

## Distribution and habitats of mosquito larvae in the Kingdom of Tonga

Jon S Harding,<sup>1\*</sup> Culum Brown,<sup>2</sup> Felicity Jones<sup>2</sup> and Russell Taylor<sup>1,3</sup>

<sup>1</sup>School of Biological Sciences, University of Canterbury, Christchurch, New Zealand

<sup>2</sup>Department of Biological Sciences, Macquarie University, Sydney, NSW 2109, Australia

<sup>3</sup>EcoCare Pacific Trust, Christchurch, New Zealand

### Abstract

Mosquitoes are a significant pest and human health issue in the Kingdom of Tonga. The occurrence of species and habitats used by mosquito larvae were investigated to determine the potential for control through larval habitat management. Forty-two sites, including 22 villages and 20 farm plantations on the six islands of Tongatapu, Pangaimotu, Vava'u, Pangaimotu (Vava'u group), 'Utungake and Nuku, were surveyed in April 2006. A total of eight mosquito species were collected: *Aedes aegypti* (Linnaeus), *Ae. horrescens* (Edwards), *Ae. nocturnus* (Theobold), *Ae. tongae* (Edwards), *Culex albinervis* (Edwards), *Cx. annulirostris* (Skuse), *Cx. quinquefasciatus* (Say) and *Cx. sitiens* (Wiedemann). Several species were widespread, particularly *Ae. aegypti* and *Ae. nocturnus* on the main island of Tongatapu, whereas *Ae. aegypti* dominated sites on islands of the Vava'u group. Comparative sampling of 17 village and 17 rural sites showed that larval habitat was more abundant in towns than in rural areas. Larvae were found in a wide range of habitats but were particularly abundant in artificial water bodies (e.g. disused concrete water tanks, 44-gallon drums and used car tyres). In rural sites, habitats were generally sparse except in rain-filled branch stems of giant taro plants. Mosquito populations in artificial habitats could be markedly reduced by seeding disused water tanks with aquatic predators already present in Tonga, using mesh-net covers over 44-gallon drums, and drilling holes in used car tyres.

**Key words** *Aedes aegypti*, artificial habitat, larva, mosquito, Tonga.

### INTRODUCTION

Mosquitoes are one of the most important vectors of disease globally. Currently, more than 2000 mosquito species have been identified worldwide, many of which are known vectors of human disease. Mosquito-borne diseases pose a major threat both to human populations and to the diversity of indigenous fauna throughout the world (Spielman & D'Antonio 2001), and are of particular concern in developing nations of the South Pacific where the ecology and distribution of mosquitoes is only partially documented. Various arboviruses and protozoan parasites have been widely reported around the Pacific (Hales *et al.* 1999). In Tonga, outbreaks of Dengue occurred in 1974, 1975, 1998 and 2003, causing several fatalities (Gubler *et al.* 1978; Muto 1998; WHO 2006). Furthermore, a number of mosquito species known to be vectors of Dengue fever, West Nile virus and Ross River virus (e.g. *Aedes aegypti* (Linnaeus) and *Ae. nocturnus* (Theobold)) have been previously reported in Tonga (Belkin 1962).

Many of the mosquito-borne diseases are zoonotic and can have dramatic effects on endemic fauna, but such effects are often overlooked because efforts are often firmly focused on the human health issues (Atkinson *et al.* 1995). The introduction of *Culex* mosquitoes to Hawaii in the early 19th century

was believed to be responsible for the establishment of avian pox virus and malaria (*Plasmodium relictum* Grassi & Feletti) in Hawaiian forest bird populations (Atkinson *et al.* 1995). Moreover, some native birds may be more susceptible to introduced diseases and have a significantly poorer survival rate than introduced birds (van Riper & van Riper 1985; Atkinson *et al.* 1995). Mosquito-borne diseases therefore may represent a serious threat to both human health and regional biodiversity.

The Kingdom of Tonga is widely recognised as having a mosquito problem (Stanley 1999); however, the challenges of conducting research in a developing country means that there is a paucity of information on Tongan mosquito species. The earliest records of mosquito taxonomy relating to Tonga come from collections by Edwards and others from which *Ae. tongae* was described (Edwards 1926). Laird (1956) published records of four species from Tongatapu (*Ae. nocturnus*, *Ae. oceanicus* (as *samoanus* Gruenberg), *Cx. annulirostris* (Skuse) and *Cx. quinquefasciatus* (Say)), while Iyengar (1955, 1960) documented eight species from collections around the capital Nuku'alofa and Vava'u. However, the most comprehensive work to date has been produced by Belkin (1962), who listed 10 species including a single endemic species *Ae. tongae* (Edwards), two indigenous species *Cx. annulirostris* and *Cx. sitiens* (Wiedemann), two introduced species *Ae. aegypti* and *Cx. quinquefasciatus*, two species which he suggests may have been spread by Tongans, *Ae. nocturnus* and *Ae. oceanicus*, and three species which he considered to be of questionable

\*jon.harding@canterbury.ac.nz

identification, *Cx. roseni* (Belkin), *Cx. albinervis* (Edwards) and *Ae. horrescens* (Edwards). Ramalingam (1976) lists 11 possible species in Tonga, though the age of these records and the fact that the majority of Tonga's islands have not been sampled for mosquitoes mean that many more species are likely to exist. To our knowledge, no studies have been published on the distribution of mosquito taxa in Tonga in the last 30 years.

In this study, we conducted a one-off survey of mosquito species and larval habitats in a subset of the main islands within the Kingdom of Tonga to improve our understanding of the current distribution of mosquito species on selected islands. The occurrence of mosquito larvae in urban and rural habitats was also compared, and we characterised the relative importance of differing artificial and natural habitats for mosquito larvae with a view to assist the development of larval mosquito control strategies.

## MATERIALS AND METHODS

### Study sites

Tonga lies between Fiji and Niue in the South Pacific Ocean, between latitude 15° and 23°30'S and longitude 173° and 177°W. The Kingdom includes three main groups of islands (Tongatapu, Ha'apai and Vava'u) that comprise approximately 170 islands, giving Tonga a collective landmass of about 718 km<sup>2</sup>. The majority of the islands are of volcanic and raised coral origin. The Kingdom has a tropical climate with warm temperatures and high rainfall (Vava'u has an annual average temperature of 24.5°C, and 2279 mm rainfall; Stanley 1999).

Six islands were surveyed on a single occasion over 10 days in April 2006 (Fig. 1), within the Tongatapu island group, the main island of Tongatapu and a smaller off-shore island Pangaimotu, and in the Vava'u island group, four islands: Vava'u, Pangaimotu, 'Utungake and Nuku. Time, funding and logistical constraints precluded multiple sampling. The urban areas of Tonga consist of the capital city of Nuku'alofa (on Tongatapu) and a large number of markedly smaller towns and villages (e.g. Tongatapu has approximately 45 small villages). The towns generally consist of 50–100 buildings and are almost entirely residential with few multistorey buildings, few businesses (except one or two small shops) or industry. The rural areas are dominated by cultivated fields of coconut trees, often with ground crops such as taro, giant taro, pumpkins, yams, sweet potato, manioc and vanilla. Rural land on the inhabited islands is almost entirely cultivated. We encountered only a single small fragment of native or regenerating forest, which we were unable to get permission to sample.

### Mosquito distribution

To determine the distribution of species, 42 sites were sampled (Fig. 1). Of these, 22 were towns and villages and within each a grid of approximately 100 m<sup>2</sup> was examined. At each urban site, we conducted a systematic search of the grid. If no larval habitats were found, we then asked local residents about

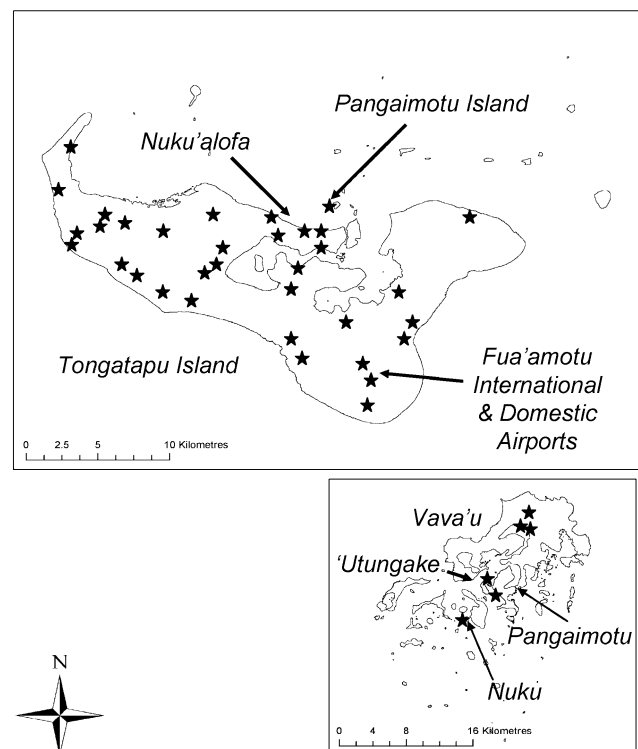


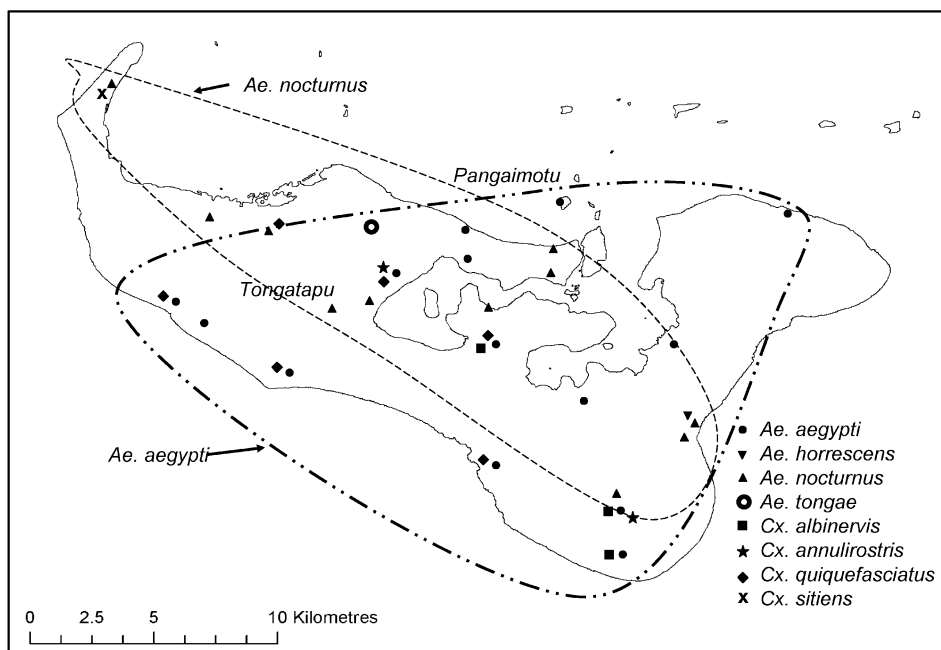
Fig. 1. Sites of the 42 sites on six Tongan islands sampled in April 2006.

known and possible mosquito habitats outside our grids. These additional habitats were then sampled to determine species that may be present in the vicinity. A further 20 sites were selected in rural areas; these were all cultivated fields with coconut trees and a grid of approximately 100 m<sup>2</sup> was searched. All ground-level standing water habitats within each grid were identified and representative mosquito larvae were collected from each water body.

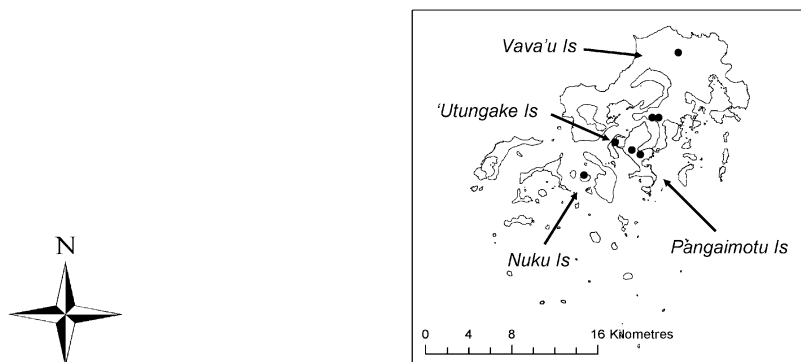
On Vava'u, the occurrence of larvae in water trapped on five common tree species was also investigated. In four different plantations, 10 trees of each of taro, giant taro, fan palms, ground palms and mango trees were randomly selected and branches and roots within 2 m of the ground were searched for the presence of trapped water and larvae.

### Urban vs. rural comparison

A total of 34 randomly selected grids (25 m × 25 m) were surveyed: 17 in towns and 17 in rural plantations of Tongatapu, Vava'u and Pangaimotu (in the Vava'u group). All ground-level larval habitats were identified in each grid and the type of habitat was noted. Representative larvae were collected at each habitat with a pipette or hand net. Many habitats (e.g. car tyres, tins, tyre tracks) could be thoroughly sampled with pipettes and sweep nets while representative larvae were collected in larger habitats (e.g. concrete water tanks). All larvae were preserved in the field in 70% ethanol and returned to the laboratory for identification. Larvae were identified using Belkin (1962).



**Fig. 2.** Occurrence of each of the eight species of mosquito larvae across the six Tongan islands. The possible range of *Aedes aegypti* and *Ae. nocturnus* are shown.



### Statistical analyses

The number of larva-positive sites per 25 m<sup>2</sup> sampling grid in towns compared with rural sites was evaluated using *t*-test, while differences between the occurrence of larvae in towns compared with rural sites and natural compared with artificial habitats were tested using a Kruskal–Wallis one-way ANOVA on ranks. Data failed normality tests following a variety of transformations.

## RESULTS

### Distribution of mosquito species

Mosquitoes were readily collected in a wide range of sites and habitats. Eight of the 42 sites sampled did not have larvae present, and six of these were rural sites. Among these 42 sites, 175 standing water habitats were sampled and mosquitoes were found in 74 habitats (42% of habitats). A total of eight mosquito

species were found, of which *Ae. aegypti* was the most widespread (Fig. 2, Table 1). Tongatapu had all eight species, while the only other island with more than one species was Pangaimotu (in the Vava'u Island group). *Aedes aegypti* was the most commonly collected species occurring at 21 sites (50% of all sites sampled), while *Ae. nocturnus* was found in 11 (26%), *Cx. quinquefasciatus* in seven (17%), and *Ae. horrescens*, *Ae. tongae* and *Cx. sitiens* were only found at one site each.

*Aedes aegypti* was collected on all six islands and was the only species found in Pangaimotu (in the Tongatapu Island group) and on Vava'u, 'Utungake and Nuku islands. Although widespread on Tongatapu, this species was not collected at any of the eight sites located in the north-western region of the island (Fig. 2). The next most abundant species, *Ae. nocturnus*, was only found on Tongatapu and was not collected on the southern coast of the island, despite nine of our sample sites covering this area. In contrast, *Cx. quinquefasciatus* seemed widely distributed in the southern and central regions of Tongatapu, but was not collected in the west.

**Table 1** Mosquito species and number of sites at which each species was collected on the six Tongan islands

	Tongatapu Islands		Vava'u Islands			
	Tongatapu	Pangiamotu	Vava'u	Pangiamotu	'Utungake	Nuku
Island area (km <sup>2</sup> )	270	0.1	89	8.7	1.1	0.08
<i>Aedes aegypti</i>	13	1	4	2	1	1
<i>Aedes horrescens</i>	1					
<i>Aedes nocturnus</i>	11					
<i>Aedes tongae</i>	1					
<i>Culex albinervis</i>	3					
<i>Culex annulirostris</i>	2					
<i>Culex quinquefasciatus</i>	6			1		
<i>Culex sitiens</i>	1					

Occurrence of multiple species at the same site was relatively uncommon with only seven sites having two or more species at a site. The Fua'amotu Domestic Airport on Tongatapu had three species, *Ae. aegypti*, *Cx. albinervis* and *Cx. annulirostris*, but these may have been distributed across several discrete habitats at this site (e.g. mud pools and mango tree roots). Several species did co-occur: of the seven sites that *Cx. quinquefasciatus* was collected, *Ae. aegypti* was present at six, and several of these were in the same 44-gallon drums. Similarly, *Ae. aegypti* was also present at all three sites at which *Cx. albinervis* was collected. Conversely, despite their relative abundance *Ae. aegypti* and *Ae. nocturnus* were never collected at the same sites.

### Urban vs. rural occurrence

The eight species collected were distributed relatively evenly across both urban and rural sites, with the three most common species *Ae. aegypti*, *Ae. nocturnus* and *Cx. quinquefasciatus* occurring frequently in both land uses (Fig. 3). A comparison of the occurrence of mosquito larvae in towns and rural areas showed that there was no significant difference between the number of larval habitats in urban areas compared with rural areas ( $t = 1.631$ ,  $P < 0.055$ ; Fig. 3). Within these two land uses larvae were found primarily in artificial habitats in towns, while in rural areas mosquitoes used natural and artificial habitats equally (Fig. 3).

### Habitat preferences

Towns had significantly more artificial habitats than natural ones, while in rural sites similar percentages of artificial and natural habitats were detected (Kruskal–Wallis one-way ANOVA,  $H = 8.92$ ,  $P < 0.03$ ; Fig. 3). The occurrence of artificial habitats in rural plantations therefore markedly increased the number of habitats available for mosquitoes (Fig. 3). A notable exception to this occurred in Giant taro plantations where larvae were found in taro branch axils (Table 2). The main artificial habitats colonised in villages and towns were pools (formed by tyre tracks), discarded car tyres, disused concrete water tanks and 44-gallon drums (Table 2). In farm plantations, mosquito larvae were commonly found in giant taro (and to a lesser extent in Mango and Fig tree roots), as

well as ground pools and 44-gallon drums. We did not find mosquito larvae in banana, coconut, palm or fan palm trees.

The three most common mosquito species were found in markedly differing habitats (Table 2). Although found in a range of habitats, *Ae. nocturnus* was most commonly found in natural and tyre-track pools (71% occurrence), whereas *Cx. quinquefasciatus* was found primarily in 44-gallon drums (66% occurrence). In contrast, *Ae. aegypti* occurred in a range of habitats, particularly car tyres, 44-gallon drums and Giant taro branch axils (Table 2).

A survey of phytotelmata in common tree species showed that 40% of giant taro plants had larvae in water collected in their leaf axils. This habitat trapped small pockets of water 2–3 cm deep which seemed to provide sufficient habitat for several larvae. Approximately 20% of mango trees also had larvae usually in pools associated with their surface root systems. Again, this water was comparatively deep (2–5 cm) and may persist without draining or evaporating for some time. The other trees surveyed did not seem to trap water in their branch axils or roots and we did not find suitable larval habitat within 2 m of the ground.

Of the 175 standing water habitats sampled, nine (mainly disused water tanks) had fish, dragonfly nymphs and/or water boatmen present and only two contained mosquito larvae (*Ae. nocturnus*), which were in very low abundances.

## DISCUSSION

### Occurrence and distribution of species

Our survey though limited by a single census in autumn confirmed the presence of at least eight species in Tonga, including confident identifications of *Cx. albinervis* and *Ae. horrescens*. Other species, including endemics are probably present and might be detected with more extensive and repetitive sampling. The three species that Belkin (1962) suggests are endemic or indigenous to Tonga (*Ae. tongae*, *Cx. annulirostris* and *Cx. sitiens*), were the rarest species in our survey, whereas the two introduced species (*Ae. aegypti* and *Cx. quinquefasciatus*) were collected at 52% of our sites. *Aedes aegypti* was almost always present at the same sites as *Cx. quinquefasciatus*, and Ramalingam (1976) notes that

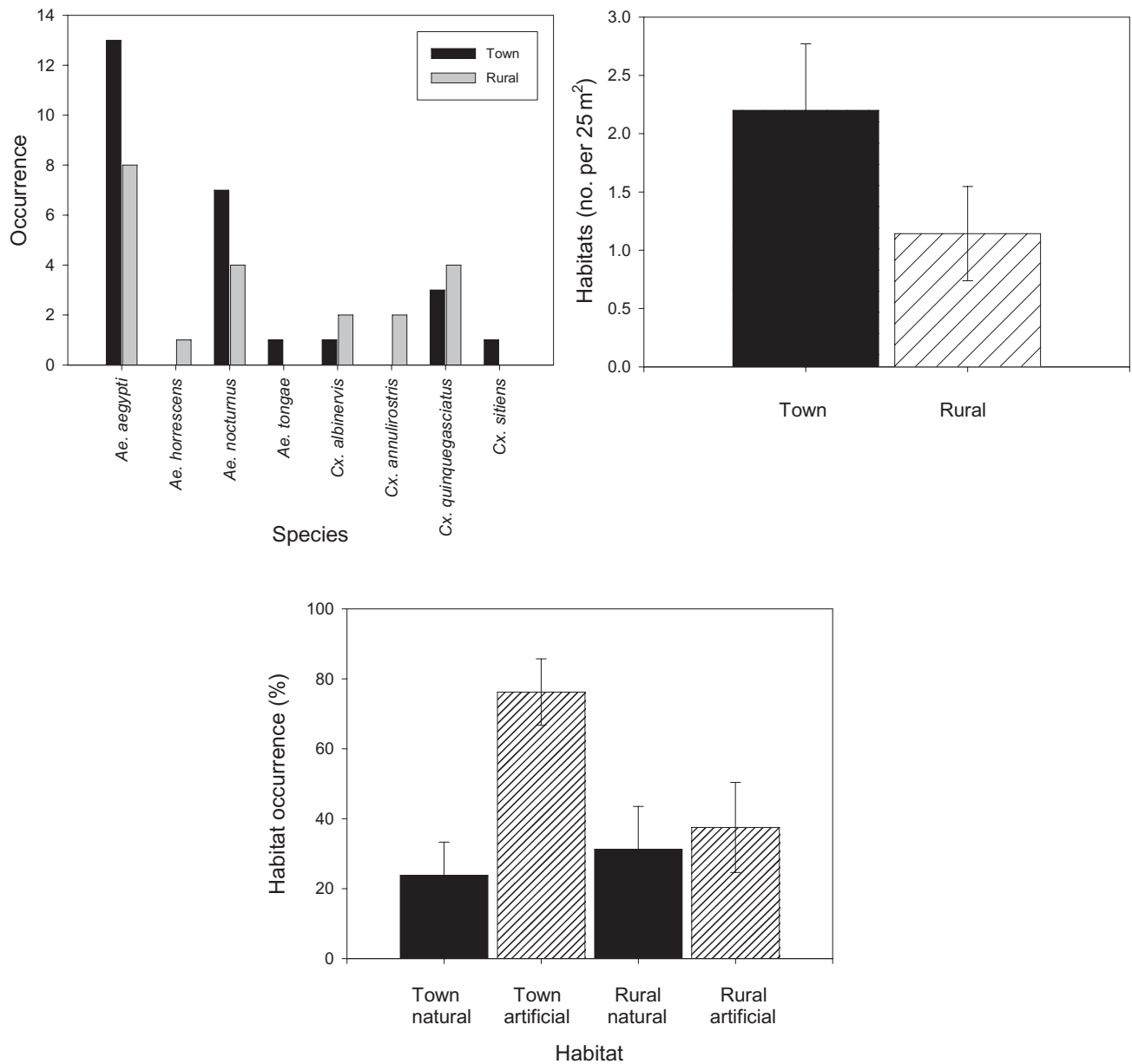


Fig. 3. Occurrence of mosquito habitat in town and rural sites in both natural habitats and artificial man-made habitats (mean ± standard error).

Table 2 Percentage (%) of micro-habitats occurring in town and rural sites in Tonga, and habitats in which three mosquito species were collected

	Town	Rural	<i>Aedes aegypti</i>	<i>Aedes nocturnus</i>	<i>Culex quinquefasciatus</i>
Pool	39.5	15.6	0	74.0	16.6
Car tyres	16.3	0	22.0	0	0
44-gallon drum	4.6	21.8	16.0	3.2	66.6
Plastic container	2.3	3.1	–	–	–
Water tank	11.6	0	8.0	64.0	8.3
Giant taro	0	46.8	30.0	0	0
Other	25.3	12.4	–	–	–
Coconuts	–	–	10.0	3.2	0
Other artificial	–	–	14.0	16.0	8.3

*Ae. aegypti* can be found breeding with *Cx. quinquefasciatus* and *Ae. tabu* Ramalingam and Belkin. The lack of co-occurrence of *Ae. nocturnus* and *Ae. aegypti* at the same sites despite their overlap in distribution is almost certainly due to the differences of larval habitats. Lee *et al.* (1982) noted that *Ae. nocturnus* is frequently found in temporary natural pools and almost never with *Ae. aegypti*. In contrast, *Ae. aegypti* is well known as a coloniser of artificial habitats (Seng & Jute 1994; Nam *et al.* 1998; Thavara *et al.* 2001). Ramalingam (1976) noted that *Ae. nocturnus* did not occur in large numbers and he only recorded its presence in a few sites on Tonga. In contrast, we found aggregations of many hundreds of larvae of this species. Furthermore, Ramalingam does not mention that *Ae. nocturnus* co-occurred with other species; however, in our survey, it was found with *Cx. quinquefasciatus*, *Cx. sitiens* and *Ae. horrescens*.

Both Belkin (1962) and Ramalingam (1976) refer to the presence of *Ae. oceanicus*; however, we did not collect this species despite sampling in similar habitats and sites with their studies. The distribution of *Ae. oceanicus* may have changed during the intervening 30 years since their work, or sampling over multiple seasons may be necessary to detect it. Ramalingam (1976) suggested that knowledge of the mosquito fauna of Tonga is extremely fragmented and despite this earlier work and the addition of our study relatively few of the 170 islands within the Kingdom have been surveyed.

Our data also provide some insights into changes in the distribution of mosquito species. Ramalingam (1976) recorded only six species on Tongatapu, while our survey added *Cx. albivervis* and *Ae. tongae* to those found previously. The most striking difference, however, is the apparent expansion of *Ae. aegypti* which had not been previously recorded in the Vava'u island group. In our survey, *Ae. aegypti* was collected on four islands in this group (Vava'u, Pangaimotu, 'Utungake and Nuku) and at all sites sampled. We might speculate that if this taxon dispersed by natural mechanisms (e.g. via wind), and assuming each island group has available habitat for mosquitoes to colonise, then we might expect mosquito diversity to be similar across all island groups. However, if dispersal is mediated by humans (e.g. via aircraft or boats) then we might find greater diversity near transport hubs (e.g. Tongatapu). This is in fact what we observed: six species were collected near the capital of Nuku'alofa and in the vicinity of the International and Domestic Airports on Tongatapu. Anecdotal evidence of human-mediated dispersal is also provided by the presence of adult mosquitoes in the passenger cabin of our plane *en route* to Vava'u from Tongatapu. Failloux *et al.* (1997) found that the genetic distance between populations of the Polynesian mosquito, *Aedes polynesiensis* (Marks), was correlated not with geographical distance but rather with commercial traffic intensity. Human assistance of the mosquito dispersal has serious implications for the spread of any mosquito-borne disease between Tongan island groups. Of the eight species recorded in this survey, seven have been associated or implicated in some form of human or animal disease. Of most concern is *Ae. aegypti* which is a significant vector of Yellow fever, Dengue fever and Dengue haemorrhagic fever (Lounibos 2002).

## Habitat preferences

The use of artificial habitats by many mosquito species has been widely documented (Ramalingam 1976; Lee *et al.* 1982; Seng & Jute 1994; Nam *et al.* 1998; Thavara *et al.* 2001), as has a high occurrence of these habitats in urban areas. Several species were collected more frequently in particular habitats. For example, 70% of water bodies occupied by *Ae. nocturnus* were natural pools or 'ruts' created by vehicle tracks, while the remainder were found in concrete water tanks, ice-cream containers and small tins. This species was not collected from car tyres, and this observation is consistent with Ramalingam (1976) and Lee *et al.* (1982) who noted the presence of this species in shallow ground pools sometimes with grass vegetation.

*Culex quinquefasciatus* was only collected in artificial habitats, primarily in 44-gallon drums and large concrete water tanks. Ramalingam (1976) also noted the presence of *Cx. quinquefasciatus* larvae in cisterns and concrete drains but did not mention finding it in natural habitats; however, Lee *et al.* (1982) suggest that it readily uses artificial and natural water bodies near human habitation in Australia. In contrast, *Ae. aegypti* was widely distributed in a range of natural and artificial habitats and was the only species collected on all six islands. Ramalingam (1976) only found larvae of *Ae. aegypti* in artificial habitats and in larger towns and Nuku'alofa. Our findings indicate that this species has dispersed widely since 1976, and its ability to use almost any available larval habitat would have undoubtedly aided its spread (Kittayapong & Strickman 1993).

## Management of larval habitat

Artificial habitats accounted for 47% of all larval habitats; consequently, active management of these could markedly reduce mosquito populations in Tonga. In particular, disused concrete water tanks were common in towns around the main island of Tongatapu. In several cases tanks were full of water and had been stocked with predatory fish (*Poecilia mexicana* Steindachner). Mosquito larvae were absent from these stocked tanks. Removal, draining or stocking water tanks with mosquito predators such as fish, dragonfly larvae and/or water boatmen would render these habitats unavailable to mosquitoes. Elsewhere copepods, notonectids and fish have all been used successfully to reduce larval numbers (Marten *et al.* 1994; Nam *et al.* 1998; Kay *et al.* 2002). Another common artificial habitat was 44-gallon drums, which are widely used to collect rainwater for irrigation purposes or for mixing concrete. Turning the drums upside-down when not in use or fitting fine mesh covers would still enable rainwater to be collected but would inhibit adult egg laying. Finally, disused car tyres were abundant in some urban areas because of inadequate rubbish disposal facilities. However, in one village we observed tyres that had holes punched in them to drain rainwater, and no larvae were present in these tyres.

The results of this survey clearly indicate that larval habitats are widespread throughout Tonga. However, the number of larval habitats has been substantially increased by the creation

and lack of management of artificial habitats. Active management of these artificial habitats would markedly reduce mosquito habitats.

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