SURVEY OF *ANOPHELES* MOSQUITOES (DIPTERA: CULICIDAE) IN WEST SUMBA DISTRICT, INDONESIA

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Abstract. In August 2007, mosquitoes were collected using four different collection methods at 2 upland interior and 2 coastal villages in West Sumba District, East Nusa Tenggara Province, Indonesia. Methods included human-baited and unbaited tent and malaise traps, human-landing collections (HLC), and unbaited CDC light traps. Mosquitoes were identified to species by morphological characters and all anophelines were tested for malaria circumsporozoite protein (CSP) using an enzyme-linked immunosorbent assay (ELISA). During six trap nights, 4,174 *Anopheles* mosquitoes belonging to 13 species were captured and identified: *An. aconitus, An. annularis, An. barbirostris, An. flavirostris,* Hyrcanus Group species, *An. indefinitus*, *An. kochi*, *An. leucosphyrus* group, *An. maculatus* s.l., *An. subpictus* s.l., *An. sundaicus* s.l., *An. tessellatus*, and *An. vagus.* Of potential disease vectors, *An. annularis, An. subpictus,* and *An. vagus* were the most frequently collected species in the upland interior sites, whereas *An. sundaicus, An. subpictus,* and *An. vagus* were most commonly found along the coast. The predominant species from evening human-landing collections (mosquitoes per human) were *An. subpictus* and *An. vagus* in the upland interior and *An. sundaicus* along the coast. All mosquitoes were non-reactive for *Plasmodium* CSP. One specimen of the *An. leucosphyrus* group was captured from indoor HLC in Tenateke Village, an upland interior location. This finding appears to represent a new collection record for Sumba Island.

Keywords: *Anopheles*, malaria vectors, Sumba Island, Indonesia

INTRODUCTION

Malaria is a major health burden in the East Nusa Tenggara Province of Indonesia. The island of Sumba is one of many malaria endemic islands forming the Lesser Sunda Archipelago, located along a southern arc extending from the central part of the country eastward. In West Sumba District, an area with perennial hypo- to mesoendemic transmission, malaria ranks first among the 10 major public health problems and is a leading cause of death

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(Syafruddin *et al*, 2006). Malaria prevalence in West Sumba has been found to vary between 6.83% during the wet season (November through April) and 4.95% during the dry period (May to October) (Asih *et al*, 2009, Syafruddin *et al*, 2009). The malaria parasites in West Sumba are predominately *Plasmodium falciparum* and *Plasmodium vivax* (Syafruddin *et al*, 2006, 2009). The inconsistent and poorly controlled use of anti-malarial drugs, spreading drug resistance, and cessation of widescale, organized vector control programs have all contributed to a resurgence of malaria in Sumba (Asih *et al*, 2009). More detailed bionomic and epidemiologic studies of malaria vectors in Sumba should provide a clearer picture of the current malaria threat and the evolving patterns of resurgent malaria. The results of a brief, single period vector mosquito survey in West Sumba, including vector species presence, their relative attraction to humans, and respective roles in malaria transmission are presented herein.

MATERIALS AND METHODS

The survey was carried out during a 10-day period in August 2007 in four villages in West Sumba District (Fig 1). The district occupies the entire western half of Sumba Island and is separated from the eastern district at 120º23′ E longitude, extending north and south from 9º18′ S to 10º20′ S latitude, reaching its western coastal limit at 108º55′ E longitude. The target sites were divided between interior villages: Lamboyadete (119.3457 E 9.7105 S; elevation 80 m bordering higher elevations), Tenateke (119.2028 E 9.5717 S; elevation 405 m), coastal villages: Patialabawa (119.3319 E 9.7508 S; elevation 76 m), and Radamata (Laura) (119.2456 E 9.3917 S elevation 30 m) locations. The wet season in West Sumba runs from December to April with an average yearly rainfall ranging from 1,200 to 2,450 mm. Depending on elevation, mean ambient temperatures vary between 18-20ºC and 25-33ºC during the wet and dry seasons, respectively. West Sumba is topographically diverse and comprised of coastal plains, mountains, low hills and valleys. Rice cultivation and subsistance agriculture is the most common profession in rural areas. The landscape of Lamboyadete is rural and hilly, consisting mostly of bush/shrub (63%), forest (12%), and grassland (11%). Tenateke village is a small village located in a heavily forested region of central West Sumba. Major land types include bush/shrub (55%), plantation (17%), and cultivated farmland (11%). Radamata is located in northern West Sumba on a coastal plain dominated by bush/shrub (42%) and grassland (28%). Patialabawa is on the southeastern coast and is mainly bush/shrub (29%), plantation (25%), and grassland (22%).

Site selection was based on previous and concurrent epidemiologic studies of malaria in West Sumba (Syafruddin *et al* 2006, 2009). Four collection methods were used: 1) all night (18.00 h to 06:00 h) human landing collections (HLC) (WHO, 1975), 2) modified human-baited tent traps and malaise traps, 3) unbaited tent traps and malaise traps (Stoops *et al*, 2010), and 4) unbaited (light without additional attractant) Centers for Disease Control (CDC) miniature light traps. Human landing collections were carried out in two houses over one or two nights for each location. Paired indoor and outdoor collections were conducted by four human collectors (2 indoor, 2 outdoor). Outdoor collection sites were 3 to 5 meters from the houses containing the indoor collectors. The human landing rate (HLR), a surro-

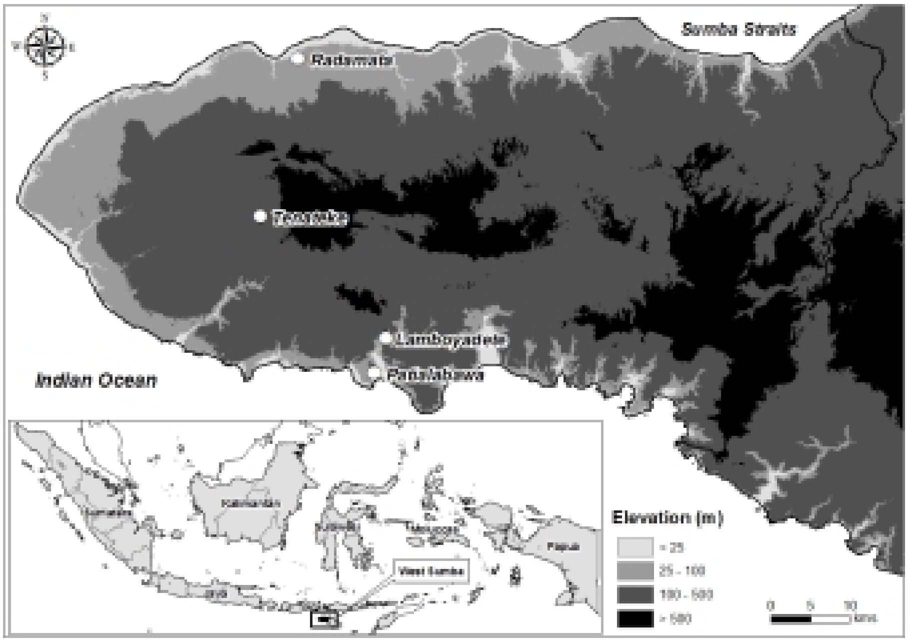

Fig 1–Map of West Sumba District, Sumba Island, East Nusa Tenggara Province, Indonesia showing the four mosquito collection sites.

gate measure of the actual 'biting' densities, was based on the total number of mosquitoes captured indoors and outdoors per 12-hour collection period averaged over two collection nights divided by total person-nights (Silver, 2008). Endophagy by species was defined as the proportion of mosquitoes captured indoors campared to total collected mosquitoes from HLC. Mosquitoes were sorted by genera and identified morphologically using keys for adult anopheline species native to Indonesia (Bonne-Webster and Swellengrebel, 1953; Reid 1968; O'Connor and Soepanto, 1989). Mosquitoes were stored individually in labeled vials over silica gel before laboratory processing.

Mosquitoes were assayed using a semi-quantitative enzyme-linked immunosorbent assay (ELISA) for the presence of *P. falciparum*, *P. vivax*, and a *P. vivax* variant (VK 247) circumsporozoite protein (CSP) antigens (Wirtz *et al*, 1987, 1992). Mosquitoes were dissected and only the head-thorax portion tested. Depending on numbers collected, specimens were tested either individually or pooled with up to 10 mosquitoes per well. The ELISA was performed following standard methodology with species-specific anti-sporozoite monoclonal antibodies (MAb) adsorbed onto the surface of individual U-bottom 96-well microtitration plates. Following a series of sequential steps and the addition of a peroxidase substrate solution all reactions were read at 405 nm using an ELISA plate reader (Benchmark Plus, BioRad Laboratories, Hercules, CA) at 30 and 60 minutes. Positive control antigens and negative controls from unifected, laboratory reared mosquitoes were run on each plate for each parasite species tested.

RESULTS

From 7 to 16 August, 2007, 5,786 female anopheline specimens were collected

Table 1
Total female anopheline species recorded by collection method in August 2007 from 2 coastal and 2 interior localities in West Sumba District, Indonesia.

	Coastal							Interior							
	Baited			Unbaited				Baited			Unbaited				Grand total
	Tent	Malaise	HLC	Tent	Malaise	Light	Total	Tent	Malaise	HLC	Tent	Malaise	Light	Total	
An. annularis	1	0	1	0	0	306	308	1	2	28	2	10	569	612	920
An. aconitus	1	0	0	0	0	0	1	0	2	96	0	0	46	144	145
An. barbirostris	2	0	1	0	1	1	5	0	6	15	6	7	14	48	53
An. flavirostris	0	0	0	0	0	0	0	0	0	15	0	0	3	18	18
An. hyrcanus gr	0	0	0	0	0	0	0	0	0	1	0	0	0	1	1
An. indefinitus	0	24	0	5	3	0	32	0	7	0	7	0	9	23	55
An. kochi	0	5	0	0	0	0	5	0	0	13	0	1	45	59	64
An. leucosphyrus gr	0	0	0	0	0	0	0	0	0	1	0	0	0	1	1
An. maculatus	0	0	0	0	0	0	0	0	0	5	0	0	4	9	9
An. subpictus	14	90	64	14	25	346	553	36	40	229	120	49	258	732	1,285
An. sundaicus	235	10	389	22	1	29	686	0	0	1	0	0	0	1	687
An. tessellatus	0	0	1	0	0	11	12	1	2	5	2	1	16	27	39
An. vagus	40	39	12	23	6	885	1,005	15	46	557	299	67	520	1,504	2,509
Total	293	168	468	64	36	1,578	2,607	53	105	966	436	135	1,484	3,179	5,786

Table 2
Anopheles captured from indoor and outdoor HLC and corresponding human landing rates (HLR= bites/person/12- hour evening collection period) in August 2007 from 2 coastal and 2 interior localities in West Sumba District, Indonesia.

Species	Lamboyadete			Patialabawa			Tenateke			Radamata			Total
	Interior			Coastal			Upland interior			Coastal			
	In	Out	HLR	In	Out	HLR	In	Out	HLR	In	Out	HLR	
An. aconitus	0	0	0	0	0	0	43	52	11.88	0	0	0	95
An. annularis	18	9	3.38	0	1	0.25	1	0	0.13	0	0	0	29
An. barbirostris	2	3	0.63	1	0	0.25	3	7	1.25	0	0	0	16
An. flavirostris	0	0	0	0	0	0	5	6	1.38	0	0	0	11
An. hyrcanus gr	0	1	0.13	0	0	0	0	0	0	0	0	0	1
An. kochi	5	4	1.13	0	0	0	2	2	0.5	0	0	0	13
An. leucosphyrus gr	0	0	0	0	0	0	1	0	0.13	0	0	0	1
An. maculatus	0	1	0.13	0	0	0	1	4	0.63	0	0	0	6
An. subpictus	131	99	28.75	35	25	15	0	0	0	1	2	0.75	293
An. sundaicus	0	1	0.13	203	181	96	0	0	0	0	0	0	385
An. tessellatus	1	2	0.38	1	0	0.25	0	1	0.13	0	0	0	5
An. vagus	113	82	24.38	8	4	3	0	1	0.13	0	0	0	208
Total	270	202		248	211		56	73		1	2		1,063

Collection periods: Lamboyadete 2 nights (7-8 Aug); Patialabawa 1 night (9 Aug); Tenateke 2 nights (13-14 Aug); Radamata 1 night (15 Aug); In, indoor; out, outdoor; HLR, human landing rate

Table 3
Number and percent of female culicine (*Aedes*, *Armigeres*, *Culex* and *Ochlerotatus*) species collected by method in August 2007 from 2 coastal and 2 interior localities in West Sumba District, Indonesia.

	Coastal							Interior							
	Baited			Unbaited				Baited			Unbaited				Grand
	Tent (%)	Malaise (%)	HLC (%)	Tent (%)	Malaise (%)	Light (%)	Total (%)	Tent (%)	Malaise (%)	HLC (%)	Tent (%)	Malaise (%)	Light (%)	Total (%)	total (%)
Ae. aegypti	0	0	0.1	0	0	0	0.1	0	0	0.8	0	0	0	0.8	9
Oc. poicilius	0	0	0.1	0	0.1	0	0.2	0	0	0	0	0	0	0	3
Ae. vexans	0.2	0	0.2	0.2	0.1	2.6	3.3	0	0.1	0.6	0.2	0.3	0.5	2.7	81
Oc. vigilax	0	0	0	0	0	0.2	0.2	0	0	0	0	0	0	0	3
Ar. malayi	0	0	0.1	0	0	0	0.1	0	0	0	0	0	0	0	2
Cx. bitaeniorhynchus	0	0	0.1	0	0	0.3	0.4	0	0	0.6	0	0	0.2	1.2	17
Cx. fuscocephala	0	0	0	0	0	1.1	1.1	0	0.3	1.0	0.2	0	3.6	12.2	127
Cx. gelidus	0	0	0	0	0	0.1	0.1	0	0	0.1	0	0	0.1	0.3	4
Cx. pseudovishnui	0	0.6	0.6	0.2	0.2	1.9	3.7	0.2	0	6.3	0	0	0.6	8.3	137
Cx. sinensis	0	0.2	0	0	0	0.2	0.4	0	0	2.0	0	0	0	2.0	24
Cx. sitiens	0.4	0.2	0.4	0	0	1.3	2.3	0	0	0	0	0	0	0	39
Cx. tritaeniorhynchus	0.1	0.1	0	0	0.1	12.4	12.7	0.1	0.1	1.6	0.3	0.2	1.5	6.8	278
Cx. vishnui	24.1	0.9	11.5	2.9	0.8	35.6	75.7	1.6	0.6	50.8	1.2	0.8	3.6	65.7	1,888
Total (*N*)	426	34	227	57	23	957	1,724	17	10	566	18	12	265	888	2,612

during 6 trap nights. Total anopheline species captured by location and collection method are presented in Table 1 and dates of collection(s) for each site provided in Table 2. *Anopheles* mosquitoes belonging to 13 species were identified as: *An. aconitus* Döenitz, *An. annularis* Van der Wulp, *An. barbirostris* Van der Wulp, *An. flavirostris* (Ludlow), Hyrcanus Group species, *An. indefinitus* (Ludlow), *An. kochi* Döenitz, *An. leucosphyrus* group Döenitz, *An. maculatus* s.l. Theobald, *An. subpictus* s.l. Grassi , *An. sundaicus* s.l. (Rodenwaldt), *An. tessellatus* Theobald, and *An. vagus* Döenitz. Definitive species identification of the single Hyrcanus Group female mosquito was not possible because of specimen damage. Of potential disease vectors, *An. annularis*, *An. subpictus*, and *An. vagus* were the most frequently collected species in the upland interior sites and *An. sundaicus*, *An. subpictus*, and *An. vagus* were predominant along the coast. The most common species from evening human-landing collections were *An. subpictus* and *An. vagus* in the upland interior and *An. sundaicus* along the coast. The numbers and percentages of non-anopheline (culicine) species captured by trapping method in the four locations are presented in Table 3.

The HLR for *An. subpictus, An. vagus,* and *An. annularis* were 28.75, 24.38, and 3.38, respectively at Lamboyadete (Table 2). Inland species with the greatest HLR also exhibited high to moderate endophagy. For coastal Patialabawa, the HLR were 96, 15, and 3 for *An. sundaicus*, *An. subpictus*, and *An. vagus*, respectively. *Anopheles sundaicu*s, a major malaria vector in many coastal locations in western Indonesia, was found to exhibit slightly stronger endophagy (0.54 total number *An. sundaicus* collected indoors/total *An. sundaicus* collected) compared to outdoor biting in Patialabawa.

The distribution frequency of the predominant species by time of day HLC (combined indoor and outdoor collections) is presented in Fig 2 (a-c). In Lamboyadete, *An. vagus* had a distinct early evening peak between the hours of 18.00 and 20.00 h, whereas *An. annularis* showed nearly the opposite behavior with peak activity seen between 02.00 and 04.00 h. *An. subpictus*, the most common species captured, also showed an early evening peak similar with *An. vagus*. All 3 species were captured throughout the entire evening. In Tenateke, *An. aconitus* showed much stronger activity the first half of the night, except at 21.00-22.00 h. In Patialabawa, *An. sundaicus* showed a modest peak of activity during the early evening (20.00-21.00 h) and persisted throughout the entire evening. *Anopheles subpictus* demonstrated two distinct peaks, one in the early evening at 20.00-21.00 h and another at 24.00-01.00 h. In all three locations, there was little difference in distribution frequencies between indoor and outdoor collections. These apparent patterns of evening activity may have been strongly influenced by the limited collection period and prevailing environmental conditions at the time of collection.

Human landing collections and CDC light traps collected more *Anopheles* species compared to tent and malaise traps. For sites with two collection nights, humans were rotated between trapping locations and devices, however there is the possibility of collection bias due to varying individual host attractiveness. CDC light traps were intentionally hung near animal enclosures which likely accounts for the high numbers of zoophilic vectors (*An. annularis, An. subpictus,* and *An. vagus*) captured. Conversely, *An. sundaicus* was collected in larger numbers using HLC and baited tent traps demonstrating this

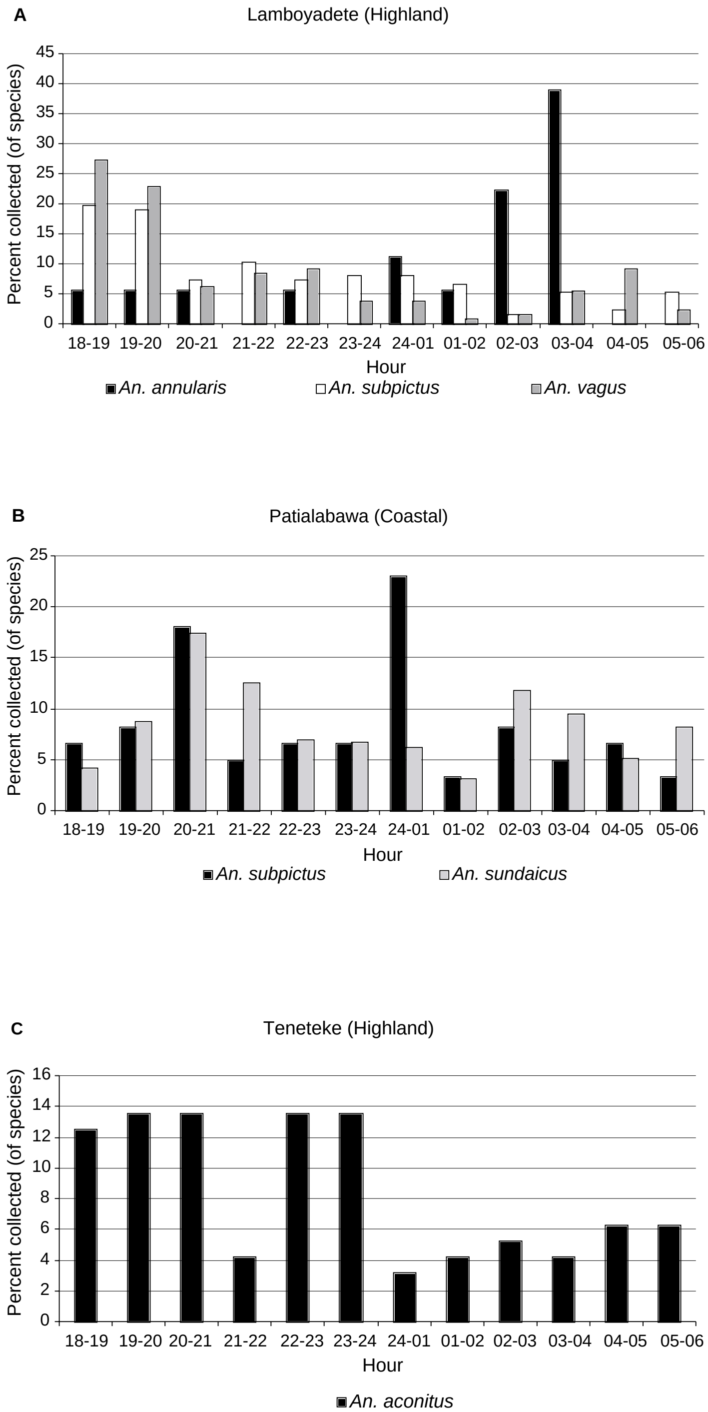

Fig 2–(A-C) Proportion of predominant human-landing mosquitoes by species, hour of capture and location from combined indoor and outdoor collections.

species's stronger anthropophilic/phagic proclivity.

DISCUSSION

All 5,786 *Anopheles* tested using the CSP-ELISA were found non-reactive for presence of malaria sporozoites. August is the dry season on Sumba, a period when malaria transmission typically declines to hypoendemic levels in most locales. Syafruddin *et al* (2009) conducted an extensive and nearly concurrent investigation of malaria seasonal prevalence in West Sumba and found nearly 5% of the human population infected in August 2007. The prevailing dry conditions contributing to likely lower transmission potential during this period may, in part, help explain the apparent absence of malaria-positive mosquitoes in collections.

There have been few published mosquito surveys in the Lesser Sunda Archipelago, and most of these are decades old. O'Connor and Sopa (1981) provided an overview of anopheline and culicine distribution (by province) in Indonesia based on pre-1981 published literature and specimen material available at the Directorate of Disease Control and Environmental Health (P2M) Jakarta. All *Anopheles* species collected in Sumba during August 2007 have been previously reported in the Lesser Sunda island chain. The *Anopheles* species encountered are consistent with collections from Bali and Lombok (Bonne-Webster and Swellengrebel, 1953; Lee *et al*, 1983; Olson *et al*, 1985; Miyagi *et al*, 1994; Kawada *et al*, 2004; Maekawa *et al*, 2009a). For example, Lee *et al* (1983) using CDC light traps and resting collections found *An annularis*, *An. subpictus* and *An. vagus* the predominant species in rice-growing areas of Bali. From 2001 to 2004, Maekawa *et al* (2009a) identified 11 anopheline species on Lombok, all of which were found on Sumba. The one minor exception is the reference to *An. balabacensis* presence on Lombok, whereas we have chosen to designate the Sumba specimen as a member of the Leucosphyrus Group. Closer to Sumba, multi-year longitudinal mosquito larval and adult collections in coastal western Sumbawa Island are also consistent with the Sumba findings (MJ Bangs, unpublished reports).

The results of this survey, combined with known disease vectors in Indonesia (Takken and Knols, 1991) suggest that *An. sundaicus* and *An. subpictus* are likely the primary vectors of malaria on Sumba. However, further indepth vector incrimination studies are required to elucidate this with more defiance. With several exceptions, most other anopheline species captured were of very low density and possibly less anthropophilic. For example, *An. barbirostris*, an important vector of malaria and lymphatic filariasis on nearby islands of Flores, Timor and Alor was poorly represented in total and human-landing collections.

An. aconitus was found to be common in one upland interior location and virtually absent from the coastal areas surveyed during two nights of HLC. This species also showed no strong preference between outdoor and indoor activity. This species is regarded as an important malaria vector in Java (Chow *et al*, 1959; NAMRU-2 unpublished data) and may play a role in malaria transmission in the interior of Sumba, especially in areas with extensive rice cultivation.

Of interest were the relatively high densities of *An. annularis* captured in both coastal and interior locations of Sumba compared to nearby Sumbawa; however, this may reflect more the different trapping methods used or seasonal effects than location. Lee *et al* (1983) also reported relatively high densities of *An. annularis* in Bali from CDC light trap and natural resting collections. In Sumba, the unbaited light traps collected the majority (> 95%) of this species compared to the other methods. *Anopheles annularis* does not appear to play an important role in malaria transmission in Indonesia and is considered to be mostly zoophagic (Bonne-Webster and Swellengrebel, 1953; Takken and Knols, 1991). However, if the vector is competent and found in high biting densities, its epidemiological importance may increase markedly (Ghosh *et al*, 1985; Baker *et al*, 1987; Somboon *et al*, 1994).

From combined trapping methods in coastal and inland areas, *An. vagus* was the most commonly captured anopheline mosquito (38.2%), followed by *An. subpictus* (25.1%). *Anopheles vagus* also presented with a relatively high indoor-outdoor HLR in Lamboyadete (24.38 mosquitoes per human/night), only exceeded by *An. subpictus* (Table 2). *Anopheles vagus* had a greater proportion of female mosquitoes captured indoors, as with *An. annularis*, if found in high enough biting densities, this species may act as a secondary malaria vector (Baker *et al*, 1987; Somboon *et al*, 1994). *Anopheles vagus* specimens have been found infected with human plasmodia on Timor Island (near Kupang City) and central Java (Purworejo, NAMRU-2 unpublished report). Incidentally, Japanese encephalitis virus was isolated from *An. annularis* and *An. vagus* on Lombok Island; however, the epidemiological significance of that finding is unknown (Olson *et al*, 1985). Given the predominately zoophilic nature of this species, it is not considered a disease vector of high importance.

Comparisons between human-baited and unbaited tent and malaise traps found baited traps far more effective (2.8 to 4.3-fold greater for tent and malaise traps, respectively) in attracting mosquitoes in coastal Patialabawa, whereas baited traps either proved less effective or near equivalent in the interior locations (Table 1). As these findings and comparisons are based on only 1 or 2 collection nights, further assessment of trapping methods is needed. Radamata and Tenateke produced far fewer mosquitoes than the other two locations. Due to the brevity of this survey, findings were likely more susceptible to chance environmental factors and natural variability in mosquito behavior, both important factors that may have impacted collection results. For example, phase of moonlight may have had a significant impact on mosquito activity (Bidlingmayer 1964). The collections in Tenateke and Radamata took place during a new moon, whereas the 2 sites with significantly higher collection success occurred during the waning last quarter phase.

One specimen of the *An. leucosphyrus* group was collected from indoor HLC in the upland site (Tenateke Village). This appears to represent a new collection record for Sumba Island and only the third island in the Nusa Tenggara region east of the Wallace Line. This species group, which includes *An. balabacensis*, has also been collected on Lombok (Miyagi *et al*, 1994; Kawada *et al*, 2004) and western Sumbawa (MJ Bangs, unpublished report, 2005; Maekawa *et al*, 2009b). For those identified as *An. leucosphyrus* group (Miyagi *et al*, 1994; Bangs, unpublished report, 2005), only a single adult female specimen was recorded from each collection. The Lombok specimen was collected from a rock pool along a stream in a forested area ~600 m elevation above sea level near Mt. Rinjani and the Sumbawa female from a HLC in a heavily forested, remote mountain location at 350 m above sea level. A single adult female and an unspecified number of *An. balabacensis* larvae that were also collected in a rock pool in Sumbawa were also found in forested areas (Maekawa *et al*, 2009b). The limited amount of material and recorded morphological variability indicates molecular genetic analysis will be needed to help clarify the precise identity of these species and their distribution on the three islands.

We acknowledge that the initial observations presented are limited on number of collections and sites; however, this represents the first vector survey of its kind on the island of Sumba and serves as a beginning for future studies. Many of the anopheline species identified in this study have been incriminated as malaria vectors elsewhere in Indonesia and Southeast Asia. Correctly identifying those mosquito species in West Sumba that play a role as either primary or secondary disease vectors will focus vector control efforts. Additionally, molecular level identification of specific members, or as yet undescribed new members, within species complexes is essential to pinpoint those mosquitoes involved in transmission. Linking vector identification with species-specific bionomic and epidemiologic factors will require further investigation in the field and laboratory.

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REFERENCES

Asih PB, Rogers WO, Susanti AI, *et al.* Seasonal distribution of anti-malarial drug resistance alleles on the island of Sumba, Indonesia. *Malar J* 2009; 8: 222-7.

Baker EZ, Beier JC, Meek SR, Wirtz RA. Detection and quantification of *Plasmodium falciparum* and *P. vivax* in Thai Kampuchean *Anopheles* (Diptera: Culicidae) by enzyme linked-immunosorbent assay. *J Med Entomol* 1987; 24: 537-41.

Bidlingmayer WL. The effect of moonlight on the flight activity of mosquitoes. *Ecology* 1964; 45: 87-94.

Bonne-Webste J, Swellengrebel NH. The anopheline mosquitoes of the Indo-Australian region. Amsterdam: JH de Bussy, 1953.

Chow CY, Ibnoe MR, Josopoero ST. Bionomics of anopheline mosquitoes in inland areas of Java, with special reference to *Anopheles aconitus* Dön. *Bull Entomol Res* 1959; 50: 647-60.

Ghosh KK, Chakraborty S, Bhattacharya S, Palit A, Tandon N, Hati AK. *Anopheles annularis* as a vector of malaria in rural West Bengal. *Indian J Malariol* 1985; 22: 65-9.

Kawada H, Maekawa Y, Tsuda Y, Takagi M. Laboratory and field evaluation of spatial repellency with metofluthrin-impregnated paper strips against mosquitoes in Lombok Island, Indonesia. *J Am Mosq Control Assoc* 2004; 20: 292-8.

Lee V, Atmosoedjono S, Rusmiarto S, Aep S, Semendra W. Mosquitoes of Bali Island, Indonesia: common species in the village environment. *Southeast Asian J Trop Med Public Health* 1983; 14: 298-307.

Maekawa Y, Tsuda Y, Dachlan YP, *et al.* Anopheline fauna and incriminatory malaria vectors in malaria endemic areas on Lombok Island, Indonesia. *Med Entomol Zool* 2009a; 60: 1-11.

Maekawa Y, Sunahara T, Dachlan YP, *et al.* First record of *Anopheles balabacensis* from western Sumbawa Island, Indonesia. *J Am Mosq Control Assoc* 2009b; 25: 203-5.

Miyagi I, Toma T, Mogi M, *et al*. Mosquito species (Diptera: Culicidae) from Lombok Island, Indonesia. *Mosq Syst* 1994; 12: 19-24.

O'Connor CT, Sopa T. A checklist of the mosquitoes of Indonesia. Special publication of the US Naval Medical Research Unit no. 2, Jakarta, Indonesia. *NAMRU-SP* 1981: 45.

O'Connor CT, Soepanto. Illustrated key to female anophelines of Indonesia (revised by Atmosoedjono S and Bangs MJ, NAMRU-2). Jakarta: Directorate of Communicable Disease, Ministry of Health Indonesia, 1989.

Olson JK, Ksiazek TG, Lee VH, Tan R, Shope RE. Isolation of Japanese encephalitis virus from *Anopheles annularis* and *Anopheles vagus* in Lombok, Indonesia. *Trans R Soc Trop Med Hyg* 1985; 79: 845-7.

Reid JA. Anopheline mosquitoes of Malaya and Borneo. *Stud Inst Med Res Malaya* 1968; 31: 353-60.

Reuben R, Tewari SC, Hiriyan J, Akiyama J. Illustrated keys to species of *Culex* (*Culex*) associated with Japanese encephalitis in Southeast Asia (Diptera: Culicidae). *Mosq Syst* 1994; 26: 75-96.

Silver JB. Mosquito ecology: field sampling methods. 3rd ed. Dordrecht: Springer, 2008: 1516 pp.

Somboon P, Suwonkerd WW, Lines JD. Susceptibility of Thai zoophilic anophelines and suspected malaria vectors to local strains of human malaria parasites. *Southeast Asian J Trop Med Public Health* 1994; 25: 766-70.

Stoops A, Gionar Y, Rusmiarto S, *et al.* Laboratory and field testing of bednet traps for mosquito (Diptera: Culicidae) collection in West Java, Indonesia. *J Vector Ecol* 2010; 35: 187-96.

Syafruddin D, Asih PB, Coutrier FN, *et al.* Malaria in Wanokaka and Loli sub-districts, West Sumba District, East Nusa Tenggara Province, Indonesia. *Am J Trop Med Hyg* 2006; 74: 733-7.

Syafruddin D, Krisin P, Asih B, *et al.* Seasonal prevalance of malaria in West Sumba district, Indonesia. *Malaria J* 2009; 8: 8-15.

Takken W, Knols BGJ. A taxonomic and bionomic review of the malaria vector of Indonesia. In: Takken W, Snellen WB, Verhave JP, Knols BGJ, Atmosoedjono S, eds. Environmental measures for malaria control in Indonesia - An historical review on species sanitation. Wageningen: Wageningen Agricultural University Press, 1991: 9-62.

World Health Organization (WHO). Manual on practical entomology in malaria. Part II. Geneva: WHO, 1975.

Wirtz RA, Burkot TR, Graves PM, Andre RG. Field evaluation of enzyme-linked immunosorbent assays for *Plasmodium falciparum* and *Plasmodium vivax* sporozoites in mosquitoes (Diptera: Culicidae) from Papua New Guinea. *J Med Entomol* 1987; 24: 433-7.

Wirtz RA, Sattabongkot J, Hall T, Burkot TR, Rosenberg R. Development and evaluation of an enzyme-linked immunosorbent assay for *Plasmodium vivax*-VK247 sporozoites. *J Med Entomol* 1992; 29: 854-7.