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Dry Season Production of Filariasis and Dengue Vectors in American Samoa and Comparison with Wet Season Production

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Abstract

Aedes polynesiensis and *Ae. aegypti* breeding site productivity in two American Samoa villages were analyzed during a dry season survey and compared with a wet season survey. Both surveys identified similar container types producing greater numbers of pupae, with buckets, drums, and tires responsible for > 50% of *Aede*s pupae during the dry season. The prevalence of containers with *Ae. polynesiensis* and the density of *Ae. polynesiensis* in discarded appliances, drums, and discarded plastic ice cream containers were significantly greater during the dry season. *Aedes aegypti* pupal densities were significantly greater in the dry season in ice cream containers and tires. Significant clustering of the most productive container types by household was only found for appliances. The high productivity for *Ae. polynesiensis* and *Ae. aegypti* pupae during the wet and dry seasons suggests that dengue and lymphatic filariasis transmission can occur throughout the year, consistent with the reporting of dengue cases.

INTRODUCTION

The most important vector-borne diseases in the South Pacific between Fiji and French Polynesia are lymphatic filariasis (LF) and dengue. Lymphatic filariasis is endemic in 16 Pacific island countries and territories. In contrast, dengue is not believed to be endemic in most of the Pacific island countries and territories including American Samoa. Instead, periodic dengue outbreaks occur upon reintroduction of the dengue virus by either infected

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humans or mosquitoes. Dengue and LF share a key epidemiologic feature in the South Pacific from Fiji through French Polynesia: both diseases are transmitted by day-time biting *Aedes* mosquitoes. *Aedes aegypti* (L.) is the primary dengue vector throughout the world, including the Pacific island countries where it is found. Dengue may also be transmitted by a number of secondary vectors, including *Ae. polynesiensis* Marks, which is found across much of the South Pacific, including the Samoan Islands.

Aedes polynesiensis is also the primary vector of LF between Fiji and French Polynesia.[1] The public health importance of LF, which is caused by infection with *Wuchereria bancrofti*, led to the formation of the Pacific Program for the Elimination of Lymphatic Filariasis (PacELF) with a goal to stop LF transmission in the 16 endemic PacELF countries and territories by 2010. The primary strategy for stopping LF transmission is by annual mass drug administration (MDA) with diethylcarbamazine (DEC) and albendazole to all healthy residents more than two years of age. However, previous MDA campaigns using DEC alone failed to eliminate transmission where *Ae. polynesiensis* is the vector. Despite reducing the LF micro-filariae (mf) prevalence to 0.14% in Samoa in the 1970s, the prevalence of LF increased upon cessation of MDA.[2] Similarly in French Polynesia, transmission continued despite more than 30 years of twice a year MDA with DEC.[3] The failure of the MDA campaigns may have been a function of several factors, including efficiency of *Ae. polynesiensis* as an LF vector.[4] *Aedes polynesiensis* is arguably the most efficient LF vector in the world because of a characteristic known as limitation in which the proportion of ingested mf that successfully reach the infectious third stage (L3) increases as mf densities decrease.[5]

MDA based campaigns may also have failed to eliminate transmission for other reasons, including the difficulty in attaining and maintaining high MDA coverage for an adequate number of years with persistent non-compliers who refuse to take the medication acting as reservoirs to infect mosquitoes and thereby maintain transmission. Such challenges argue that there is a need for adjunct control measures to MDA to achieve elimination.[4,6] In the near future, control of the vectors of LF is the only possible adjunct strategy with the potential for implementation.

Aedes polynesiensis and *Ae. aegypti* are day-time biting mosquitoes that use containers as breeding sites for oviposition.[7–9] Both mosquitoes also have limited flight ranges.[7,10] These characteristics suggest that elimination of breeding sites in villages might be effective in controlling these vectors and the diseases that they transmit. This strategy has been advocated as a community-based approach for the control of dengue vectored by *Ae. aegypti*.[11] Identifying the most productive breeding sites for vectors based on the numbers of pupae found in different container categories can enable source reduction campaigns to target the removal or destruction of the most productive breeding sites, thereby minimizing vector densities and limiting transmission.[12] This targeted source reduction approach was further refined by the recognition that a small proportion of key premises (households) could be responsible for the generation of a disproportionate number of vector mosquitoes.[13]

Previous work in American Samoa during the wet season established that *Ae. polynesiensis* uses both human-made and natural containers with equal frequency whereas *Ae. aegypti* pupae are found predominantly in human-made containers.[1] The most productive wet season human-made breeding sites for *Ae. polynesiensis* in American Samoa were buckets, tires, and plastic ice cream containers. Most *Ae. aegypti* were produced in 44-gallon drums, buckets, and tires. However, a number of important pieces of information critical to the implementation of a source reduction campaign in American Samoa were not addressed by the previous study. Information on the relative productivity of containers in the wet season relative to the dry season and on the distribution of productive containers and those households responsible for the production of most of the vectors was not collected. Such data would be useful for planning and carrying out source reduction campaigns. We present the results of a dry season survey in America Samoa and an analysis of the productivity of containers compared with that found in a survey conducted previously in the same villages during the wet season.

MATERIALS AND METHODS

Study sites

Study villages were located on the main island of Tutuila in American Samoa. Tutuila has a wet season from approximately October through May, with a mean daily rainfall of 9.7 mm and a dry season from approximately June through September with mean daily rainfall of 5.4 mm.[14] A pupal survey of domestic containers was undertaken during the dry season in the villages of Fagasa, Pago Pago, Aoloau, and Malaeloa from June 1 through July 29, 2004. Fagasa and Pago Pago are sentinel villages for monitoring the American Samoan LF elimination efforts and were previously surveyed during the wet season in 2002. The villages of Aoloau and Malaeloa were included at the request of the American Samoa Department of Health.

A daily average of 4.6 mm of precipitation fell during the period from 14 days before the start of the 2002 wet season survey in Fagasa and Pago Pago until the end of the survey period (February 4–March 7, 2002) (precipitation ranged from 0 mm to 52.1 mm with measureable precipitation falling on 62% of the days during this period). In contrast, a mean of 1.74 mm of rain fell from May 18, 2004 (14 days before the dry season survey began) through the end of the survey in these two villages (June 28, 2004).[14] Daily precipitation ranged from 0 mm to 14.5 mm with measureable rainfall recorded on 57% of days during this dry season period. Thus, although rainfall patterns observed were consistent with the designations of wet and dry seasons (mean daily rainfall during the wet season sampling period was more than 2.6-fold greater than the mean rainfall during the 2004 dry season survey), the rainfall observed was less than half of the observed historical rainfalls reported for both the wet and dry seasons.

Field methods

Container surveys were undertaken in all households within randomly selected clusters. To select clusters, villages were first partitioned into groups of 25 households and two clusters were randomly selected for surveys in each village (50 households in each village). All households in selected clusters were mapped by a global positioning system. In Fagasa and Pago Pago, all water holding containers (i.e., potential breeding sites) associated with households in the selected clusters were sampled and information on each container was recorded, including the container type and the water volume held. The presence of mosquito larvae was recorded and the number of mosquito pupae counted. Up to 30 pupae from individual breeding sites were transported to the laboratory for further processing.

In the laboratory, pupae were held inside incubators at 25°C and a relative humidity of 90% in 13-hour light/11-hour dark cycles for 48 hours to enable mosquitoes to emerge. Adult mosquitoes were killed by freezing before being identified to species using morphologic characteristics.[15,16] Counts of each species identified from emerged adults were recorded.

Surveyed containers were dichotomized as either natural or human-made. Natural containers included sea shells, coconuts, rock pools, and tree holes. Human-made containers were categorized as appliances, buckets, drums, ice cream containers, metal, plastic, tin cans, tires, or other. The other category included containers made of glass, polystyrene, paper, and cardboard cartons and soda cans, concrete holes, gutters, shoes, and car batteries.

Associations between characteristics of containers and pupal productivity were examined in the villages of Aoloau and Malaeloa. In these two villages, the six most productive container types for *Ae. polynesiensis* and *Ae. aegypti* from the Fagasa and Pago Pago surveys were analyzed using additional data on water quality and the amount of sun exposure. Container exposure to sunlight was categorized as either full to mostly sun-exposed or full to mostly shaded. Water quality was condensed into three categories: clean, organic, and other. The clean category was water that was clear without any suspended material in it, and the organic classification was water with either suspended or settled organic debris, such as leaves. The other category included water with rust, motor oil, grease, or paint thinner.

Statistical analysis

Poisson regression implementing the generalized estimating equations procedure to adjust for correlation among multiple containers at the same house was used to compare the number of mosquitoes by village, season (wet versus dry), container type, and water quality (organic versus clear). Logistic regression also implementing the generalized estimating equations procedure was used to compare the proportion of positive containers at various classification levels. In the event of sparse data, Fisher's exact test was used to compare prevalence rates of containers. All analyses were performed using SAS version 9.1 (SAS institute, Cary, NC). When considering multiple comparisons, the *P* value was adjusted using the Bonferroni correction. Statistical significance was set at alpha = 0.05.

The six most productive container categories for *Ae. polynesiensis* and *Ae. aegypti* were analyzed for associations between productivity and water quality and sun/shade score using chisquare tests. Containers with water classified as other were excluded from the analysis because of the range of conditions represented.

The spatial distributions of houses with the most productive container types (discarded appliances, buckets, drums, folded plastic sheets, discarded plastic ice cream containers, and tires) were analyzed to determine whether households with a particular container type were significantly clustered among the set of all households. To test for clustering, a test statistic that calculates the number of additional households with at least one of a particular container type among the *k* nearest neighbors of each household bearing that container type was used.[17] This count is compared with the distribution of container bearing households among the *k* nearest neighbors of each container bearing household location determined under a null hypothesis of random allocation of the observed number of container bearing households among the set of all observed household locations. The analysis was conducted separately on each of the village clusters.

Because the two clusters in Fagasa were contiguous, these two clusters were combined for the analysis. Using the same methods, we also assessed the spatial distribution of houses with at least one *Ae. polynesiensis* pupa present versus none and with at least one *Ae. aegypti* pupa present versus none, and the houses in the highest quintel for *Ae. polynesiensis* or *Ae. aegypti* counts versus lower counts. The correlations between the number of pupae found in a household and the number of highly productive containers at that household or the total number of containers at that household were examined using the Spearman correlation coefficient. Associations between number of pupae found in a household and number of containers and number of the three most productive container types were assessed using Poisson regression.

RESULTS

A total of 1414 containers were analyzed for mosquito productivity during the dry season (Table 1). Plastic receptacles, including discarded chairs, trays, and bottles, were the most abundant water-bearing containers, comprising 22% of the containers in Fagasa and Pago Pago. Discarded appliances were the least abundant, accounting for only 2% of containers sampled. Overall, 28% of containers were positive for mosquito larvae. A significant difference in the percentage of containers positive for larvae between container types was present in Fagasa ($P < 0.0001$) and Pago Pago ($P < 0.0001$).

Overall, 14% of water-holding containers in Fagasa and Pago Pago were positive for *Aedes* pupae, with 12% and 5% of containers positive for *Ae. polynesiensis* and *Ae. aegypti* pupae, respectively (Table 1). In Fagasa, the mean number of *Ae. polynesiensis* pupae per container was 1.44, ranging from 0.50 in plastic containers to 7.29 in drums. In Pago Pago, the mean number of pupae per container was 1.76, with a range from 0.21 in other containers to 11.79 in drums (Table 1). For *Ae. aegypti* , the mean number of pupae per container ranged from 0 in appliances to 3.36 in folded plastic sheets in Fagasa. Mean number of *Ae. aegypti* pupae in Pago Pago containers ranged from 0 in natural containers and ice cream containers to 2.76 in drums. Although 53% of containers were categorized as other containers (including soda cans, glass bottles, and polystyrene containers) or plastic containers, only 13% of *Ae. polynesiensis* and 6% of *Ae. aegypti* pupae were found in these container categories. In contrast, 76.9% of *Ae. polynesiensis* pupae (64.2% in Fagasa and 87.5% in Pago Pago) were found in buckets, tires, drums, ice cream containers, folded plastic sheets, and appliances. These container categories made up 27.4% of all containers sampled. For *Ae. aegypti*, 34.2% of containers (buckets, tires, drums, folded plastic, tin cans, and ice cream containers) produced 87.5% of the pupae collected (79.3% in Pago Pago and 91.7% in Fagasa).

Containers associated with households were almost twice as likely to be positive for *Ae. polynesiensis* pupae in the dry compared with the wet season (11.95% and 6.05%, respectively; $P < 0.0001$). For *Ae. aegypti*, the seasonal effect was dependent upon village: Fagasa containers were much more likely to be positive during the dry season than the wet season (5.85% versus 1.13%, respectively; $P = 0.0005$). Pago Pago showed no seasonal difference (4.60% versus 5.71%, dry season versus wet season, respectively). Overall, appliances, natural containers, and tin cans had higher prevalences for *Ae. polynesiensis* pupae during the dry season than during the wet season (37.04% versus 5.88%; $P = 0.04$; 10.00% versus 2.41%; $P = 0.012$; and 14.63% versus 3.57%; $P < 0.01$, respectively). Buckets and ice cream containers had significantly higher prevalences for *Ae polynesiensis* pupae during the dry season (34.19% versus 10.95%; $P < 0.001$ and 33.33% versus 7.79%; $P < 0.001$, respectively; Table 2). For *Ae. aegypti* pupae, the prevalence in drums and tires was higher but not significantly greater (after Bonferroni correction) during the dry season compared with the wet season (22.22% versus 2.70%; $P = 0.04$; and 14.81% versus 6.08%; $P = 0.02$, respectively).

For containers in Fagasa and Pago Pago combined, appliances, drums, and ice cream containers had significantly greater mean densities of *Ae. polynesiensis* pupae during the dry season than during the wet season (6.25 versus 0.03; $P < 0.0001$; 10.04 versus 0.84; $P < 0.05$; 4.97 versus 0.48; $P < 0.0001$) (Table 3). In addition, buckets ($P = 0.0005$) and tin cans ($P < 0.0005$) had greater mean densities of *Ae. polynesiensis* pupae in Fagasa during the dry season than during the wet season. Ice cream containers and tires had significantly greater mean densities of *Ae. aegypti* pupae during the dry season (0.97 versus 0.04; $P < 0.0001$ and 0.75 versus 0.18; $P = 0.0013$, respectively), but metal containers were more productive during the wet season (0.20 versus 5.56; $P < 0.0005$, dry versus wet, respectively; Table 3).

When analyzed by households, there were significantly more *Ae. polynesiensis* pupae during the dry season than during the wet season in American Samoa, with means of 22.6 and 4.8 pupae, respectively ($P < 0.0025$). Although almost twice as many *Ae. aegypti* pupae were found on average per household during the dry season (mean = 5.8) compared with the wet season (mean = 2.8), this difference was not statistically significant ($P > 0.05$).

Although not statistically significant, higher proportions of appliances, buckets, drums, ice cream containers, folded plastic sheets, and tires were positive for *Ae. polynesiensis* pupae when located in mostly shady locations than in sunny locations. The prevalence of *Ae. aegypti* pupae in appliances, buckets, drums, and tires was greater when these containers were located in the sun, and ice cream containers and folded plastic sheets had higher proportions containing *Ae. aegypti* pupae when located in the shade. These differences were also not statistically significant.

The mean numbers of *Ae. polynesiensis* pupae in appliances, buckets, drums, ice cream containers, folded plastic sheets, and tires with organic water were significantly greater compared with the same container categories with clear water (Table 4). Appliances, buckets, drums, ice cream containers, folded plastic sheets, and tires with organic water all had higher mean numbers of *Ae. aegypti* pupae compared with the same container categories with clear water, although the differences for drums and tires were not significant.

Distribution of containers and numbers of *Ae. polynesiensis* and *Ae. aegypti* pupae in the dry season were analyzed at the household level. With the exception of appliances in the village of Fagasa ($P = 0.024$), significant clustering of households for prevalence of the most abundant container types was not found. Evidence of significant clustering of households positive for either *Ae. polynesiensis* or *Ae. aegypti* pupae was also not found. Residences of Fagasa and Pago Pago were categorized into one of five quintiles based on the number of pupae present at the time of survey (0, 1–0, 11–20, 20–50, and > 50) (Table 5). When households were analyzed by quintiles for density of *Ae. polynesiensis* or *Ae. aegypti*, evidence for significant clustering of households in the highest quintile was not found. Greater than 50% of all *Ae. polynesiensis* and *Ae. aegypti* pupae were found in three container types: buckets, drums, and tires. The total number of containers and the total number of the three most productive container types were significantly associated with the number of pupae found in the households ($P < 0.0001$ and $P = 0.0029$, respectively). Altogether, 25% of the variation in the number of pupae at the household level can be explained by the variation in the number of the three most productive containers found per household, and 16% can be explained by the variation in the total number of containers of all categories associated with a household.

The percentage contribution to the total number of *Aedes* pupae in the villages of Fagasa and Pago Pago was calculated by household. Despite greater use of containers by these two species, 18% of households did not harbor pupae of either *Ae. polynesiensis* or *Ae. aegypti* during this dry season survey despite such households having a median of 5.5 containers per household. In contrast, the 20 most productive households had a median of 19.5 containers with a median of 49 *Ae. polynesiensis* and 5.9 *Ae. aegypti* pupae per household and contributed 63% of the *Aedes* pupae (1,781 of 2,839) found in the 100 households surveyed.

DISCUSSION

Vector control has a role in both LF elimination and dengue control programs.[4,11] The failure of MDA alone to eliminate LF in areas where *Ae. polynesiensis* is the primary vector suggests that additional measures to limit transmission will be required if elimination is to be achieved. At the present time, vector control is the only intervention with the potential for immediate implementation with MDA. Theoretical and empirical studies have indicated that vector control integrated with MDA campaigns may reduce the number of years of MDA required to eliminate transmission and reduce the likelihood of recrudescence.[18–20]

Similarly, the absence of either drugs or vaccines to prevent dengue leaves vector control of *Aedes* spp. vectors as the only presently available option to prevent and control dengue outbreaks.[11] The biology of the *Aedes* vectors will determine which control strategies are most likely to be successful. Although insecticide-treated mosquito nets are not likely to be as protective against the day-time feeding *Ae. aegypti* and *Ae. polynesiensis* as against night-time feeding mosquitoes, insecticide-treated nets have reduced *Ae. aegypti* larval and pupal indices in Haiti.[21] Insecticide applications inside houses can provide short-term control of the indoor resting *Ae. aegypti*,[22,23] but are less likely to be effective in controlling *Ae. polynesiensis*, which prefers to be outside of houses.[24,25]

Because *Ae. aegypti* and *Ae. polynesiensis* are weak fliers that use human-made and natural containers for breeding sites, a logical intervention to reduce the potential for dengue and LF transmission would be to limit mosquito breeding in containers in and near villages. The present study describes the relative productivity of various container types, between-season differences in productivity, and the distribution of productive containers among households in American Samoan villages.

Similarities were found in the productivity of *Ae. polynesiensis* and *Ae. aegypti* during the wet and dry seasons. In both seasons, only a few key container types produced a disproportionate number of adult mosquitoes, as estimated by pupal numbers. Although a previous study in independent Samoa found that *Ae. polynesiensis* breeding stopped in smaller containers during the driest month of the year,[9] our studies in American Samoa found an overall higher prevalence and greater density of *Ae. polynesiensis* pupae in containers during the dry season compared with the wet season. The density of *Ae. polynesiensis* pupae in a number of the most common container categories was significantly greater in the dry compared with the wet season with greater productivity of containers for pupae being associated with containers in predominantly shady locations or with water containing suspended or settled organic matter. The less pronounced impact of seasonality on *Ae. aegypti* container productivity might be related to the tendency of this species to occur in larger containers.[1]

Unlike many areas where dengue is transmitted, in American Samoa household storage of water is less likely to be a prominent factor in potentially maintaining dengue vector populations during the dry season because only 4% of households in the surveyed villages use a catchment system to store any portion of their water supply; > 97% of households in American Samoa receive water from a piped water system.[26]

Our observations in American Samoa suggest that in the absence of a significant level of household water storage, dry season rainfall may be sufficiently frequent and abundant for vector populations and the risk of dengue outbreaks and LF transmission to be as high or higher at times than during the wet season. Supporting this hypothesis of continuing high risk of dengue transmission during the dry season is the fact that both the 2001–2002 dengue type 1 and the 2008–2009 dengue type 4 outbreaks appeared to begin during the dry season and continued through the wet season and into the subsequent dry season (American Samoa Department of Health, unpublished data).

Although it is often assumed that increased rainfall results in increased mosquito production, this is not always the case, and mechanisms by which rainfall reduces populations of container-breeding mosquitoes have been proposed. Buxton and Hopkins[27] reported that long periods of heavy rain, as often occur in Samoa, reduce oviposition by *Ae. polynesiensis*. Koenraadt and Harrington[28] found that simulated rainfall could wash *Culex pipiens* L., but not *Ae. aegypti*, pupae out of container habitats. Frank and Curtis[29] and Teesdale[30] observed that rainfall expelled eggs of *Wyeomyia vanduzeei* Dyar and Knab from bromeliad leaf axils and *Aedes simpsoni* (Theobald) eggs from banana leaf axils, respectively. These observations on immature stages are supported by studies in independent Samoa where low biting rates for *Ae. polynesiensis* occurred during periods of high rainfall with higher biting rates after these periods.[25,31] Such direct effects of rainfall on natality and mortality could explain the reduced productivity measured in our rainy season samples, but further research is needed to determine if these effects occur with *Aedes* spp. in American Samoa. Conditions appear more than adequate to produce sufficient numbers of *Ae. polynesiensis* and *Ae. aegypti* for dengue and LF transmission in the wet and dry seasons in American Samoa.

Negative correlations between larval densities and rainfall have also been reported for mosquitoes breeding in rice fields[32] and ponds.[33] In the former case, *Anopheles* and *Culex* larval populations in the sampled habitats varied with changes in availability of other flooded habitats resulting from seasonal timing of rainfall and irrigation water management. In the latter case, it was suspected that desiccation of alternative habitats in the dry season resulted in increased *Anopheles* oviposition in the sampled ponds, which remained flooded throughout the study. In the present study, because all potential breeding sites in the villages were sampled, differences in availability of alternative, unsampled habitats does not seem a likely explanation for the differences in productivity observed between seasons.

Although our study identified key premises responsible for greater production of *Ae. polynesiensis* and *Ae. aegypti*,[13] these premises were not clustered in distribution. Surprisingly, numbers of containers in the most productive (key) container categories only explained a minor proportion of the variation in productivity of households. It appears that households that are highly productive for *Aedes* result from both a greater overall number of containers and a larger number of the more productive containers. The lack of clustering of either the most productive container categories or the most productive households suggests that source reduction campaigns must target the entire village to be effective.

Recently, Morrison and others reported that methods to control adult vectors of dengue are needed to limit dengue outbreaks because of ineffectiveness of attempts to control the larval stages.[34] The difficulty in providing adequate resources for the removal or destruction of *Aedes* breeding sites in villages in American Samoa suggests that multiple interventions will be needed to prevent or interrupt transmission of dengue.

However, because LF is less efficiently transmitted than dengue, source reduction campaigns for control could potentially be more effective in interrupting LF transmission even where *Aedes* are the vectors, in a manner analogous to the way that the unsuccessful malaria eradication program interrupted LF transmission in the Solomon Islands. Although the DDT-based indoor residual spray program failed to eliminate malaria in the Solomon Islands, this program succeeded in eliminating LF, which is transmitted by the same mosquitoes.[35]

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Table 1

Prevalence of larvae and density and prevalence of *Aedes polynesiensis* and *Ae. aegypti* pupae by container category and village during the dry season in American Samoa

			Fagasa Village					
			Ae. polynesiensis			***Ae. aegypti***		
Container	**No. containers**	**% Containers positive for larvae**	**Total no. pupae**	**Mean (SE) no. pupae**	**% Containers positive for pupae**	**Total no. pupae**	**Mean (SE) no. pupae**	**% Containers positive for pupae**
Appliance	9	67	10	1.09 (0.51)	44	0	0 (0)	0
Bucket	66	65	272	4.13 (1.0)	39	158	2.4 (1.3)	14
Drum	14	57	102	7.29 (5.2)	14	13	0.92 (0.55)	21
Folded plastic	15	33	105	6.98 (3.9)	27	50	3.36 (3.3)	13
Ice cream	18	39	73	4.07 (2.6)	22	43	2.41 (1.5)	17
Metal	28	39	27	0.96 (0.93)	7	13	0.46 (0.30)	11
Natural	60	20	38	0.63 (0.36)	10	4	0.07 (0.06)	2
Other	209	17	118	0.56 (0.37)	6	4	0.02 (0.01)	1
Plastic	185	16	93	0.5 (0.23)	5	11	0.06 (0.03)	3
Tin can	51	27	95	1.86 (0.93)	16	41	0.81 (0.69)	8
Tire	63	57	102	1.62 (0.77)	14	44	0.69 (0.24)	16
Total	718	29	1035	1.44 (0.24)	12	381	0.53 (0.15)	6

			Pago Pago Village					
			Ae. polynesiensis			***Ae. aegypti***		
Container	**No. containers**	**% Containers positive for larvae**	**Total no. pupae**	**Mean (SE) no. pupae**	**% Containers positive for pupae**	**Total no. pupae**	**Mean (SE) no. pupae**	**% Containers positive for pupae**
Appliance	18	72	159	8.82 (5.2)	33	25	1.4 (0.87)	28
Bucket	51	49	168	3.29 (1.1)	27	44	0.87 (0.55)	10
Drum	22	36	259	11.79 (9.1)	18	61	2.76 (1.4)	23
Folded plastic	38	34	74	1.95 (1.3)	8	12	0.33 (0.25)	5
Ice cream	27	59	150	5.57 (1.7)	41	0	0 (0)	0
Metal	42	26	15	0.36 (0.18)	12	1	0.02 (0.02)	2
Natural	20	15	5	0.25 (0.17)	10	0	0 (0)	0
Other	236	14	49	0.21 (0.09)	4	11	0.05 (0.03)	1
Plastic	125	14	44	0.35 (0.14)	6	4	0.03 (0.02)	3
Tin can	72	38	39	0.54 (0.21)	14	4	0.06 (0.05)	1
Tire	45	53	261	5.79 (2.9)	24	37	0.83 (0.39)	13
Total	696	27	1223	1.76 (0.40)	12	200	0.29 (0.07)	5

Table 2

Prevalence of containers positive for *Aedes polynesiensis* and *Ae. aegypti* pupae during the dry and wet seasons in American Samoa

			Ae. aegypti			*Ae. polynesiensis*		
	No. of containers		**% Containers positive**			**% Containers positive**		
Container	**Dry**	**Wet**	**Dry**	**Wet**	***P***	**Dry**	**Wet**	***P***
Appliances	27	17	18.52	5.88	0.2546	37.04	5.88	0.0413
Buckets	117	137	11.97	5.84	0.1510	34.19	10.95	0.0006
Drums	36	37	22.22	2.70	0.0422	16.67	2.70	0.1022
Folded plastic sheets	53	0	7.55	–	–	13.21	–	–
Ice cream	45	154	6.67	3.25	0.5897	33.33	7.79	0.0003
Metal	70	9	5.71	11.11	0.3408	10.00	0.00	1.0000
Natural	80	291	1.25	0.00	0.2156	10.00	2.41	0.0124
Other	445	112	1.12	3.57	0.1144	4.94	5.36	0.9369
Plastic	310	7	2.90	0.00	1.0000	5.16	14.29	0.3625
Tin cans	123	112	4.07	0.89	0.1476	14.63	3.57	0.0089
Tires	108	148	14.81	6.08	0.0216	18.52	10.14	0.0700
All containers	1414	1024	5.23	2.93	0.0049	11.95	6.05	< 0.0001

Table 3

Dry and wet season pupae densities of *Aedes polynesiensis* and *Ae. aegypti* pupae in American Samoa

			Ae. aegypti			***Ae. polynesiensis***		
	No. containers		**Mean (SE) density**			**Mean (SE) density**		
Container	**Dry**	**Wet**	**Dry**	**Wet**	***P***	**Dry**	**Wet**	***P***
Appliances	27	17	0.93 (0.59)	0.55 (0.55)	0.6547	6.25 (3.5)	0.03 (0.03)	< 0.0001
Buckets	117	137	1.73 (0.77)	2.65 (1.3)	0.3995	3.76 (0.74)	3.49 (2.8)	0.8059
Drums	36	37	2.05 (0.89)	0.51 (0.51)	0.2685	10.04 (5.8)	0.84 (0.84)	0.0385
Folded plastic sheets	53	0	1.18 (0.93)	–	–	3.37 (1.47)	–	–
Ice cream	45	154	0.97 (0.63)	0.04 (0.02)	0.0001	4.97 (1.4)	0.48 (0.18)	< 0.0001
Metal	70	9	0.20 (0.12)	5.56 (5.5)	0.0003	0.60 (0.38)	0.00 (0)	0.1345
Natural	80	291	0.05 (0.05)	0.00 (0)	0.0562	0.54 (0.27)	0.13 (0.06)	0.1022
Other	445	112	0.03 (0.02)	0.24 (0.18)	0.0290	0.38 (0.17)	0.41 (0.28)	0.9731
Plastic	310	7	0.05 (0.01)	0.00 (0)	0.3053	0.44 (0.15)	0.14 (0.14)	0.3164
Tin cans	123	112	0.37 (0.29)	0.17 (0.17)	0.5082	1.09 (0.40)	0.42 (0.29)	0.2365
Tires	108	148	0.75 (0.22)	0.18 (0.07)	0.0013	3.36 (1.3)	1.18 (0.44)	0.1255
All containers	1,414	1,024	0.41 (0.08)	0.51 (0.19)	0.9242	1.60 (0.23)	0.87 (0.38)	0.0481

Table 4

Comparison of number of *Aedes polynesiensis* and *Ae. aegypti* pupae between organic and clean water quality for highly productive containers in Aoloau and Malaeloa villages in American Samoa

	Water quality				
	Organic		**Clean**		
Mosquito, container	**No.**	**Mean (SE)**	**No.**	**Mean (SE)**	***P***
Ae. polynesiensis					
Appliance	17	9.75 (5.49)	10	0.28 (0.20)	0.0056
Bucket	171	6.11 (1.18)	35	0.29 (0.23)	0.0025
Drum	24	12.34 (8.68)	29	1.12 (0.84)	0.0076
Folded plastic	23	7.77 (3.21)	30	0 (0)	< 0.0001
Ice cream	71	5.49 (1.44)	14	0.84 (0.70)	0.0216
Tire	268	5.91 (1.28)	16	0.54 (0.47)	0.0005
Ae. aegypti					
Appliance	17	1.42 (0.92)	10	0.10 (0.1)	0.0322
Bucket	171	1.38 (0.43)	35	0 (0)	0.0001
Drum	24	1.83 (1.20)	29	1.61 (0.63)	0.6784
Folded plastic	23	2.73 (2.14)	30	0 (0)	0.0001
Ice cream	71	1.10 (0.44)	14	0 (0)	0.0134
Tire	268	1.35 (0.39)	16	1.28 (0.68)	0.6566

Table 5

Median pupal density by household (HH) in relation to overall container number and the most productive container categories in American Samoa

Quintile	No. of HHs	Pupae/HH, median (minimum–maximum)	*Aedes polynesiensis*/HH, median (minimin–maximum)	*Ae. aegypti*/HH, median (minimum–maximum)	Containers/HH, median (minimum–maximum)	Containers with pupae, median (minimum–maximum)	Three most productive containers, median (minimum–maximum)	Pupae in most productive containers, median (minimum–maximum)
0	18	0 (0)	0 (0)	0 (0)	5.5 (1–20)	0 (0)	0 (0–5)	0 (0)
1	20	6 (1–10)	1.6 (0–9)	0 (0–9)	9 (1–62)	1 (1–3)	2 (0–6)	1.5 (0–9)
2	20	15.5 (11–20)	12 (2–19)	1 (0–16)	11 (1–42)	2 (1–5)	2 (0–12)	5 (0–19)
3	22	35.5 (21–49)	24.7 (5.4–47.8)	2.3 (0–28)	11.5 (3–42)	2 (1–5)	2.5 (0–24)	9 (0–40)
4	20	84.5 (53–409)	49 (0–218.7)	5.9 (0–80.5)	19.5 (4–66)	4 (2–14)	3.5 (1–10)	53.5 (0–301)