

INVESTIGATION OF MOSQUITOES WITH EMPHASIS ON *Aedes (Finlaya) poicilius*, PUTATIVE VECTOR OF BANCROFTIAN FILARIASIS ON PANAY ISLAND, THE PHILIPPINES

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Abstract. Entomological investigations were conducted in four remaining lymphatic filariasis endemic provinces of Panay Island, The Philippines to determine mosquito species present in these areas and to identify host preference and biting activity of *Aedes (Finlaya) poicilius* Theobald, primary vector of nocturnal periodic *Wuchereria bancrofti* in The Philippines. Sampling techniques targeted nocturnally active mosquito species using a carabao-baited trap (CBT) and human-landing collection (HLC), the latter taking place from 06:00 to 12:00 PM. A total of 25,536 mosquitoes comprising 42 species and 7 genera were collected from CBT, whilst HLC acquired 6,486 mosquitoes comprising 28 species and 5 genera. Three known or potential vectors of human filarial were collected, namely, *Aedes poicilius*, *Culex quinquefasciatus* and *Mansonia uniformis*. The peak landing (biting) activity for *Ae. poicilius* was between 09:00 and 11:00 PM. Comparisons between CBT and HLC yields showed this species to be more zoophilic. Based on observed mosquito behavior and interviews with residents, vector-host contact was promoted by the local practice of staying overnight in makeshift shelters in high risk areas without adequate protection against mosquito bites. Results of this survey will augment information for integrating vector control and mass drug administration into an island-wide lymphatic filariasis elimination program.

Keywords: *Aedes poicilius*, behavior, lymphatic filariasis, mosquito species, Panay Island, The Philippines

INTRODUCTION

Despite decades of efforts to control lymphatic filariasis, it still remains a public health concern in The Philippines (Ca-

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brera and Arambulo, 1973; Cabrera and Valeza, 1978; Hernandez, 1993, Amarillo *et al*, 2008; DOH, 2008). Both *Wuchereria bancrofti* and *Brugia malayi* are present in the country; however, only autochthonous Bancroftian filariasis is known to occur in Panay Island. Since 2015, there were 44/81 provinces in the country that have been designated as 'filarial-free' while the remaining 33 provinces are currently

under an elimination phase. The remaining four provinces from Region VI, which comprises the remaining four provinces on Panay Island, namely, Aklan, Antique, Capiz and Iloilo, are under a control category (DOH, 2013).

In 2007, a serendipitous detection of microfilaria from a patient in Iloilo City subsequently resulted in blood surveys among residents in areas suspected of residual infection or active transmission (Pendon, 2012; DOH, 2013). The surveys yielded several patent human infections in three municipalities of Iloilo, *ie*, Concepcion, Janiuay and Lambunao. Nighttime blood surveys were extended to other provinces of Panay Island and additional filaria cases were detected in the municipalities of Bugasong and Valderama in Antique Province; Libacao, Madalag and Malinao in Aklan Province; and Tapaz in Capiz Province (<http://balita.ph/2009/11/27/health-authorities-strengthen-campaign-vs-lymphatic-filariasis/>, 2009). All filarial infections revealed only *W. bancrofti*.

Aedes (Finlaya) poicilius Theobald (Knight and Stone, 1977; Wilkerson *et al*, 2015) is the principal vector of Bancroftian filariasis in The Philippines, especially in areas where abaca and banana cultivations are abundant (Suguri *et al*, 1985; Schultz, 1988; Hernandez, 1993; Walker *et al*, 1998). As for other mosquito-borne diseases of importance, with the exception of Antique Province (Semirara Island), Panay Island is regarded as malaria-free with no reported indigenous cases in the previous 5-10 years, placing Panay Island in the pre-elimination phase of control (DOH, 2014).

In order to further investigate the risk of filariasis transmission on Panay Island, entomological surveys in the four above-

mentioned provinces were conducted from September 2011 to October 2012 with the following objectives: (1) identification of potential filariasis mosquito vectors in areas with positive human cases, (2) determination of distribution of mosquito species with emphasis on filariasis vectors, and (3) determination of vector behavior and bionomics contributing to filariasis transmission in each area.

Entomological surveys of this type contribute substantially to the Philippine National Filariasis Elimination Program towards the development of site-specific, evidence-based, integrated vector control strategies (DOH, 2013). Although the vector species on Panay Island may be similar to other areas in The Philippines, site-specific spatial variation in mosquito behavior may be present, which might be critical to the understanding of disease transmission and implementation of cost-effective vector control measures. Moreover, a better understanding of local community activities, which may promote transmission could be targeted to minimize vector-human contact.

MATERIALS AND METHODS

Study sites and activities

The island of Panay (11° 09'N, 122° 29'E) is in Western Visayas Region of The Philippines and comprises four provinces, namely, Aklan, Antique, Capiz and Iloilo, on the main island and Guimaras Island, southeast of Panay Island. Panay is surrounded by various bodies of sea and the land mass is characterized by relatively wide stretches of coastal lowlands combined with rugged hills and mountains in the interior. Panay possesses a diverse marine life, inland fisheries and various agricultural products. Its large agricul-

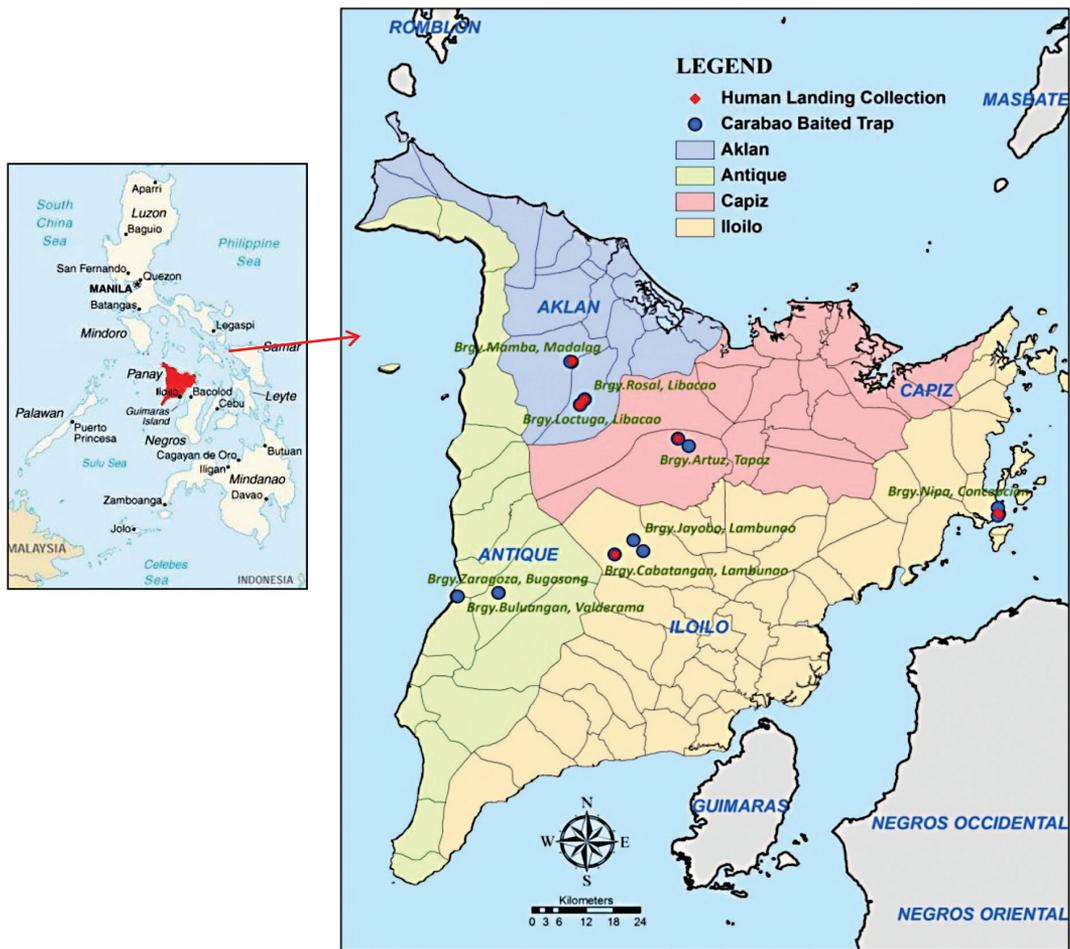


Fig 1–Map of mosquito collection sites in the four provinces of Panay Island, The Philippines.

tural areas produce high quantities of rice, coconut, mango, pineapple, banana, various root crops, vegetables and sugar. Abaca and rattan crafts are also important industries.

Collection sites were chosen based on the presence of filariasis positive cases, accessibility and security. Working closely with the regional entomology staff and the Filaria Program Coordinator of Department of Health, Regional Health Office - Region VI, candidate collection sites were identified and representative mosquito collection sites in Aklan, Antique, Capiz and Iloilo Provinces (Fig 1)

were sampled for 2 nights per month from September 2011 to October 2012. Entomological sampling was conducted in locations that were identified as filarial positive by active case detection surveys completed in 2007-2009 (Pendon, 2012; DOH, 2013). These included Aklan Province (Barangays Loctuga and Rosal from the municipality of Libacoo and Brgy Mamba from the municipality of Madalag); two municipalities from Antique Province, namely Bugasong (Brgy. Zaragoza) and Valderama (Brgy. Buluangan I); the municipality of Tapaz in Capiz Province (Brgy. Artuz and Bacahan); and

municipalities of Concepcion (Sitio Dadoy and Sitio Puntalis from Brgy. Nipa) and Lambunao (Barangays Jayobo and Cabatangan) from Iloilo Province.

The primary goal was to conduct vector collections in and around the primary residence and/or work area of known filariasis-positive cases and interview key informants, such as local health workers, barangay (village) health workers, *kagawads* (elected councilor for the smallest political unit in The Philippines, the barangay), and midwives to gain a better understanding of the local cultural habits and behavioral patterns.

All mosquito collections were performed in locations that are relatively flat landscapes between hills with open canopy transition forest. Other vegetation types in the near vicinity of collections included grass, rice, low shrub, bamboo, scattered trees, abaca ('manila hemp', *Musa textilis*), banana (*Musa* spp), and *Pandanus*. Potential mosquito larval habitats, such as creeks and streams, ponds, ground depressions, rice fields and irrigation canals were inspected for immature stages of mosquitoes around the immediate collection sites (within 100 m).

GPS coordinates of all mosquito collection sites (Table 1) for adults and immature samples were recorded using a Garmin™ 76C GPS unit (Garmin, Olathe, KS). All sites were mapped using ArcGIS 9.1 (2005 software) (ESRI®, Redlands, CA). Coordinates will serve as future reference should there be a need to revisit these sites. Habitat description of each collection site was recorded as baseline reference to account for changes in the environment over time.

Collection of mosquito larvae

Collections of mosquito larvae covered a wide range of possible breeding sites

within the vicinity of primary residences of filaria-positive cases (DOH, 2013). Known filariasis vector species in the country belong within four genera (*Aedes*, *Anopheles*, *Culex*, and *Mansonia*), each with specific breeding site preferences; therefore all possible sources of mosquito larvae were inspected using various techniques (WHO, 1975). Inspected breeding sites include margins of slow moving creeks, streams, irrigation canals, ponds, ground pools, roots of aquatic plants and natural containers. Particular attention was paid to sampling plant leaf axils, such as abaca, banana, *Pandanus*, taro (*Colocasia esculenta*) and other similar plants that are the preferred breeding habitats of *Aedes poicilius*.

Collection of mosquitoes by carabao-baited traps (CBT)

Animal-baited traps generally collect a wider diversity and number of mosquito species than most other collection methods per trapping session and is dependent on host size, number and defense behavior (Silver, 2008). CBT was composed of white, nylon (non-insecticide-treated) mosquito net (4 m x 3 m x 3 m in dimension). A door panel was provided on one side measuring 1 m x 3 m. The white color of the net contrasts with the dark mosquito coloration thereby assisting the collector to locate and capture more efficiently those resting on the net surface.

A minimum of one trap-night per collection site was conducted. CBT was set-up in a relatively flat area and temporarily secured to poles or trees and pegs gathered from around the collection site. The open panel of the net was positioned in areas that face possible breeding sites (eg, surface bodies, streams, abaca plantation, or dense vegetation). The side panel of CBT was weighed down with rocks or other articles to minimize detrimental

Table 1
GPS coordinates of mosquito collection sites of entomological survey of Panay Island,
The Philippines, September 2011 to October 2012.

Collection site	Month of collection	Collection type	Latitude	Longitude
Jayobo Proper, Lambunao, Iloilo	Sep 2011	CBT	11°05'53.34"	122°24'56.5"
Tagbakan, Jayobo, Lambunao, Iloilo	Sep 2011	CBT	11°07'14.6"	122°23'46.2"
Cabatangan Proper, Lambunao, Iloilo	Nov 2011	HLC	11°05'29.3"	122°21'32.7"
Cabatangan Proper, Lambunao, Iloilo	Nov 2011	CBT1	11°05'28.9"	122°21'31.6"
Sitio Dadoy, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	CBT1	11°10'29.06"	123°07'28.96"
Sitio Dadoy, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	HLC1	11°10'27.01"	123°07'29.35"
Nipa Proper, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	CBT2	11°10'15.67"	123°07'20.71"
Nipa Proper, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	HLC2	11°10'36.25"	123°07'19.85"
Sitio Puntalis, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	CBT3	11°11'14.35"	123°07'21.54"
Sitio Puntalis, Brgy. Nipa, Concepcion, Iloilo	Apr 2012	HLC3	11°10'36.3"	123°07'19.8"
Artuz Proper, Tapaz, Capiz	Sep 2012	CBT1	11°19'43.8'	122°29'05.8'
Artuz Proper, Tapaz, Capiz	Sep 2012	HLC1	11°19'45.8'	122°29'04.8'
Bacahan, Brgy. Artuz, Tapaz, Capiz	Sep 2012	CBT2	11°18'50.9'	122°30'24.1'
Brgy. Zaragoza, Bugasong, Antique	Jul 2012	CBT1	11°00'20.2"	122°02'45.2"
Brgy. Zaragoza, Bugasong, Antique	Jul 2012	CBT2	11°00'19.8"	122°02'39.2"
Brgy. Zaragoza, Bugasong, Antique	Jul 2012	CBT1	11°00'20.1"	122°02'45.3"
Brgy. Buluangan I, Valderrama, Antique	Jul 2012	CBT1	11°00'45.2"	122°07'38.6"
Brgy. Buluangan I, Valderrama, Antique	Jul 2012	CBT2	11°00'45.3"	122°07'36.0"
Brgy. Rosal, Libacao, Aklan	Oct 2012	HH w/case	11°24'34.8"	122°17'55.0"
Brgy. Rosal, Libacao, Aklan	Oct 2012	CBT1	11°24'35.4"	122°17'57.7"
Brgy. Rosal, Libacao, Aklan	Oct 2012	CBT2	11°24'33.9"	122°17'54.9"
Brgy. Rosal, Libacao, Aklan	Oct 2012	HLC1	11°24'30.8"	122°17'48.6"
Brgy. Rosal, Libacao, Aklan	Oct 2012	HLC2	11°24'32.6"	122°17'50.7"
Brgy. Rosal, Libacao, Aklan	Oct 2012	HLC3	11°24'35.7"	122°17'55.8"
Brgy. Luctoga, Libacao, Aklan	Oct 2012	CBT1	11°23'55.9"	122°17'23.0"
Brgy. Luctoga, Libacao, Aklan	Oct 2012	CBT2	11°23'54.6"	122°17'23.2"
Brgy. Luctoga, Libacao, Aklan	Oct 2012	HLC1	11°23'52.1"	122°17'20.3"
Brgy. Luctoga, Libacao, Aklan	Oct 2012	HLC2	11°23'52.6"	122°17'17.5"
Brgy. Luctoga, Libacao, Aklan	Oct 2012	HLC3	11°23'52.9"	122°17'17.2"
Mamba Proper, Brgy. Madalag, Aklan	Oct 2012	CBT1	11°29'13.1"	122°16'22.2"
Sitio Tagpi, Brgy. Mamba, Madalag, Aklan	Oct 2012	CBT2	11°29'15.1"	122°16'13.6"
Sitio Tagpi, Brgy. Mamba, Madalag, Aklan	Oct 2012	HLC1	11°29'17.3"	122°16'26.9"
Sitio Tagpi, Brgy. Mamba, Madalag, Aklan	Oct 2012	HLC2	11°29'18.9"	122°16'27.6"
Sitio Tagpi, Brgy. Mamba, Madalag, Aklan	Oct 2012	HLC3	11°29'19.1"	122°16'23.3"

CBT, carabao-baited trap; HLC, human-landing collection; HH w/case -meaning households with member/s who contracted filariasis before the conduct of entomological investigation.

impact from strong winds. A local adult water buffalo (*Bubalus bubalis*) or carabao served as the animal bait and was readily available in the farming communities

sampled. These docile animals are ideal baits and can attract a wider range of mosquito species in higher numbers than most other collection methods. The panel was

left open overnight while a carabao was tethered in the middle of the netted space from 06:00 PM to 04:00 AM of the following morning. After removal of the carabao in the morning, the panel was lowered and secured by Velcro® strips to prevent the trapped mosquitoes from escaping before collection began at sunrise.

Mosquitoes were collected individually from CBT using a mouth aspirator and placed in a labeled collection cup. Mosquitoes were initially sorted by relative size and resting position and placed in separate collection cups. The open end of cups were covered with a polyester netting material, which then secured with a rubber band. A 1 cm hole in the middle of the netting allowed transfer of mosquitoes in and out of the cup using an aspirator; otherwise the hole was kept closed with a plug of cotton wool. Sugar-solution soaked cotton balls were placed on top of the netting for mosquitoes to freely feed upon. The cotton balls were held in place by another piece of netting secured over the bottom layer of netting with a rubber band.

Human-landing collection (HLC) of mosquitoes

In order to determine mosquito species that are attracted to humans and thus make them more likely disease vectors, HLC was performed inside and outside of occupied houses for half night collections from 06:00 to 12:00 PM. From each barangay, 1 to 3 houses were sampled by each of the assigned local volunteer collector. Each volunteer signed an informed consent before being enrolled in the study. At each collection site, indoor and outdoor household stations were chosen based on the presence or proximity to households of recent filaria-positive cases. Hourly collections from each

station were recorded to determine frequency of activity and peak biting time of mosquitoes attempting to blood feed on the collector. Individual collection cups were labeled for each hour interval (eg, 06:00 to 07:00 PM), house number, indoor or outdoor collection site, and name of collector. All information was recorded on written field data sheets and entered electronically at a later time.

Mosquito identification

All collected mosquitoes were killed by freezing or by exposure to direct sunlight (at locations lacking electricity) while still in the collection cups. Contents of each cup were examined under a stereomicroscope and mosquitoes were sorted according to genera and species using morphological criteria, with each cup being processed separately. Final sorting as to collection method, place, date and time was made prior to transport to RITM, Alabang, Muntinlupa City, The Philippines.

Collected larvae were reared to adults, which were identified using standard morphological characteristics. Identification of *Anopheles* larvae and adults followed the morphological key of Cagampang-Ramos and Darsie (1970). Species of other genera were identified using a series of mosquito taxonomic keys for Thailand (Rattanarithikul *et al*, 2005a; *ibid*, 2005b; *ibid*, 2006; *ibid*, 2010) in combination with known distribution records for The Philippines (Apiwathnasorn, 1986). Specimens were further analyzed using pictorial keys from the Walter Reed Biosystematics Unit (<http://wrbu.si.edu/VecIDResourcesMQ.html>).

RESULTS

Social factors influencing vector contact

Interviews with local health workers,

Table 2
 Female mosquito species collected using carabao-baited traps (CBT) on Panay Island, The Philippines, September 2011 to October 2012.

Mosquito species	Iloilo Province			Capiz Province		Aklan Province			Antique Province							
	Cabatangan	Jayobo	Tagbakan	Dadoy	Nipa	Concepcion	Artuz	Bacahan	Loctuga	Mada-lag	Rosal	CBT1	CBT2	CBT3	Valderama	
<i>Aedes</i> spp ^a	1 ^b	---	---	---	---	---	---	---	---	---	---	---	4	15	15	---
<i>Ae. albopictus</i>	1	---	---	---	---	---	4	2	---	5	---	1	---	1	---	---
<i>Ae. poicilius</i>	27	44	127	1	---	---	1	31	---	4	1	---	---	---	32	4
<i>Ae. vexans</i>	---	---	---	---	---	---	8	13	---	---	---	---	---	---	---	---
<i>Ae. vigilax</i>	---	---	---	---	---	---	---	---	2	6	---	19	---	---	30	14
<i>Armigeres</i> spp ^a	2	88	3	---	---	---	---	---	---	---	---	---	---	---	---	---
<i>Ar. digitatus</i>	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	1
<i>Ar. jugraensis</i>	---	---	---	---	---	---	2	---	---	---	---	---	---	---	---	---
<i>Ar. kuchingensis</i>	---	---	---	---	---	---	---	---	---	2	---	---	---	---	---	---
<i>Ar. malayi</i>	---	---	---	---	---	---	2	---	---	---	---	---	---	---	---	---
<i>Ar. subalbatus</i>	---	---	---	---	---	---	6	3	---	11	4	3	6	3	3	3
<i>Ar. theobaldi</i>	---	---	---	---	---	---	---	---	---	---	---	---	---	1	1	---
<i>Anopheles</i> spp ^a	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---
<i>An. annularis</i>	42	---	---	---	---	---	6	---	---	15	---	47	78	89	217	64
<i>An. franciscoi</i>	211	5	17	---	---	---	---	---	2	2	1	---	---	---	---	---
<i>An. indefinitus</i>	75	4	11	---	---	---	8	5	---	---	---	2	---	---	---	---
<i>An. karwari</i>	---	---	---	---	---	---	8	---	---	---	---	---	---	---	---	---
<i>An. kochi</i>	56	58	82	---	---	---	2	19	---	17	1	---	1	---	---	---
<i>An. lesteri</i>	---	---	---	---	---	---	---	---	---	---	1	---	---	---	---	---
<i>An. litoralis</i>	---	---	---	249	97	71	---	---	---	---	---	---	---	---	---	---
<i>An. ludlowae</i>	---	---	---	64	37	23	---	---	14	8	11	---	---	---	---	4
<i>An. maculatus</i>	19	11	27	12	9	8	7	31	---	37	---	---	---	2	2	6
<i>An. mangyanus</i>	---	---	---	---	---	---	---	---	1	---	---	---	---	---	---	---
<i>An. pedataeniatus</i>	---	---	---	---	---	---	1	---	---	6	---	---	---	---	---	---
<i>An. pseudobarbitrostris</i>	27	4	5	---	---	---	1	---	---	19	---	---	---	---	---	---
<i>An. samarensis</i>	---	---	---	---	---	---	1	---	---	10	5	2	---	---	---	---

<i>An. subpictus</i>	96	42	59	---	---	---	8	18	---	2	---	7	9	18	3	---
<i>An. tessellatus</i>	132	14	9	---	---	---	2	23	---	1	---	---	---	---	---	---
<i>An. vagus</i>	377	4	27	13	9	14	0	5	---	6	---	7	15	10	---	1
<i>An. vanus</i>	---	---	---	---	---	---	1	20	---	---	---	---	---	---	---	---
<i>Coquilletidia</i> spp ^a	---	1	2	---	---	---	---	---	---	---	---	---	---	---	---	---
<i>Culex</i> spp ^a	2,994	---	---	---	---	---	---	---	---	---	---	140	31	90	254	31
<i>Culex fuscocephala</i>	---	---	---	153	103	96	50	57	16	19	---	---	---	---	---	---
<i>Cx. gelidus</i>	---	---	---	---	---	---	---	5	16	43	1	---	---	---	---	---
<i>Cx. quinquefasciatus</i>	336	---	---	23	29	35	1	2	---	7	---	---	---	---	1	---
<i>Cx. sitiens</i> spp complex	---	---	---	1,115	572	455	34	71	21	60	3	---	---	---	---	---
<i>Cx. tritaeniorhynchus</i>	---	---	---	171	83	22	---	---	---	3	---	---	---	---	---	---
<i>Cx. vishnui</i>	---	---	---	558	477	336	38	156	36	56	---	22	1	2	---	---
<i>Cx. whitmorei</i>	---	---	---	---	---	---	34	225	41	26	---	---	---	6	---	---
<i>Cx. fuscatus</i>	---	---	---	4	---	---	---	---	---	---	---	---	---	---	---	---
<i>Mansonia uniformis</i>	2	11	34	---	---	---	28	1	---	---	---	---	---	---	---	---
<i>Mimomyia hybrida</i>	---	---	---	17	16	14	---	---	---	---	---	---	---	---	---	---
Total	4,398	286	403	2,380	1,432	1,074	249	685	149	365	28	250	145	237	560	127

^aSpecies not identified. ^bNumber of specimens.

recent filarial patients, and other local residents revealed that the common source of livelihood at the collection sites were abaca fiber processing, banana cultivation, copra production, weaving sleeping mats, and forest product gathering, activities requiring residents to access elevated areas with dense vegetation and where potential filaria vectors may be present. Additionally, because of the inaccessibility of many of these areas, residents typically conduct initial processing of materials away from residence with a tendency of staying overnight in makeshift shelters for varying periods of time. Field workers did not have bed nets with them during the harvest of abaca. Their clothes were basic, well-worn short sleeved shirts and cut-off pants, which provide minimal protection from biting mosquitoes. They stayed in make-shift houses that provide some protection from the wind and rain but have many openings and entry points for insects and other host-seeking arthropods to gain access. For the most part, the population did not practice any form of personal protection while away from their primary residences.

Mosquito collections

A total of 25,536 adult female mosquitoes were collected from CBTs, comprising 7 genera and 42 species. HLC resulted in 6,486 mosquitoes, comprising 5 genera and 28 species. Eight known or potential human filarial vectors in The Philippines were collected: *Ae. poicilius*, *Culex quinquefasciatus*, *C. sitiens* complex, *Anopheles maculatus*, *An. subpictus*, *An. tessellatus*, *An. vagus*, and *Mansonia uniformis* (Hernandez 1993; Manguin *et al*, 2010). *Ma. uniformis* has been associated primarily with *Brugia malayi* transmission in Southeast Asia (Sasa, 1976). The peak landing (biting) activity for *Ae. poicilius* was between

09:00 and 11:00 PM. Comparison between CBT and HLC yields indicated that this species fed on both humans and carabao, as well as indoor and outdoor for humans but this varied according to locality.

Mosquito larvae collections

Collections of immature mosquitoes were conducted in both natural habitats and in natural containers. All collections were made during the rainy season with the exception of one collection from banana leaf axil in Nipa (Concepcion, Iloilo) that was conducted during the dry season (April, 2012). Five putative filaria vectors were obtained from the larvae collections: *Ae. poicilius*, *An. maculatus*, *Cx. quinquefasciatus*, *Cx. sitiens* complex, and *Ma. uniformis*. Each of these species has specific breeding site requirements that varied in number and distribution among collection sites. The primary filaria vector, *Ae. poicilius*, was found only in *Musa* spp (abaca and banana) leaf axils, which were common in all areas surveyed. *Mansonia* spp were collected from aquatic plants, such as kangkong (*Ipomea aquatica*) and *Pistia*, while *Ma. uniformis* was found in irrigation canals containing aquatic plants, such as *Ipomea aquatica* (water spinach). *An. maculatus* was found in irrigation canals (intended for rice cultivation) with emerging and floating vegetation, and in slow-moving mud-bed streams lined with riverine rocks. *Cx. quinquefasciatus* typically was found in coconut shells and in edges of stagnant irrigation canals, while *Cx. sitiens* was common in temporary ponds and fresh water-filled ground depressions.

Mosquito collections from CBT

A far greater range of evening-active mosquito species were collected using CBT: 7 mosquito genera representing 42 species (Table 2). *Anopheles* with 17 species

dominated the CBT catch, followed by *Culex* (7 species including *Cx. quinquefasciatus*), *Aedes* (5 species including *Ae. poicilius*) and *Armigeres* (4 species). Presence of *Ae. poicilius* was observed in all sites with approximately 1 to 4 specimens per site. On the other hand, *Ma. uniformis* was not collected in Aklan and Antique while *Cx. quinquefasciatus* was fewest in Antique and Capiz.

Mosquito collections from HLC

Mosquito species that are attracted to humans and therefore are potential vectors of disease were collected using HLC for subsequent identification. HLC allowed measurements of vector distribution relative to human habitation (indoor or outdoor), and peak vector activity in relation to host seeking behavior during evening up to 12:00 PM. HLC stations were set up in and near nipa huts, the most common domiciliary structures in the area. A hut consisted of a nipa palm frond roof, split bamboo walls and bamboo slat floors with open spaces between slats. Wide and open windows were variously placed around the walls of these huts especially in the dining, kitchen and receiving areas.

A total of 28 mosquito species belonging to 5 genera were collected, with *Ae. poicilius*, *An. maculatus*, *An. subpictus*, *An. tessellatus*, *An. vagus*, *Cx. quinquefasciatus*, and *Cx. sitiens* species complex being potential vectors of *W. bancrofti* (Table 3). With the exception of sites in Capiz Province, the percent indoor HLC yield showed a greater abundance of *Cx. quinquefasciatus* compared to the primary vector, *Ae. poicilius* (Fig 3). *Ae. poicilius* was collected at all periods from 06:00 to 12:00 PM, with peak landing activity occurring between 09:00 to 11:00 PM. The average HLC outdoor hourly yield of

Table 3
Female mosquito species collected from human landing collections (HLC) and resting collections (RC) on Panay Island, The Philippines, September 2011 to October 2012.

Mosquito Species	Cabatangan		Concepcion		Capiz	Aklan			Antique	
	HLC	RC	Dadoy	Nipa	Artuz	Luctoga	Rosal	Mada- lag	Buga- song	Valde- rama
<i>Ae. aegypti</i>	---	---	1 ^a	---	---	---	---	3	2	1
<i>Ae. albopictus</i>	---	---	---	---	---	---	---	2	2	1
<i>Ae. poicilius</i>	12	12	---	---	32	8	8	10	1	27
<i>Ae. vexans</i>	---	---	---	---	1	---	---	---	---	---
<i>Ae. vigilax</i>	---	---	---	---	---	1	---	---	---	3
<i>Armigeres spp^b</i>	---	1	---	---	---	---	---	---	---	---
<i>Ar. jugraensis</i>	---	---	---	---	1	---	---	---	---	---
<i>Ar. malayi</i>	---	---	---	1	---	---	---	---	---	---
<i>Ar. moultoni</i>	---	---	---	1	---	---	---	---	---	---
<i>Ar. subalbatu</i>	---	---	---	---	1	16	17	---	9	7
<i>Anopheles spp^b</i>	---	---	---	---	---	---	---	---	2	---
<i>An. kochi</i>	---	---	---	---	4	---	---	2	1	---
<i>An. litoralis</i>	---	---	41	35	---	---	---	---	---	---
<i>An. ludlowae</i>	---	---	---	---	---	---	2	1	---	---
<i>An. maculatus</i>	---	---	---	---	---	---	---	1	1	---
<i>An. pseudobarbistrois</i>	---	1	---	---	---	---	2	2	---	---
<i>An. samarensis</i>	---	---	---	---	---	1	2	---	---	---
<i>An. subpictus</i>	---	---	---	16	1	---	---	---	---	---
<i>An. tessellatus</i>	---	---	---	---	---	---	---	---	---	1
<i>An. vagus</i>	---	---	---	---	---	---	---	---	2	---
<i>Ae. (Bothaella) sp</i>	---	---	---	1	---	---	---	---	---	---
<i>Culex fuscocephala</i>	---	---	---	4	4	1	11	4	---	---
<i>Cx. gelidus</i>	---	---	---	---	---	2	5	7	---	---
<i>Cx. quinquefasciatus</i>	19	119	---	---	3	36	17	1	5	74
<i>Cx. sitiens spp complex</i>	---	---	1,202	1,009	87	4	23	118	---	---
<i>Cx. tritaeniorhynchus</i>	---	---	---	---	---	1	---	---	---	---
<i>Cx. vishnui</i>	---	---	---	21	24	9	26	28	---	37
<i>Cx. whitmorei</i>	---	---	---	---	35	4	1	2	---	---
Total	31	133	1,244	1,088	193	83	114	181	25	151

^aNumber of specimens. ^bSpecies not identified.

Ae. poicilius in the different provinces in Panay showed no distinct pattern of activity. This species was also collected from outdoor stations at all periods from 06:00 to 12:00 PM, with only a slight increase in densities between 07:00 and 08:00 PM and between 10:00 and 11:00 PM.

DISCUSSION

Anopheles species suspected as vectors were relatively rare in HLC compared to CBT collections (17 and 9 *Anopheles* spp, respectively). *An. flavirostris* was not encountered in any location. This species

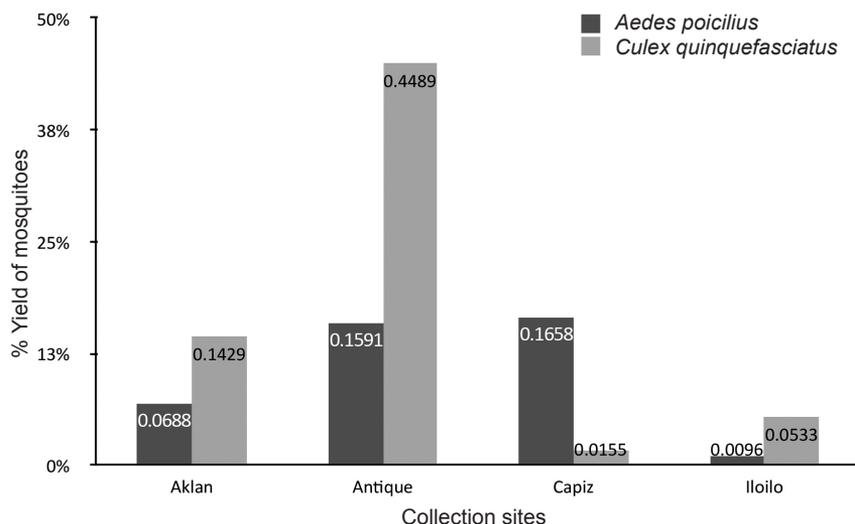


Fig 2—Proportion (percentage) of potential filaria vectors among mosquitoes from human landing collections (HLC) on Panay Island, The Philippines, September 2011 to October 2012.

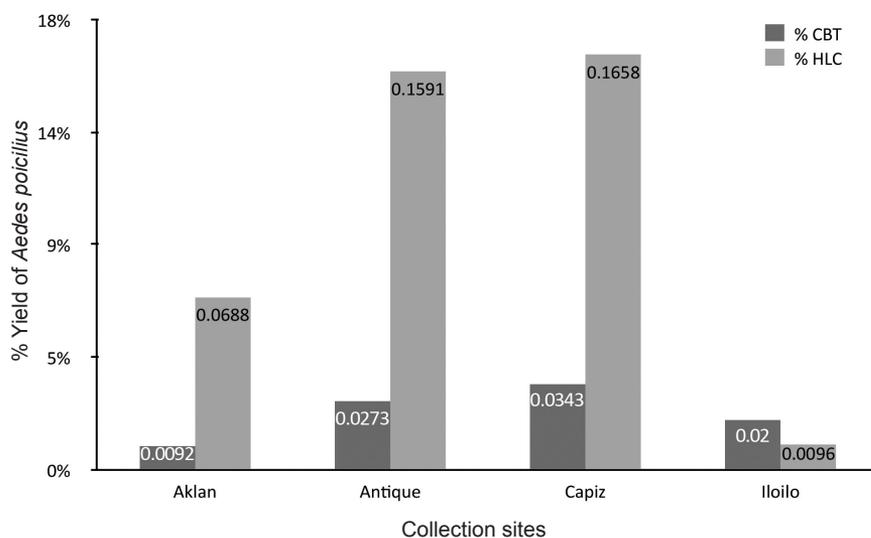


Fig 3—Mean proportion (percentage) of *Aedes poicilius* collected in paired carabao-baited traps (CBT) and human landing collections (HLC) on Panay Island, The Philippines, September 2011 to October 2012.

The peak landing activity of *Ae. poicilius* (09:00 to 11:00 PM) coincided with the time during which the majority of the resident population retired indoors, especially women and children. This suggests that sleeping under a mosquito net would likely provide adequate protection against this vector species. However, interviews with homeowners at the collection sites revealed that the majority of the residents do not possess bed nets, while others indicated a dislike for using bed nets for a variety of reasons (claustrophobic feeling, uncomfortably warm), thereby not conducive to a restful sleep. In addition, as *Ae. poicilius* is active outdoors throughout the first half of the night, people with outdoor activities during this period are at greater risk of acquiring filariasis if they do not

is not only a suitable secondary vector of Bancroftian filariasis (Valeza and Grove, 1979) but is also the most important vector of human malaria throughout The Philippines (DOH, 2013).

employ the use of some form of protection against insect bites.

Although female *Ae. poicilius* were attracted to the carabao, CBT collection is not representative of the potential biting

intensity of *Ae. poicilius* on humans (Walker *et al*, 1998). Comparing the proportion of *Ae. poicilius* collected from both CBT and HLC in relation to other mosquito species in the different provinces, the CBT data were transformed by dividing them into two to take into account that CBT collections were 10-hour collections while HLC was only conducted for the first 6-hour of the night. The numbers of *Ae. poicilius* taken from indoor and outdoor human baits (two persons per collection period) were found to be significantly greater than those collected from CBT. Considering individual numbers alone, far more *Ae. poicilius* specimens were collected using CBT method, but in relation to other mosquito species collected, a much higher relative proportion was obtained from HLC than CBT. However, as a collection method, CBT clearly attracted more species (diversity) and numbers of mosquitoes than HLC. Besides host preferences (*ie*, anthropophily *vs* zoophily) factor, the apparent enhanced attraction of most species to carabao was the greater types of host cues (odor, heat, carbon dioxide) produced and larger surface area of the animal bait.

There are several limitations to this study. Mosquito collections took place over a 1 year period but were limited in scope and duration because of the number of sites (3 sampling nights per site) and the influence of season and specific time of collection. Thus the data do not serve as a definitive description of the bionomics of the mosquitoes encountered but only list species found and their relative abundance. Species identification was hampered by a lack of updated and comprehensive morphological identification keys for The Philippines. In some cases, the reliance on published keys produced for aedine species occurring on mainland

Southeast Asia (Thailand) may have resulted in errors in identification and must await confirmation. In particular, *Armigeres* spp tentatively identified as *Ar. jugraensis*, *Ar. moultoni*, and *Ar. theobaldi* and one putative member in the subgenus *Bothaella* [possibly *Catatassomyia meronephada*, a species present in The Philippines (Knight and Stone, 1977; Rattanaarithikul *et al*, 2005a; *ibid* 2005b; *ibid* 2006; *ibid* 2010)] may represent new distribution records in the country. No attempt was made to incriminate mosquitoes collected as actual vectors of *W. bancrofti*. This was partly due to logistical constraints encountered by the team in some of the more remote locations to perform manual dissections. Additionally, all areas surveyed recently had received mass drug administration for filariasis and follow-up active case detection failed to identify a single case (DOH, 2013). Therefore the likelihood of detecting infected mosquitoes would have been remote. Given the importance of *Ae. poicilius* as a filariasis vector in The Philippines, we chose to focus on this species with the assumption that it was likely the primary vector in Panay. In Antique Province, the majority of anopheline and *Culex* specimens were not identified at the species level because many of the specimens were damaged by water exposure during the sampling periods performed during the rainy season.

From a series of mosquito surveys in filariasis endemic provinces on Panay Island, the presence of the primary, putative filariasis vector, *Ae. poicilius*, had been confirmed from all surveyed sites. The highest abundance of the species was observed during the rainy season (July to November) while the dry season rendered the species almost undetectable. This suggests that, as a predominantly leaf axil breeder (*eg*, *Musa* spp), eggs of the species

can either withstand desiccation sufficient to survive the dry summer months and/or that adult females are able to survive the inclement period and resume activity after the rains return. As this species relies heavily on abaca and bananas, the density of adult populations is a reflection of the number of these 'indicator' plants in an area (Baisas, 1972). The fact that bananas are commonly planted near houses, both in villages and towns, filarial transmission risk is enhanced in both areas and this should be taken into consideration when planning a control program (Cabrera and Valeza, 1978). We found this species to have a more zoophilic tendency, although it will also readily feed on humans. Previous findings have shown a high percentage of this species as human biters (Baisas, 1972). The peak indoor biting time was between 09:00 PM and 11:00 PM coinciding with the normal sleeping schedule of local residents. When collections from all sites were combined, the species showed no strong preference for biting either indoors or outdoors. The common practice of forest product gathering for livelihood likely increased human-vector contact as residents commonly remain overnight in forested locations without adequate protection against mosquito bites. Based on these observations, it would appear that distribution of insecticide-treated bed nets in filaria-endemic areas in rural areas of Panay would help to reduce disease risk. However, without strong and sustainable information campaigns on the importance of bed net use to increase community understanding and acceptance of the proper use and maintenance of such bed nets, this type of control would likely provide limited benefit. This study therefore provides baseline information for integrating vector control into the island-wide lymphatic filariasis elimination program.

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