ASSessment of Malaria transmission
In an area with very low mosquito density

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INTRODUCTION

The increase in world travel in recent years, especially to and from areas where vector-borne diseases are endemic, has resulted in a substantial rise in imported cases of those diseases. In particular, malaria is a cause of concern. In those countries at the edge of its distribution, it can be difficult to distinguish between autochthonous and imported cases. However, distinguishing between the two is important because of the different allocation of resources to combat the disease that each requires.

In general, observation of the various stages of parasite development in wild-caught female mosquitoes is considered evidence of autochthonous transmission. Observation of oocysts in the mosquito mid-gut testifies that mosquitoes are susceptible to infection but conclusions cannot be reached about their ability to complete the transmission cycle. Perhaps the best indication of autochthonous transmission is microscopic observation of sporozoites in mosquito salivary glands, since this detects parasites ready to be inoculated. Detection of circumsporozoite protein (CSP) in dry mosquito thoraxes, by Enzyme Linked Immunosorbent Assay (ELISA) is also widely used to determine transmission, especially when large numbers of mosquitoes need to be processed. Such assays provide information about the parasite species infecting the mosquito. They are, however, considered less accurate than dissection since sporozoites in salivary glands cannot be distinguished from those in the haemocel. Detection of antibodies against CSP-protein by ELISA were used to assess mosquito mediated malaria transmission in the village of Achada Leite on the archipelago of Cabo Verde, an area with very low mosquito densities where a clonal Plasmodium falciparum outbreak occurred in October 1995. Anti-CSP antibodies were found among individuals who had previously been malaria patients, none of whom had travelled out of the outbreak area. The malaria vector Anopheles arabiensis was collected in both wet and dry seasons.

KEY WORDS: Plasmodium falciparum, Anopheles arabiensis, malaria transmission, Cabo Verde, anti-CSP ELISA.

In many marginal zones of transmission, the collection of infected mosquitoes is, however, extremely difficult. Under such circumstances alternative measures are required to establish the presence of local transmission. These have only been reported from Santiago, the main island of the group. In October 1995, a clonal outbreak of Plasmodium falciparum Welch, 1897, malaria was recorded in the small village of Achada Leite (Arez et al., 1997) but entomological studies were unable to fully demonstrate autochthonous malaria transmission. Collections of potential vectors and assessment of anti-sporozoite antibodies in residents was therefore undertaken to confirm that transmission was taking place in the village.

MATERIAL AND METHODS

Achada Leite, 40 kilometers Northwest from the capital, Praia, on Santiago island, has approximately 200 inhabitants living in 35 brick-walled, zinc-roofed houses. These are distributed along the high edges of an isolated deep green valley. A deserted plateau surrounds the valley on three sides with the seashore on the fourth. A freshwater stream flows in the middle of this valley during the rainy season. The climate is characterised by a dry (December to June) and a wet season (July to November) with very low rainfall. Strong winds are common. Two anopheline species have been recorded for the archipelago, Anopheles pretoriensis Theobald, 1903 and Anopheles melas Theobald, 1903.
TRARCA

Gambiae s.l. Giles, 1902, mosquitoes were kept dried at room temperature. The outdoor traps were hung close to windows or by animal shelters. None of the adult specimens collected were infected with Plasmodium. Despite the dearth of adult mosquitoes, larvae were relatively common. Six positive habitats were identified and in the dry season and three in the wet season. Altogether, 674 An. pretoriensis and 211 An. gambiae s.l. were obtained from larval collections. Culex ethiopicus Edwards, 1912, Culex pipiens s.l. Linnaeus, 1758, Culex tigripes De Grandpre et De Charmoy, 1900, Aedes aegypti Linnaeus, 1762, and Culiseta longiareolata Macquart, 1838, were also collected from these larval sites. In both seasons, An. gambiae s.l. eggs, all larval stages and pupae, were found together in the same breeding sites.

All of the An. gambiae s.l. identified by either PCR or cytogenetical means were An. arabiensis. Three mosquitoes were negative by PCR. These were further analysed with a primer specific for Anopheles melas Theobald, 1903, but remained negative. The chromosomal inversions recorded in 6 readable specimens of An. arabiensis were 2Ra and 2Rb, both found as inverted homozygotes, and 3Ra, which was polymorphic.

Six (43%) of the 14 former malaria patients tested for anti-CSP Ab were positive. The cut-off value of the ELISA assay was 0.09 and the absorbances of the positive controls ranged from 0.556 to 0.897 (Table 1). Both children and adults were positive (< 5 years, n = 2; 5-14 years, n = 1; and 50 years, n = 3). Three of the positive cases belonged to the same family (subjects 1, 5 and 12) but there was no evidence of family ties amongst the other positive cases and there was no history of any of these subjects travelling out of the area.

**DISCUSSION**

Malaria transmission in the area was difficult to assess, since even though a variety of collection methods were employed, only a small sample of adult mosquitoes was obtained.
According to Gillies & De Meillon (1968), very few An. pretoriensis have been found infected with malaria parasites and, considering the zoophilic habits of this species, those were more likely to be of non-human origin. Although unlikely, available data are not enough to completely exclude An. pretoriensis from playing a role in transmitting malaria in the study area.

Nevertheless, entomological data, although scarce, support the idea that An. arabiensis should be the only potential vector of malaria in the area. The fact that larvae were collected in both wet and dry seasons also indicates that low-level malaria transmission is possible throughout the year.

Previously, Ribeiro et al. (1980) concluded that the characteristic unstable malaria of the archipelago of Cabo Verde was due to a low vectorial capacity of the local An. arabiensis populations, rather than to the presence of a lesser effective malaria vector, An. pretoriensis.

Adult mosquito densities are likely to remain low throughout the year and human blood meal sources may be difficult to obtain, due to inclement weather conditions (strong winds are common), altitude and well-built houses.

An. arabiensis, together with An. gambiae s.s., are the most anthropophilic and widespread members of the An. gambiae complex and constitute with Anopheles funestus Giles, 1900, the most efficient vectorial system available in the world for P. falciparum (Coluzzi, 1994). An. arabiensis is known to successfully exploit dry habitats, such as arid savannahs and steppes (Coluzzi et al., 1979). Indeed, the capacity of An. arabiensis to persist and maintain malaria transmission during the dry season in hot-dry savannah regions and even to maintain a certain level of malaria transmission has been stressed by several authors (Omer & Cloudsley-Thompson, 1970; Taylor et al., 1993; Touré et al., 1996).

Although the small number of chromosomally processed specimens does not allow a thorough analysis, the An. arabiensis karyotypes recorded in this study were similar to those described by Cambournac, Petrarca & Coluzzi (1982) in various islands of the archipelago during July-October 1981. Thus, the An. arabiensis population of these islands is apparently very stable. It has been proposed that An. arabiensis was introduced to the islands from the nearest continental area, Dakar-Senegal (Cambournac, Petrarca & Coluzzi, 1982); and samples from the area of St. Louis, Senegal had a karyotype composition which is very similar to those of Cabo Verde, the main difference being the higher degree of polymorphism of the continental An. arabiensis (Petrarca, Vercruysse & Coluzzi, 1987). The lower degree of polymorphism in Cabo Verde is presumably the result of the founder principle and/or canalising selection (Mayr, 1970). However, the high frequency of the inverted arrangement 2R in Cabo Verde suggests that selection and adaptation are involved, since it has been shown that the frequency of the carriers of this arrangement increases in more arid sampling sites (Coluzzi et al., 1979; Petrarca, Vercruysse & Coluzzi, 1987).

In conclusion, this study suffered from a very small entomological sample, which did not allow determination of the intensity of malaria transmission based on standard measurements in the local mosquito population. Therefore, entomological inoculation rates or the vectorial capacity could not be estimated. However, the presence of An. arabiensis, one of the most powerful African malaria vectors, with apparently perennial breeding, together with the finding of antibodies to circumsporozoite protein among the inhabitants, are indicators of malaria transmission in a non-endemic area where a clonal P. falciparum outbreak occurred and persisted for a year.

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