

INTRODUCTION

Keys for the identification of *Anopheles* mosquitoes are required for studies on the epidemiology and transmission of malaria. Many of the illustrated keys to the *Anopheles* of Thailand (Peyton and Scanlon, 1966; Rattarithikul and Harrison, 1973) are of limited value, as these were published more than 20 years ago and significant advances in our knowledge of the *Anopheles* mosquitoes have occurred in the intervening years. The purpose of the keys presented in this paper is to assist entomologists to identify larvae and adult female *Anopheles* mosquitoes. The keys can be used to initially identify specimens to species group and then to species. Discriminating characteristics are highlighted in drawings and, whenever possible, were chosen so that they could be differentiated using a hand lens (10x) or dissecting microscope (10-40x). The morphological characters used here are based on original observations and previous usage in the literature. The following references were especially helpful: Christophers (1933), Colless (1956, 1957), Reid (1968), Harrison (1972, 1980), Harrison and Scanlon (1975), Rattarithikul and Green (1986), Harbach *et al.* (2005), Linton *et al.* (2005), and Sallum *et al.* (2005). Nomenclature for morphological characters follows Harrison and Scanlon (1975), Harbach and Knight (1980, 1982), and Wilkerson and Peyton (1990). Generic and subgeneric abbreviations are those of Reinert (2001), Tanaka (2003), and Harbach *et al.* (2005).

SIBLING SPECIES AND GENETIC VARIATION IN ANOPHELINE MOSQUITOES

Combinations of morphological and other systematics methods have proven very useful in the recognition of sibling species in many groups of insects, most notably the medically important anopheline mosquitoes. Many anopheline taxa previously recognized as medically important in Southeast Asia have recently been found to be complexes of morphologically indistinct species. These discoveries suggest that in many Asian countries there is a need for the reassessment of primary vector species that were originally recognized solely on morphological methods. Important vector species should be reconfirmed using a combination of other appropriate techniques, including cytogenetic, biochemical, and molecular methods as exemplified by Baimai (1988a-d), Green (1982), Green *et al.* (1992), Panyim *et al.* (1988), and Rongnoparut *et al.* (1996, 1998, 1999), rather than relying on morphological criteria alone. The non-morphological methods are particularly useful if one has access to adult progeny (with associated larval and

pupal exuviae) reared from feral females. For example, a wild-caught female can be pinned and her morphological characters compared with those of her progeny. Wild-caught females can also be identified by ovarian polytene chromosome banding patterns or by PCR methodology, and also checked for sporogonic-stage malaria parasites using a sporozoite antigen panel assay kit, or by dissecting the salivary glands and examining them for sporozoites. Such approaches can be very revealing.

Until the late 1970s, *An. balabacensis* Baisas was regarded as an important vector of human malaria in Thailand and much of Southeast Asia. However, since then what was previously considered *An. balabacensis* on mainland Southeast Asia has been shown by morphological and non-morphological techniques to be a number of sibling species, namely *An. baimaii* Sallum and Peyton (2005), *An. cracens* Sallum and Peyton (2005), *An. dirus* Peyton and Harrison (1979), *An. nemophilous* Peyton and Ramalingam (1988), and *An. scanloni* Sallum and Peyton (2005), and *An. latens* Sallum and Peyton (2005) of the Leucosphyrus Complex. Recognition of the Dirus Complex prompted a reassessment of the distribution of *An. balabacensis*, which belongs to the Leucosphyrus Complex (Peyton, 1990) and is now restricted to certain islands in the Philippines, Indonesia, and Malaysia. *Anopheles dirus* and *An. baimaii* (Green *et al.*, 1991, as *dirus* D) are now regarded as the principal malaria vectors in Thailand. Another example of the value of using multiple methods, is the combination of morphological, cytogenetic, and related studies that revealed *An. maculatus* E (Delorme *et al.*, 1989; Kittayapong *et al.*, 1992) and *An. culicifacies* A (Subbarao, 1988) are the major vectors of human malaria parasites in Malaysia and India, respectively.

A major concern of individuals interested in the systematics of anophelines is how to deal with chromosomal forms of species that have been designated by letters of the alphabet. If these prove to be distinct species, the International Code of Zoological Nomenclature provides guidelines for taxonomists to establish a name and diagnostic characters for identifying them. However, each putative species has to be shown to be distinct from currently named species. For example, four genetic forms (A, B, C, D) of *An. jeyporiensis* are recognized in Thailand (Baimai *et al.*, 1996a). The question must be asked, which of these, if any, is conspecific with *An. jeyporiensis* James, 1902? This can only be resolved by studying the molecular genetics of *An. jeyporiensis* specimens from the type locality (Nagpur, Jeypur, Orissa and Maharashta States, Central Provinces, India) (Knight and Stone, 1977) and then comparing the Thai chromosomal forms with the species that James described as *An. jeyporiensis* from the type locality (Table 1). If one of the Thai chromosomal forms is identical to the species that James described, then the

other three Thai chromosomal forms will need to be studied further to determine if they represent distinct species. It may turn out that all of the *An. jeyporiensis* chromosomal forms reported from Thailand are distinct from the species described by James. In this case, one or more of the forms in Thailand would need to be formally named and *An. jeyporiensis* James would be deleted from the Thai records. Two recent studies (Rattanarithikul and Harbach, 1990; Linton *et al.*, 2001) involving *An. maculatus* Theobald and *An. sundaicus* (Rodenwaldt), respectively, provide approaches for resolving such problems.

ROLE OF ANOPHELINE MOSQUITOES AS DISEASE VECTORS IN THAILAND

Malaria

Despite decades of successful control programs and dramatic reductions in morbidity and mortality, malaria remains one of the most important infectious diseases in Thailand (Chareonviriyaphap *et al.*, 2000). Malaria remains prevalent along the undeveloped borders of eastern Myanmar, western Cambodia, and northern Malaysia. Although reported malaria cases have declined from a peak of 349,291 in 1988 to 85,625 in 1995, the number of cases has since risen annually (Chareonviriyaphap *et al.*, 2000). All four known human malaria parasites are present in Thailand, with *Plasmodium falciparum* (Welch) and *P. vivax* (Grassi and Feletti) predominant (Gingrich *et al.*, 1990; Snounou *et al.*, 1993). Multi-drug resistant *P. falciparum* occurs in Thailand, with widespread resistance to chloroquine, sulfadoxine-pyrimethamine, 4-aminoquinoline, and mefloquine (Faver *et al.*, 1999). Currently, antimalarial drugs that are used alone or in combination for the radical cure of falciparum malaria in Thailand include mefloquine, primaquine, quinine, tetracycline, and artemeter/artesunate compounds, whereas chloroquine and primaquine remain the choice for radical treatment of *P. vivax*, despite increasing reports of chloroquine resistance in the region (Chareonviriyaphap *et al.*, 2000).

Historically, malaria control in Thailand consisted of a combination of (i) prompt diagnosis and treatment with appropriate antimalarial drugs in government health clinics and in almost 550 specialized malaria clinics, (ii) health education in schools and in the general community, and (iii) an aggressive mosquito control program that relies on country-wide intradomiciliary insecticide spraying once or twice a year with DDT or a synthetic pyrethroid and, if appropriate, the distribution of pyrethroid impregnated bed nets (Chareonviriyaphap *et al.*, 2000). The increased resistance of parasite populations to