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Host choice by indoor-resting *Anopheles arabiensis* in Ethiopia

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Abstract

The host preference of indoor resting *Anopheles arabiensis* has been determined using a direct enzyme-linked immunosorbent assay. A total of 611 specimens, 258 from human dwellings, 179 from mixed dwellings, and 174 from cattle sheds, was examined. The proportion of human blood meals identified was highest from mosquitoes caught in human dwellings (91·5%), followed by those from mixed dwellings (20·2%) and cattle sheds (3·5%) ($P<0·0001$). The smaller proportion of human blood meals from mixed dwellings suggests that cattle may protect humans from *A. arabiensis*.

Keywords: malaria, *Plasmodium* spp., *Anopheles arabiensis*, blood meals, cattle, Ethiopia

Introduction

Two members of the *Anopheles gambiae* complex are known to exist in Ethiopia: *A. arabiensis* and *A. quadriannulatus* (see WHITE *et al.*, 1980). The complex is found in all administrative regions (O'CONNOR, 1967). *A. arabiensis* is the most important malaria vector in the country (WHITE *et al.*, 1980). It is also a vector of filariasis in Gambella, western Ethiopia, the only place where filariasis is endemic (MCCONNELL & SCHMIDT, 1973).

The primary objective of identifying blood meals is to obtain an indication of the degree of biting contact between a specified mosquito population and humans. This information could be useful in planning and evaluating malaria control measures (GARRETT-JONES *et al.*, 1980). The presence of domestic animals has been associated with a decrease in malaria transmission rates due to 'zoophilic deviation' (BRUCE-CHWATT *et al.*, 1966). In some parts of Africa zooprophylaxis is used against mosquitoes such as *A. arabiensis*; cattle are intentionally kept near or inside houses to divert mosquitoes from humans to cattle (BURKOT, 1988); this is common in southern Ethiopia. The need to assess the impact of community practices deliberately to deflect mosquitoes from humans has been emphasized by GARRETT-JONES *et al.* (1980).

In this paper we report the results of an investigation of the host preferences of indoor resting *A. arabiensis* in 6 areas of Ethiopia where malaria is endemic.

Materials and Methods

Study areas

This study was carried out from May 1995 to October 1995 in 6 malarious localities in east, south, and west Ethiopia. Ledi and Alibeti are localities in the east; the people are mainly pastoralists, occasionally practising small scale agriculture. The inhabitants of Sille in the south are mostly state farm workers; in some families a few cattle are usually kept; in Erbore, further south, the people are pastoralist. Itang and Jawe are both in western Ethiopia. Cattle keeping is predominant in Itang while in Jawe the main activity is subsistence level agriculture, honey gathering and hunting.

Mosquito collection

Mosquitoes were collected from Ledi, Alibeti, Sille and Erbore in May 1995, and from Itang and Jawe in October 1995. Indoor resting mosquitoes were collected between 05:00 and 07:00, using mouth aspirators. Mosquitoes collected from human dwellings, cattle sheds and mixed dwellings were labelled and kept separately.

The morphological keys of VERRONE (1962) were used to identify *A. gambiae s.l.* Mosquitoes were then kept in cages for one day to increase the number of half-gravid females. A representative group was kept separately for later cytogenetic identification (COLUZZI *et al.*, 1979) after anaesthetizing them with chloroform. Most of the specimens were dried in paper cups and then transferred to small vials for blood meal identification.

Blood meal identification

The direct enzyme-linked immunosorbent assay (ELISA) described by BEIER *et al.* (1988), with a slight modification, was used to test mosquitoes for human and bovine immunoglobulin (Ig). Each mosquito's abdomen was crushed in 125 µL of phosphate-buffered saline (PBS) and 50 µL of the triturate was added to flat-bottomed 96-well microtitre plates (Linbro, USA); 50 µL of peroxidase-conjugated anti-human IgG (A-8419, Sigma, USA) or the same volume of peroxidase-conjugated anti-bovine IgG (A-8917, Sigma, USA) were then added. Both conjugates were diluted 1:2000 in 0·5% boiled casein containing 0·025% Tween-20™ (Sigma, USA). Finally, 100 µL of 2,2′-azino-bis-[3-ethylbenzthiazoline-6-sulphonate]) were added to each well.

Absorbance at 405 nm was recorded with an ELISA reader (Labsystems Multiscan™ MCC 340, Finland) 30 min after the addition of ABTS. Human and bovine blood dried on filter paper were used as positive controls, while male anopheline mosquitoes served as negative controls; both controls were included on each plate. Samples were considered positive when their absorbance values were greater than the mean value plus 3 times the standard deviation obtained with 4 negative control wells.

Results

Of 611 *A. arabiensis* collected from different habitats, 325 (53·2%) gave a positive reaction to either human or bovine blood, or both. The highest proportion of human blood was found in mosquitoes collected from human dwellings, followed by mixed dwellings and cattle sheds (Table); the differences were statistically significant ($P<0·0001$).

Some bovine meals were recorded from human dwellings, and some human meals from cattle sheds. The highest proportion of mixed feeds came from mixed dwellings (i.e., those inhabited by both humans and cattle). The differences in the proportions of mixed feeds were not significant, except for that between mixed and human dwellings ($P=0·05$). Among specimens from mixed dwellings, the bovine proportion was more than 3 times higher than the human-fed proportion (Table).

Discussion

Higher proportions of human blood meals in mosquitoes collected from human dwellings than from the mixed dwellings and cattle sheds is to be expected. WHITE *et al.* (1980) reported a human blood index of 100% for *A. gambiae s.l.* collected from houses in Jimma, and KRAFSUR (1971) also reported a similar result in Gambella, both in Ethiopia. In another study which

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Table. Identification[a] of blood meals of indoor resting *Anopheles arabiensis* collected from different habitats in Ethiopia

Origin	No. tested	Proportion identified	Blood meal[b] Human	Bovine	Mixed
Human dwelling	258	50·4%	119(91·5%)	9 (6·9%)	2(1·5%)
Cattle shed	174	49·4%	3 (3·5%)	82(95·3%)	1(1·2%)
Mixed dwelling	179	60·9%	22(20·2%)	79(72·5%)	8(7·3%)

[a]Identified by enzyme-linked immunosorbent assay.
[b]Percentages shown are calculated in relation to the total number identified.

involved different habitats around Jimma, the highest human blood proportion was found in mosquitoes from human dwellings followed by those from mixed dwellings, animal sheds and pit shelters (GARRETT-JONES *et al.*, 1980).

ADUGNA & PETROS (1996), in Ethiopia, recorded a higher proportion of human blood meals in *A. gambiae s.l.* from both mixed dwellings (88%) and cattle sheds (43%) than we and other workers (GARRETT-JONES *et al.*, 1980) have found, and, in India, JOSHI *et al.* (1988) found 4% and 93% of human and bovine blood meals, respectively, in *A. culicifacies* collected from human dwellings. In a world-wide review of mosquito blood-feeding, GARRETT-JONES (1964) reported that the percentage of anophelines collected from human dwellings which contained human blood varied from 15·8% to 97·5%, while in samples from animal sheds it was 0·2–35%; the differences were significant at the 5% level for all samples except for *A. superpictus* in Cyprus. The presence of bovine-fed mosquitoes in human dwellings and human-fed mosquitoes in cattle sheds is possibly due to mosquitoes which have fed outdoors on one or other of these hosts later resting in cattle sheds or human dwellings.

Mixed feeding by mosquitoes is common (SHALABY 1969; GARRETT-JONES *et al.*, 1980; RUBIO-PALIS *et al.*, 1994), and its epidemiological implication is controversial (BOREHAM & GARRETT-JONES, 1973; BURKOT *et al.*, 1988). However, the loss of a certain number of sporozoites in non-human hosts during mixed feeding could be of importance in malaria control.

The fact that we found a higher proportion of bovine feeds than human feeds in mosquitoes caught in mixed (bovine plus human) dwellings indicates a preference of *A. arabiensis* for cattle. This finding is in accordance with previous studies in Nigeria, Madagascar, Tanzania, Burkina Faso, Kenya and elsewhere (WHITE *et al.*, 1972; WHITE & ROSEN, 1973; WHITE, 1974; HIGHTON *et al.*, 1979). These reports suggest that cattle could play a role in reducing transmission of malaria by *A. arabiensis* by distracting the vector.

On the other hand, SCHULTZ (1989) has observed that the human-biting rate of *A. flavirostris* in the Philippines increased in the presence of other animals and HEWITT *et al.* (1994) observed this with *A. stephensi* in Pakistan. Additionally, the prevalence of malaria transmitted by *A. aconitus* in Indonesia was found to be higher when cattle were nearby (KIRNOWORDOYO & SUPALIN, 1986) and a study in Pakistan gave similar results (BOUMA & ROWLAND, 1995). In our study, the lower proportion of human than bovine blood meals in mosquitoes collected from mixed dwellings is contrary to the findings of SCHULTZ *et al.* (1989) and HEWITT *et al.* (1994). It is clear that conclusions regarding zooprophylaxis must be confined to specific hosts and mosquito species. These conflicting reports indicate the need for further controlled studies on the potential of livestock to divert mosquitoes from humans.

However, previous reports (WHITE *et al.*, 1972; WHITE & ROSEN, 1973; WHITE, 1974; HIGHTON *et al.*, 1979; GARRETT-JONES *et al.*, 1980) and the present study indicating a preference of *A. arabiensis* for cattle suggest that this species, at least in certain areas, is a possible candidate for zooprophylaxis of malaria.

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Announcements

ROYAL SOCIETY OF TROPICAL MEDICINE AND HYGIENE
Denis Burkitt Fellowships

The Denis Burkitt Fund was set up by his family in memory of Denis Burkitt, FRS, who died in 1993; it is administered by the Royal Society of Tropical Medicine and Hygiene.

Two Fellowships (maximum value £3000 each) are awarded annually for practical training, travel, or direct assistance with a specific project (preferably clinico-pathological, geographical or epidemiological studies of non-communicable diseases in Africa).

Application forms, **which must be returned at least six months before the commencement of the proposed study (by 15 March or 15 September in each year)**, are available from the Administrator, Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W1N 4EY, UK.

ROYAL SOCIETY OF TROPICAL MEDICINE AND HYGIENE
Robert Cochrane Fund for Leprosy

The fund, in memory of the great leprologist Rochert Cochrane, is administered by the Royal Society of Tropical Medicine and Hygiene. It is used to finance up to three travel fellowships each year to a maximum value of £1000 each.

The Fund will support travel for
**Leprosy workers who need to obtain practical training in field work or in research
**Experienced leprologists to provide practical clinical training in a developing country
There is no restriction on the country of origin or destination providing the above requirements are met.

Applications must be made at least six months ahead of the proposed trip, sponsored by a suitable representative of the applicant's employer or study centre and agreed by the host organization. A short report on the travel/study should be submitted, within one month of the recipient's return. Application forms are available from the Administrator, Royal Society of Tropical Medicine and Hygiene, Manson House, 26 Portland Place, London, W1N 4EY, UK; fax +44 (0)171 436 1389.

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