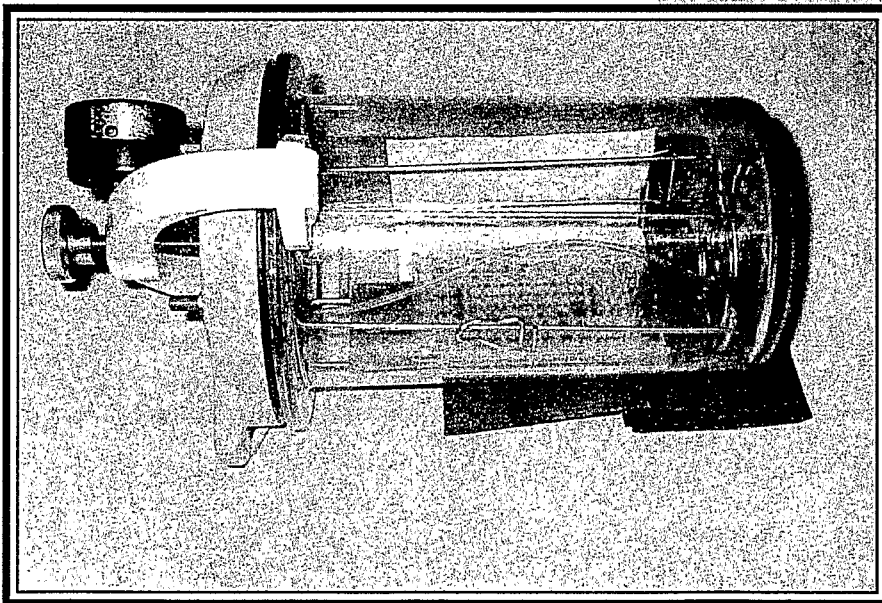
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