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First evidence of spatial clustering of lymphatic filariasis in an *Aedes polynesiensis* endemic area

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ABSTRACT

Successful elimination of lymphatic filariasis (LF) requires accurate identification of residual foci of transmission and stringent surveillance strategies to combat potential resurgence. This is challenging in areas where the day-biting *Aedes polynesiensis* is endemic, such as Samoa, since in previous studies no geographical clustering of infection has been demonstrated. Another challenge for this low prevalence phase is the choice of diagnostic assay as testing for circulating filarial antigen (CFA) or microfilariae (Mf) alone may not have adequate sensitivity. This could be solved by using the commercially available filariasis Cellabs enzyme linked immunosorbent assay (CELISA) to measure antibody. In the current study five Samoan villages were chosen based on previous epidemiological assessments to represent a range of infection prevalences. CFA, Mf, and antibody levels in children ≤10 years had been recorded and results linked to household of residence and/or primary school of attendance. To ascertain the location of exposure, two scenarios based on potential foci of transmission around communities and schools were explored. Both scenarios revealed significant spatial clusters of households with infected individuals and a relationship to antibody positive children when they were included in the spatial analysis. Fasitoo-Tai had the highest LF prevalence and largest geographical spatial clusters for both scenarios. In Falefa, spatial clusters were detected only for the primary school scenario. In Tafua, which spanned an area of 19.5 km^2, no spatial clusters were detected. Lastly, in Siufaga, the village with the lowest LF prevalence, significant clustering of infected individuals was observed and, for the primary school scenario, this was geographically related to exposure. These promising findings are the first published evidence of spatial clustering of LF in a day-biting *Ae. polynesiensis* endemic area.

1. Introduction

Elimination of lymphatic filariasis (LF) as a public health problem in the South Pacific by the year 2010 was the primary goal of the Pacific Program for the Elimination of Lymphatic Filariasis (PacELF) (PacELF, 2006). Countries which achieved target reductions in LF infection entered monitoring and active surveillance mode up until 2017, depending on future surveys, whereas other countries with >1% circulating filarial antigen (CFA) prevalence of population planned further control efforts (WHO, 2007). Countries with persistent transmission require sensitive diagnostic assays, such as antibody serology, and sampling methods to identify residual foci of infection. These methods are especially applicable during active surveillance. These foci can be defined using spatial mapping.

During the initial stages of programmatic planning, spatial mapping was used on a large scale to predict areas of endemnicity in order for program managers to target mass drug administrations (MDAs) effectively and to plan elimination strategies (WHO, 1998). The large-scale spatial mapping using filariasis surveys was based on either 25 km × 25 km or 50 km × 50 km grids, depending on the geographical area studied, since assessment of every community would be cumbersome and expensive (WHO, 1998). Therefore, by using these methods, an estimation of the distribution of filariasis could be ascertained, similar to that achieved in other neglected tropical diseases (Brooker et al., 2009). Now that the prevalence of LF is declining, it is necessary to revisit spatial mapping in order to gain information regarding transmission patterns at finer scales

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since, as a mosquito-borne disease, LF is expected to show a high degree of heterogeneity over very small areas because of differences in vector distribution and breeding habitats (Gambhir et al., 2010).

There is a difference between mapping the geographic distribution of LF and fine scale spatial mapping using spatial statistical software to understand transmission dynamics and identify "hotspot" clustering. Fine scale spatial mapping, also referred to as micro-spatial mapping, has been successfully implemented in a number of diseases to infer likelihood risks, risk factors, extent of the disease, vector control in vector-borne diseases, surveillance, and to gain information on targeting control efforts since fine scale spatial mapping is at a similar scale to that at which control measures are implemented (Brooker and Clements, 2009; Clements et al., 2009; Eisen and Lozano-Fuentes, 2009). The understanding of transmission dynamics at the micro-spatial level would be extremely useful for effectively targeting residual LF endemic "hotspots" to understand the extent of their effect on the surrounding areas. It would also be useful in the future to delimit the areas around the zones of ongoing transmission that would require control efforts. This appears feasible in night-biting vector endemic areas since spatial clustering of LF has long been established (Walter, 1974).

To date, no spatial clustering has been detected in areas where the day-biting *Aedes polynesiensis* is the predominant vector (Mladonicky et al., 2009). This makes defining geographical areas of ongoing transmission a potential challenge in these vector endemic areas. *Ae. polynesiensis*, a highly efficient vector when intensity of transmission is low (Snow et al., 2006), is endemic in Samoa. Since the formation of PacELF, Samoa has completed five rounds of MDA from 1999 to 2003, and, after demonstrating persistent antigenaemia, completed 6th and 7th rounds in 2006 and 2008 (Huppatz et al., 2009; Ichimori and Crump, 2005). The persistent antigenaemia is of concern and requires immediate attention in order to successfully eliminate LF in Samoa.

As antibody production in response to LF exposure occurs during the first few years of life (Gao et al., 1994), children can serve as a sensitive indicator of LF transmission; because in the absence of transmission children should be antibody negative (Lammie et al., 1998; Weil et al., 1999). The persistent transmission in Samoa drives the need to investigate these areas of residual foci to define transmission patterns by ascertaining if infected individuals and/or exposed children are geographically clustered. Evidence of clustering would be useful for guiding surveillance efforts. Therefore, it was the aim of this research to establish if spatial clustering of LF existed in a day-biting vector endemic area and, if so, was there a relationship with exposed children. By doing so, the feasibility of using antibody serology as a means to complement future surveying strategies can be ascertained.

2. Materials and methods

2.1. Study area and population

This research study was conducted in May 2008, prior to the 7th MDA round in June 2008, on both islands of Samoa (Fig. 1). Any infected individuals found during the study were followed up during the 7th MDA round. Study areas chosen on the island of Upolu were Fasitoo-Tai, Siufaga and Falefa. Study areas chosen on the island of Savai'i were Tafua and Puapua. It was the aim of the research to screen every individual residing in the villages of Tafua, Puapua and Siufaga ≥2 years, and coverage rates achieved ranged from 79% to 84% of the population (Table 1). The villages of Fasitoo-Tai and Falefa had populations exceeding 1000 and it was the aim of the study to screen a minimum of 500 residents.

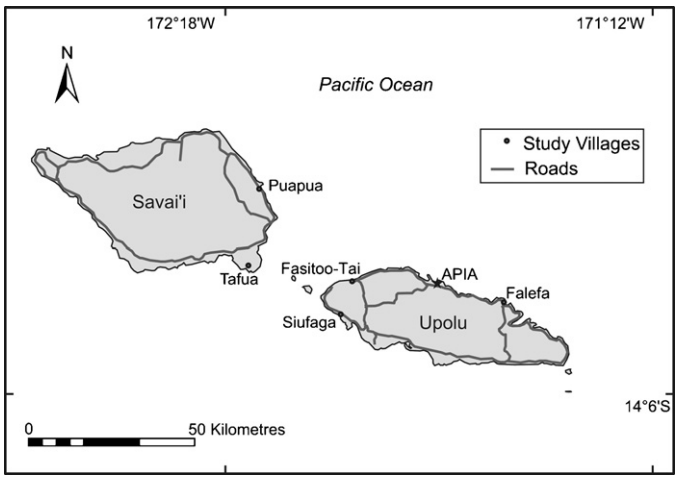

Fig. 1. Location of the five study villages in Samoa. On Upolu the three villages chosen were Fasitoo-Tai, Siufaga and Falefa. On Savai'i the two villages were Tafua and Puapua. The capital city, Apia, is included on the map as a reference.

The selection criteria for the latter villages related to a previous LF epidemiological survey completed in 2007. An individual from each village, who tested CFA positive in the previous 2007 survey, was randomly selected. Their household of residence was deemed the central point and, radiating out, every household was included in the survey until approximately 500 individuals were registered and screened. Since surveying occurred during the daytime, school children registered in the study by their guardians, after visiting their household of residence, were followed up at their respective primary schools.

In this research, any statement regarding "children" will refer to participants ≤10 years. The reasoning for choosing a target population of ≤10 years was due to the timing of the initial MDA. MDAs, under the guidance of PacELF, began in Samoa in 1999 (Ichimori and Crump, 2005) and targeting children born after the initial MDA placed their age at approximately 9 years old at the time of the study. Unfortunately, in most situations it was apparent that dates of birth were not recorded for children, thus the research was based on grade level for children who attended school. Children aged 9 or 10 years corresponded to grade five, thus any child equivalent to grade five was included in the study.

The study was conducted under human ethics approval number H1423, as approved by the James Cook University Research Human Ethics Committee. The study protocol was also approved by the Health Research Committee of the Samoan Ministry of Health prior to commencing the research.

Table 1
Demographics of the 5 Samoan villages chosen for the study.

Characteristic	Upolu			Savai'i	
	Fasitoo-Tai	Falefa	Siufaga	Puapua	Tafua
Male >10 years of age	232	197	190	162	131
Female >10 years of age	227	206	174	160	127
Male child ≤10 years	84	89	80	67	47
Female child ≤10 years	74	78	51	59	39
Total tested	617	570	495	448	344
Population census (2006)	1393	1388	629	552	408
% population screened[a]	44%	41%	79%	81%	84%
Median age (years)	19	18	23	18	20
Age range (years)	2–90	2–86	2–92	2–85	2–84

Note: for Fasitoo-Tai and Falefa it was the aim of the study to test a minimum of 500 individuals radially from a central house.

[a] Based on population census 2006.

Table 2
Prevalence of antigen (CFA), microfilariae (Mf), and anti-filarial antibodies (Ab) in each of the 5 villages (%) and 95% confidence intervals.

	Upolu			Savai'i	
	Fasitoo-Tai	Falefa	Siufaga	Puapua	Tafua
Mf prevalence (%)	3.2 (2.0–5.0)	0[a] (0–0.7)	0[a] (0–0.7)	0[a] (0–0.8)	0.6 (0.1–2.1)
CFA prevalence (%)	14.6 (11.9–17.6)	5.1 (3.4–7.2)	1.6 (0.7–3.2)	2.5 (1.2–4.4)	8.4 (5.7–11.9)
Ab prevalence children (%)	62 (54.0–69.6)	51.5 (43.6–59.3)	46.6 (37.8–55.5)	7.9 (3.9–14.1)	12.8 (6.6–21.7)
CFA prevalence children (%)	9.5 (5.4–15.2)	4.2 (1.7–8.5)	0 (0–2.8)	0.8 (0.2–4.3)	3.5 (0.7–9.9)

[a] Although 0% Mf prevalence was recorded, Mf testing was only performed on CFA positive individuals and not the entire population.

2.2. Blood collection

Blood was collected by fingerprick. CFA was measured in the field using the immunochromatographic test (ICT) as previously described (Weil et al., 1997). Positives were re-bled for confirmatory Og4C3 testing (Tropbio Pty Ltd, QLD, Australia) and 60 μL was used to make a three-line thick blood smear for Mf examination. Mf testing was performed during daylight hours, between 0800 h and 2000 h according to peak levels of microfilariae and biting tendencies of *Ae. polynesiensis* (Ramalingam, 1968). Children ≤10 years were also bled for antibody testing. Blood was collected onto a Tropbio filter paper disc (Tropbio Pty Ltd, QLD, Australia), dried, and transported back to Australia for storage at −20 °C for antibody testing. Anti-filarial IgG_4 antibodies were assayed using the commercially available Filariasis CELISA kit (Cellabs Pty Ltd, Manly, Australia), according to the manufacturer's instructions and as previously described (Joseph and Melrose, 2010).

2.3. Spatial data collection

Every household within the village was mapped using a global positioning system (GPS) handheld device and assigned a unique identifier. The GPS used was a handheld eTrex Legend™ (Garmin International Ltd., USA). Although the unit specifications for accuracy were <15 m root-mean-square (RMS) (95% of the readings within 15 m radius), it was found that accurate GPS readings could be obtained over 1–2 m. The unit had a 12-channel all-in-view tracking and National Marine Electronics Association (NMEA) 0183 GPS protocol.

When a family had more than one living area on their land, a reading was taken from the centre of their property. If an individual had multiple residencies in different parts of the village, their place of residence was defined as the place where they slept the majority of the time. GPS measurements were converted into decimal degrees for statistical analysis using SaTScan™ Version 7.0 (Kulldorff et al., 2007). For accurate mapping, the decimal degrees were further converted into a projected datum (WGS84 Zone 2S) using the GIS package ArcGIS 9.3 (ESRI, 2008).

2.4. Statistical analysis

Spatial clustering was assessed using SaTScan™, with the spanning window set for circular, as previously used in LF spatial studies (Washington et al., 2004). If the analysis identified significant primary "most likely" clusters ($P < 0.05$) as well as over-lapping secondary clusters that were significant, only the "most likely" cluster was included in the results.

The two scenarios explored by virtual analysis were "community-based" whereby exposure could be occurring around the household, or "school-based" whereby a child is being exposed whilst at school.

The case definitions used for both scenarios were:

(1) to identify microfilariae clustering: microfilariaemics as the case, every other individual in the house defined as a control;
(2) to identify antigen clustering: CFA positives as the case, every other individual in the house defined as a control;
(3) to identify antibody clustering: antibody positive child aged 10 years and below defined as the case, every other individual in the house defined as a control;
(4) to identify clustering of antigen and antibody cases: the cases were both CFA positive individuals of any age and an antibody positive child aged 10 years and below, every other individual in the house defined as a control; and,
(5) to identify clustering of microfilaria and antibody positive cases: the cases were both microfilaraemic individuals and an antibody positive children aged 10 years and below, every other individual in the house defined as a control.

Clusters identified for definitions 1–3 will be termed 'cluster'. Clusters identified from definitions 4–5 will be termed 'dual clusters' and refer to a cluster of two properties complementing each other, rather than representing two clusters merged together.

All villages were mapped using ArcGIS 9.3 (ESRI, 2008). Using the radius and centroid outputs from the SaTScan™ analysis, clusters were included on the map, to scale, using the software extension XTools Pro V 4.1.

3. Results

3.1. Study population

The prevalence rates observed for each of the villages are tabulated (Table 2). The highest prevalence was in the village of Fasitoo-Tai, where Mf prevalence reached 3.2% and antibody prevalence in children was 62%.

3.2. Spatial clustering

The relative risk (RR) and size of each primary cluster are outlined in Table 3. The relative risk defined the likelihood of an individual within this area of being infected or exposed, depending on the analysis explored from Section 2.

3.2.1. "Community-based" scenario

Clustering of cases is summarised in Table 3. Households containing infected (CFA positive) individuals showed spatial clustering in the villages of Fasitoo-Tai, Siufaga and Puapua (Fig. 2a, c, and d). LF exposure, defined by anti-filarial IgG_4 antibody positive children, was only evident in Fasitoo-Tai (Fig. 2a and b).

Dual clustering, including households with infected individuals and antibody positive children, was observed in Fasitoo-Tai and Puapua (Fig. 2a and d). Mf positives were detected in Tafua and Fasitoo-Tai (Table 2). Only in Fasitoo-Tai there was a dual cluster between households with Mf positive individuals and antibody positive children.

No significant spatial patterns were identified for either Falefa or Tafua, each with a CFA prevalence of 5.1% and 8.4% respectively (Table 2), although it was observed that a higher number of indi-

Table 3
Summary spatial data of the 5 villages examined. Data presented here include whether a spatial cluster was observed (Y/N), the relative risk (RR) of individuals living within the cluster, and how far the cluster extends in metres (Rad).

Island	Village	Number of HH	"Community-based"					"School-based"		
			Mf	CFA	Bm14	Mf and Bm14	CFA and Bm14	Bm14	Mf and Bm14	CFA and Bm14
Upolu	Fasitoo-Tai $A = 6.8\ km^2$	92	N	Y RR = 3.881 $P = 0.001$ Rad = 1160 $A = 4.2\ km^2$	Y RR = 2.371 $P = 0.004$ Rad = 1160 $A = 4.2\ km^2$	Y RR = 2.517 $P = 0.001$ Rad = 1340 $A = 5.6\ km^2$	Y RR = 2.799 $P = 0.001$ Rad = 640 $A = 1.3\ km^2$	Y RR = 7.292 $P = 0.001$ Rad = 80 $A = 0.02\ km^2$	Y RR = 4.736 $P = 0.001$ Rad = 1380 $A = 6\ km^2$	Y RR = 1.983 $P = 0.001$ Rad = 280 $A = 0.2\ km^2$
	Falefa $A = 6.4\ km^2$	70	N/A	N	N	N/A	N	Y RR = 6.038 $P = 0.001$ Rad = 220 $A = 0.15\ km^2$	N/A	Y RR = 2.781 $P = 0.001$ Rad = 1150 $A = 4.1\ km^2$
	Siufaga $A = 4\ km^2$	75	N/A	Y RR = 82.167 $P = 0.006$ Rad = 0 $A = 0$	N	N/A	N	Y RR = 11.290 $P = 0.001$ Rad = 0 $A = 0$	N/A	Y RR = 7.988 $P = 0.001$ Rad = 60 $A = 0.01\ km^2$
Savai'i	Puapua $A = 6.2\ km^2$	88	N/A	Y RR = 14.167 $P = 0.009$ Rad = 470 $A = 0.7\ km^2$	N	N/A	Y RR = 6.833 $P = 0.003$ Rad = 700 $A = 1.5\ km^2$	Y RR = 12.715 $P = 0.027$ Rad = 0 $A = 0$	N/A	Y RR = 24.556 $P = 0.007$ Rad = 0 $A = 0$
	Tafua $A = 19.5\ km^2$	62	N	N	N	N	N	N	N	N

A, size of geographical study area (village) or cluster area, in square kilometres, estimated by ArcGIS software V 9.3. N/A, not applicable; HH, households; Mf, microfilariae; CFA, circulating filarial antigen; Bm14, anti-filarial IgG4 antibodies.

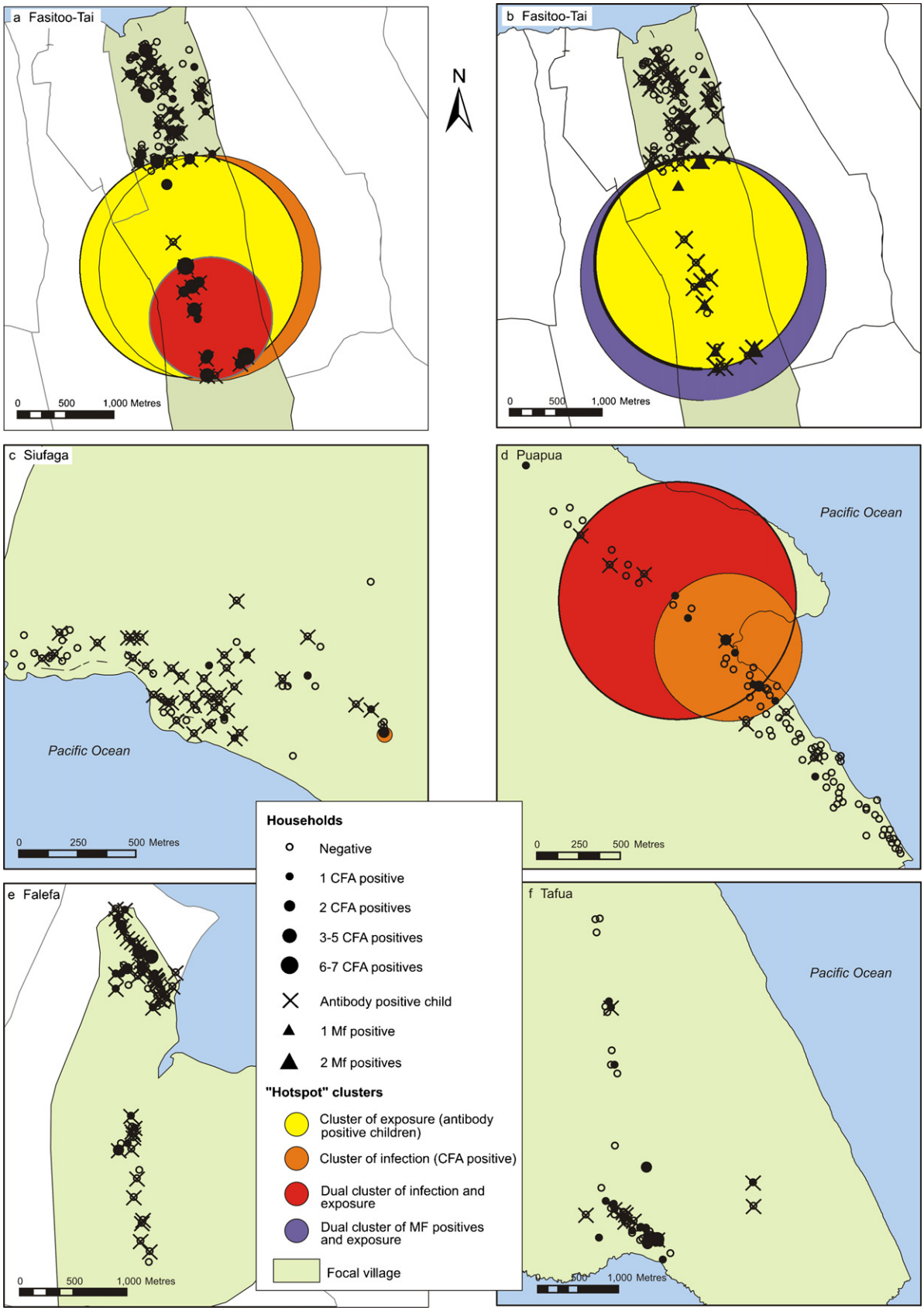

Fig. 2. The "community-based" analysis highlighting the spatial clusters of LF exposure and/or infection in each of the five villages: (a) Fasitoo-Tai, (b) Fasitoo-Tai, (c) Siufaga, (d) Puapua, (e) Falefa, and (f) Tafua. The size of the household (●) or (▲) is relative to the number of circulating filarial antigen (CFA) positive individuals or microfilariae (Mf) positive individuals respectively. Households designated with a cross (×) contained antibody positive children. There were four types of clusters identified including households with CFA positive individuals, with children exposed to LF, a dual cluster of CFA positive individuals and exposed children, and a dual cluster of Mf/CFA positive individuals and exposed children.

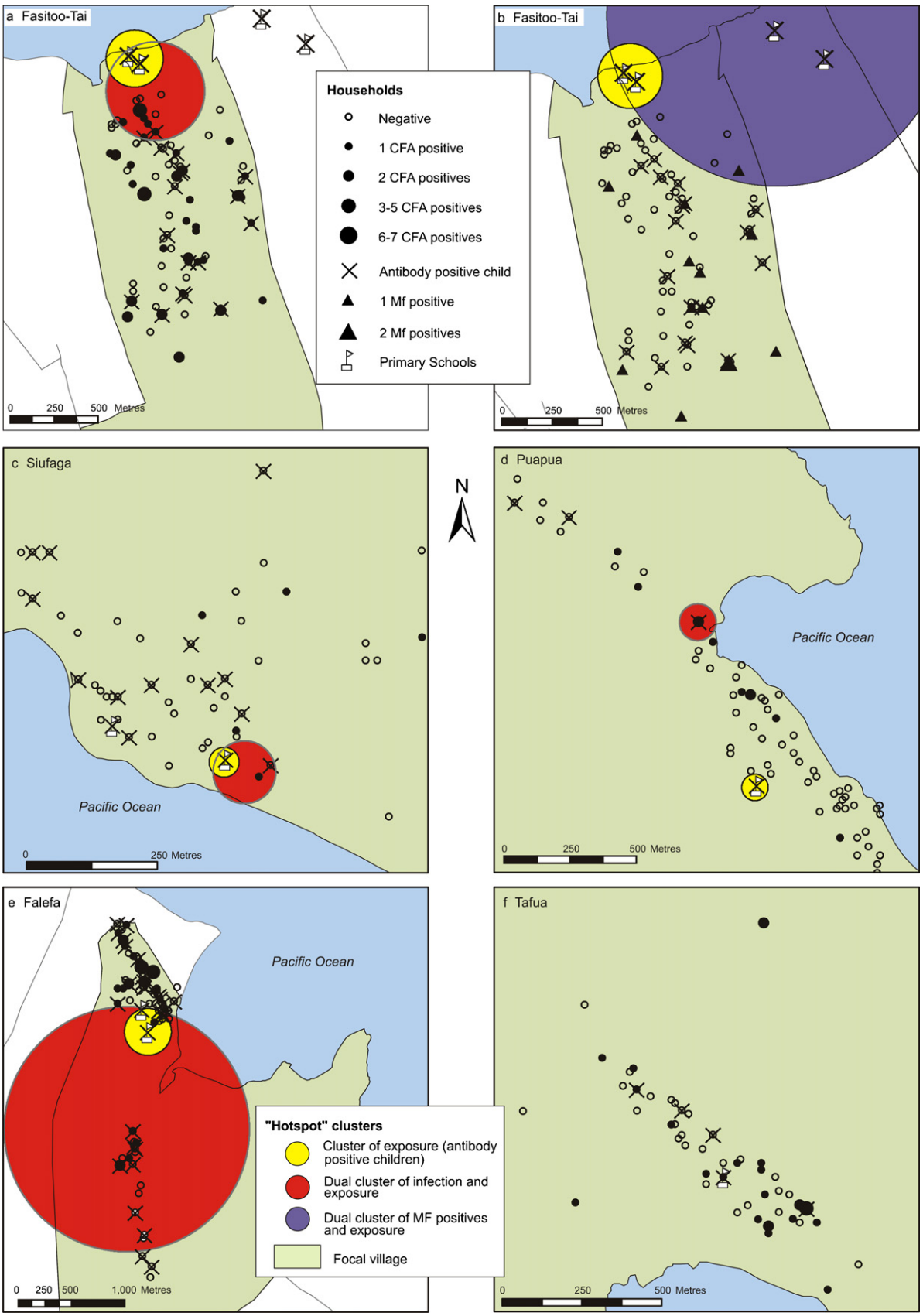

Fig. 3. The "school-based" analysis highlighting the spatial clusters of LF exposure and/or infection in each of the five villages: (a) Fasitoo-Tai, (b) Fasitoo-Tai, (c) Siufaga, (d) Puapua, (e) Falefa, and (f) Tafua. The size of the household (●) or (▲) is relative to the number of CFA positive individuals or Mf positive individuals respectively. Households also bearing a cross (×) contained antibody positive children. Primary schools are designated with a flag. There were four types of clusters identified including households with CFA positive individuals, with children exposed to LF, a dual cluster of CFA positive individuals and exposed children, and a dual cluster of Mf/CFA positive individuals and exposed children. Interestingly, (e) Puapua revealed two separate discrete clusters for antibody exposure and a dual cluster of antibody exposure and CFA positive individuals.

viduals with CFA lived within approximately 400 m of the coast for both villages (Fig. 2e and f).

3.2.2. "School-based" scenario

Different spatial patterns from the "community-based" scenario were observed for the "school-based" scenario. For four villages, excluding Tafua, a spatial cluster of antibody positive children was observed incorporating either one or two primary schools (Table 3 and Fig. 3). In Fasitoo-Tai, Siufaga and Falefa this spatial cluster extended to a dual cluster when infected individuals were included in the analysis (Fig. 3a, c and e), thus widening the geographical limits of the hypothetical area of transmission. In Puapua this dual cluster existed separate from the cluster of antibody positive children (Fig. 3d). Lastly, again, no spatial clustering was observed for Tafua (Table 3 and Fig. 3f).

4. Discussion

This study provides the first evidence of spatial clustering of LF in an *Ae. polynesiensis* vector endemic area and holds profound implications for the future of LF control and active surveillance strategies in Samoa and, indeed, other countries where this vector is endemic. Previously it was believed that the usefulness of vector control was limited in day-biting vector endemic areas and it would require countrywide MDAs to control persistent residual foci of LF (Bockarie et al., 2009). This is because spatial clustering of LF in these areas had not been previously identified, which was concluded to be due to the mobility of infected individuals during the day (Mladonicky et al., 2009). On the contrary, the current study has demonstrated that the household and/or primary school of attendance could still serve as a major site of LF exposure. Therefore there is the potential for introducing vector source reduction campaigns in these foci without needing to target the entire village or country.

The potential for both "community-based" and "school-based" clustering to be observed could relate to the vectors in Samoa; *Ae. polynesiensis* (day-biter) and *Aedes samoanus* (night-biter) (Samarawickrema et al., 1985). Hypothetically, *Ae. polynesiensis* could be responsible for exposure in the "school-based" scenario whereas both vectors could contribute to exposure around the household in the "community-based" scenario. This is because *Ae. samoanus* bites at night and the biting frequency of *Ae. polynesiensis* peaks just after sunrise until approximately 0800 h and again before sunset when individuals are likely to be at home (Bockarie et al., 2009; Samarawickrema et al., 1985). Differences in vector distribution would explain the presence of two discrete clusters observed for the "school-based" scenario in Puapua. However, without an in-depth entomological study within these communities, conclusions as to specific vector contributions to the transmission dynamics cannot be ascertained. Conclusions can only be drawn for the presence of spatial clustering.

Household clustering of LF in a night-biting vector endemic area has long been established (Walter, 1974) and studies have highlighted the necessity in these areas to treat other members of the same household and the nearest neighbours (Washington et al., 2004; Weil et al., 1999). The current data agree with this, however, the spatial clusters observed in Samoa extend past the nearest neighbour suggesting a larger geographical area of potential exposure and treatment. The total area required to target treatment differed in each village, possibly due to the intensity of transmission, since it was clear that the village with the highest prevalence (Fasitoo-Tai) had a larger radial risk of CFA positive individuals and antibody positive children. When the analysis included both CFA positive individuals and antibody positive children a dual cluster was observed with a smaller geographical area. This could potentially represent an area of intense transmission, since rising IgG_4 levels are associated with late exposure or early pre-patent infection (Kwan-Lim et al., 1990; Ottesen et al., 1985).

The data presented here also highlight the need to move away from the original spatial mapping (50 km × 50 km and 25 km × 25 km grids), proposed by the WHO during the beginning of program mapping, to much smaller areas spanning 1 km^2 during active surveillance. This concurs with previous research in Papua New Guinea which demonstrated that the spatial correlation of *Wuchereria bancrofti* Mf density reduced by half over 1.7 km (Alexander et al., 2003). Furthermore, in India, it was recommended to reduce the 25 km × 25 km WHO grids to <10 km^2 (Srividya et al., 2002). These studies correlate with new findings highlighting the heterogeneity of LF transmission over small geographical areas (Gambhir et al., 2010). By reducing the area analysed for spatial mapping, spatial patterns around households can be ascertained, such as the case with Samoa. This allowed detection of ongoing LF transmission in the chosen study areas.

Surveillance strategies rely on the accurate and sensitive detection of LF transmission. The current study suggests the use of antibody serology in children, since a significant relationship was observed between this parameter and Mf/CFA positive individuals within the community. A surveillance strategy tailored to the Pacific called the "draft LF active surveillance strategy for the Pacific Islands and Communities (PICT)", was developed in 2007 and revised in October 2008 (WHO, 2007). This proposed strategy for the Pacific is based on the detection of CFA positive children and, once identified, tracing the potential source of infection from the child's home by testing surrounding households (24 houses or a radius of 200 m) referred to as "close contact testing" (WHO, 2007). The results from the current research highlight the potential for children to be exposed to LF either during the day, whilst at school, or when at home. Therefore, in Samoa, tracing the potential source of infection should occur both from the child's home and the primary school of attendance, which will affect the current surveillance strategy.

Additionally the results from the current study indicate that the affected area could exceed this arbitrary figure of 200 m, such as in Fasitoo-Tai where spatial clusters extended over 1 km, and that additional close contact testing may be required radiating out from the household of the Mf positive individual once identified. The latter conclusion is due to the observation of dual clusters between Mf positive individuals and exposed (antibody positive) children. It could be speculated that the wider radii obtained in the current study may be due to the presence of >1 Mf positive individual contributing to the ongoing transmission, since CFA prevalence exceeded 1% in all villages studied. This would widen the limits of the geographical area where residents are potentially exposed. Consequently, it could be recommended to modify the current LF active surveillance strategy to extend the suggested radius of 200 m if CFA prevalence exceeds 1%.

Another crucial finding from the current study was the dual clustering of infected (CFA positive) and exposed (antibody positive) as well as Mf positive and exposed (antibody positive) children. This favours the potential for antibody serology to be incorporated into the surveillance strategy, which requires validation in other epidemiological settings both in the Pacific and other LF endemic regions of the world. Children would represent the prime cohort as a measure of incidence, which has proven useful in other parasitic diseases, including onchocerciasis (Lindblade et al., 2007).

The differences in spatial cluster radii between the five villages could be due to vector distribution (Samarawickrema et al., 1987), the geographical layout of a village (coastal vs. inland) (Rakai et al., 1974), differing environmental circumstances such as wind patterns (Mahoney and Kessel, 1971), differences in socioenvironmental composite risk indicators (Bonfim et al., 2009), or

the intra-community variations in transmission observed over small distances such as a few households (Gambhir et al., 2010). This may relate to the flight dynamics of the vector, which have been shown to range from a weak flier, only being able to fly a few hundred metres (Reiter et al., 1995), to up to distances of 800 m in the extreme (Honorio et al., 2003). This requires further investigation using climate-based risk models and entomological studies.

It was a concern that the "school-based" scenario may bias the data, since placing the children in their respective schools as their "household" for data analysis could result in a cluster of exposure by default. However, excluding Tafua, there were ≥4 primary or pre-schools recorded for each village, resulting in a broad geographic distribution of cases and controls. There was only one primary school attended in Tafua and no spatial clusters were identified here, possibly because Tafua was the village encompassing the largest geographical area (19.5 km^2). Higher antibody prevalence in children did not appear to influence the detection of clusters, since Puapua had a lower antibody prevalence than the four other villages and clusters were still detectable. The lack of clustering observed for Falefa in the "community-based" scenario may be due to the relative contributions of the vectors to transmission since clustering was observed for the "school-based" scenario. Without entomological studies this cannot be ascertained.

4.1. Conclusions

Collectively, these promising findings are the first evidence of spatial clustering of LF in a day-biting *Ae. polynesiensis* vector endemic area. This research provides important information to give health personnel a starting point for finding Mf index cases as the root of the residual endemnicity or during surveillance, allow for targeted treatment efforts, and potentially incorporate vector control campaigns. This would help staff revise current policies to include: (1) treating households within a certain radius from the index case and (2) possible introduction of vector control, which has been shown to (a) potentially reduce the number of years of MDA required to eliminate transmission, (b) be necessary in those areas where *Ae. polynesiensis* is endemic, (c) be necessary in areas with high vector density, and, (d) lessen the likelihood of acquiring drug resistance (Burkot et al., 2006, 2002; Das and Subramanian, 2002; Das and Vanamail, 2008; Kyelem et al., 2008; Lambdin et al., 2009; Michael et al., 2004; Molyneux et al., 1999; Reuben et al., 2001). Lastly, the spatial relationships observed between antibody positive children and CFA positive or Mf positive individuals enable the opportunity to further explore the use of antibody serology in active surveillance strategies. Future research could also be conducted in a different *Ae. polynesiensis* endemic country, such as French Polynesia, to validate these results.

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