Available online at www.sciencedirect.com
ScienceDirect
Acta Tropica 103 (2007) 102–107
ACTA TROPICA
www.elsevier.com/locate/actatropica

Genetic diversity and gene flow of humans, *Plasmodium falciparum*, and *Anopheles farauti s.s.* of Vanuatu: Inferred malaria dispersal and implications for malaria control

J.K. Lum [a,b,c,1], A. Kaneko [d], G. Taleo [e], M. Amos [e], D.M. Reiff [a,b,*,1]

[a] *Laboratory of Evolutionary Anthropology and Health, Binghamton University, USA*
[b] *Department of Anthropology, Binghamton University, P.O. Box 6000, Binghamton, NY 13902-6000, USA*
[c] *Department of Biological Sciences, Binghamton University, P.O. Box 6000, Binghamton, NY 13902-6000, USA*
[d] *Unit of Infectious Disease, Department of Medicine, Karolinska Institute, Stockholm, Sweden*
[e] *Vanuatu Ministry of Health, Government of the Republic of Vanuatu, Port Vila, Vanuatu*

Received 20 May 2007; accepted 22 May 2007
Available online 25 May 2007

Abstract

A comparison of the patterns of gene flow within and between islands and the genetic diversities of the three species required for malaria transmission (humans, *Plasmodium falciparum*, and *Anopheles farauti s.s.*) within the model island system of Vanuatu, shows that the active dispersal of *An. farauti s.s.* is responsible for within island movement of parasites. In contrast, since both *P. falciparum* and *An. farauti s.s.* populations are largely restricted to islands, movement of parasites between islands is likely due to human transport. Thus, control of vectors is crucial for controlling malaria within islands, while control of human movement is essential to control malaria transmission across the archipelago.

Keywords: Vanuatu; Gene flow; *Anopheles farauti s.s.*; Malaria

1. Introduction

Malaria is a vector borne disease responsible for the death of over one million people each year (Guinovart et al., 2006). Vector borne diseases involve the interaction of three species; in the case of malaria these species include the human host, a variety of regionally specific *Anopheles* vectors and at least four *Plasmodium* parasite species.

The *Anopheles punctulatus* species complex is the primary vector of malaria in the Pacific. Although traditional proboscis morphology identified four *An. punctulatus* species (Rozeboom and Knight, 1946; Beebe and Cooper, 2000), subsequent cross mating experiments, as well as allozyme electrophoresis, have identified 12 species in the *An. punctulatus* complex with distinct ecological and behavioral characteristics (Beebe and Cooper, 2000; Harbach, 2004). These 12 species have overlapping proboscis morphologies, making it impossible to precisely identify species in the field (Beebe and Cooper, 2002). The development of polymerase chain reaction–restriction fragment length polymorphism (PCR–RFLP) analysis of the ribosomal

* Corresponding author. Tel.: +1 607 777 2737.
E-mail address: dreiff1@binghamton.edu (D.M. Reiff).
[1] J.K. Lum and D.M. Reiff contributed equally to the manuscript.

0001-706X/$ – see front matter © 2007 Elsevier B.V. All rights reserved.
doi:10.1016/j.actatropica.2007.05.012

internal transcribed spacer 2 (ITS2) region provided an accurate identification method for these cryptic species (Beebe and Saul, 1995; Cooper et al., 2002).

Human settlement of the Pacific involved two distinct stages. Pleistocene colonization established human populations as far as the Solomon Islands by 32,000 years ago (Wickler and Spriggs, 1988). A second human migration began 3500 years ago, settling the remaining islands of the Pacific (Spriggs and Anderson, 1993; Green, 1999). In the Pacific, Vanuatu is the eastern limit of *Anopheles*, and thus malaria. The time depth of human settlement corresponds with *Anopheles* species diversity across Melanesia (Lum et al., 2004): areas settled in the Pleistocene have all 12 *An. punctulatus* species, while Vanuatu, settled in the Holocene, is thought to contain only one species of *Anopheles*.

Anopheles farauti s.s. had been previously identified as the only member of the *An. punctulatus* species complex on Vanuatu based on morphology (Rozeboom and Knight, 1946), allozyme electrophoresis (Foley et al., 1994), and ITS2 analyses (Beebe and Saul, 1995; Beebe et al., 2000). Molecular techniques were restricted to four north central islands. Recently, more extensive sampling across the geographical range of the archipelago (from 14 to 19° latitude) confirmed *An. farauti s.s.* as the single species of Vanuatu using ITS2 analyses (Reiff et al., 2007). This species is salt water tolerant making it ideal to stowaway on native canoes or modern boats (Sweeney, 1987; Service, 1997; Beebe and Cooper, 2002). The dispersal ability of *An. farauti s.s.* is reflected in its current distribution; it has the largest range of any *An. punctulatus* species, extending from New Guinea to Vanuatu (Beebe and Cooper, 2002).

Human and *P. falciparum* gene flow patterns were previously examined in four islands of Northern Vanuatu (Lum et al., 2004). Human gene flow was found to be necessary for parasite movement among islands (Lum et al., 2004). Within islands, genetically distinct human populations shared parasite populations, implicating the active dispersal of the vector for within island dispersal of the malaria parasite (Lum et al., 2004). Investigating the contribution of the *Anopheles* mosquito to malarial dispersal on Vanuatu became the next step in understanding the interaction between the three species required for malaria transmission. Thus, population and gene flow patterns of *An. farauti s.s.* were recently examined in five islands of Vanuatu (Reiff et al., 2007). Gene flow was high among populations within islands. When variance was partitioned at different levels of the population structure, variation between sites within islands was minimal (2.3%) (Reiff et al., 2007). Gene flow was also very restricted among islands, with a large portion of the variance partitioned among islands (46.4%) (Reiff et al., 2007).

In the present study, we determine the relationship between the three species involved in malaria within the model island system of Vanuatu. In particular, we examine if the *Anopheles* mosquito is responsible for within island parasite gene flow, as predicted based on analyses of patterns of human and *Plasmodium* genetic diversity (Lum et al., 2004). Here, we compare genetic data for humans and parasites (Lum et al., 2004) to *An. farauti s.s.* populations collected from the same four Northern islands of Vanuatu.

2. Materials and methods

Genetic data was generated from three species: *Homo sapiens, P. falciparum,* and *An. farauti s.s.* Human and *P. falciparum* DNA were obtained from human blood collected during malariometric surveys from seven sites on four islands of Northern Vanuatu (Gaua: $n=2$, Santo: $n=1$, Malekula: $n=2$, Pentecost: $n=2$) (Fig. 1)

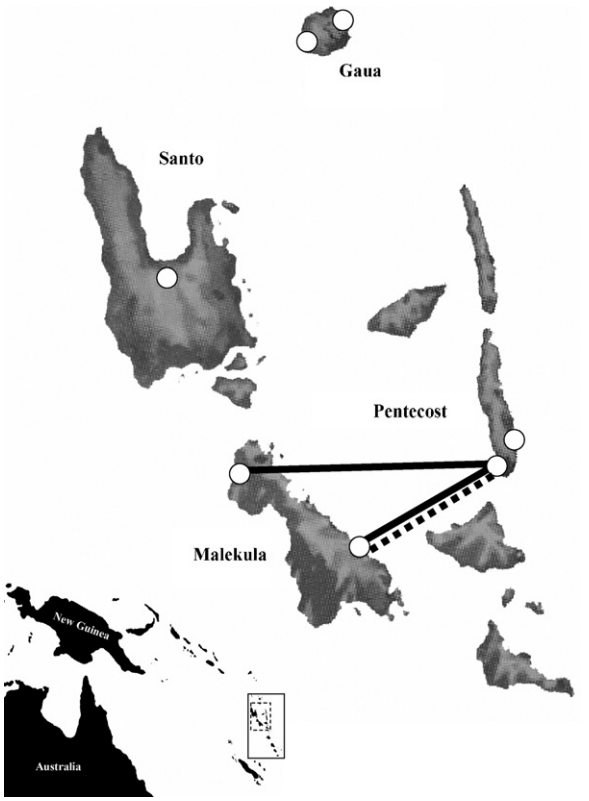

Fig. 1. Map of Vanuatu showing seven collection sites on four islands (open circles). The lines connect populations that are not significantly distinct ($P>0.05$). The solid lines represents among island *P. falciparum* gene flow, the dashed line *H. sapiens* gene flow. All *An. farauti s.s.* among island populations are significantly distinct ($P<0.01$).