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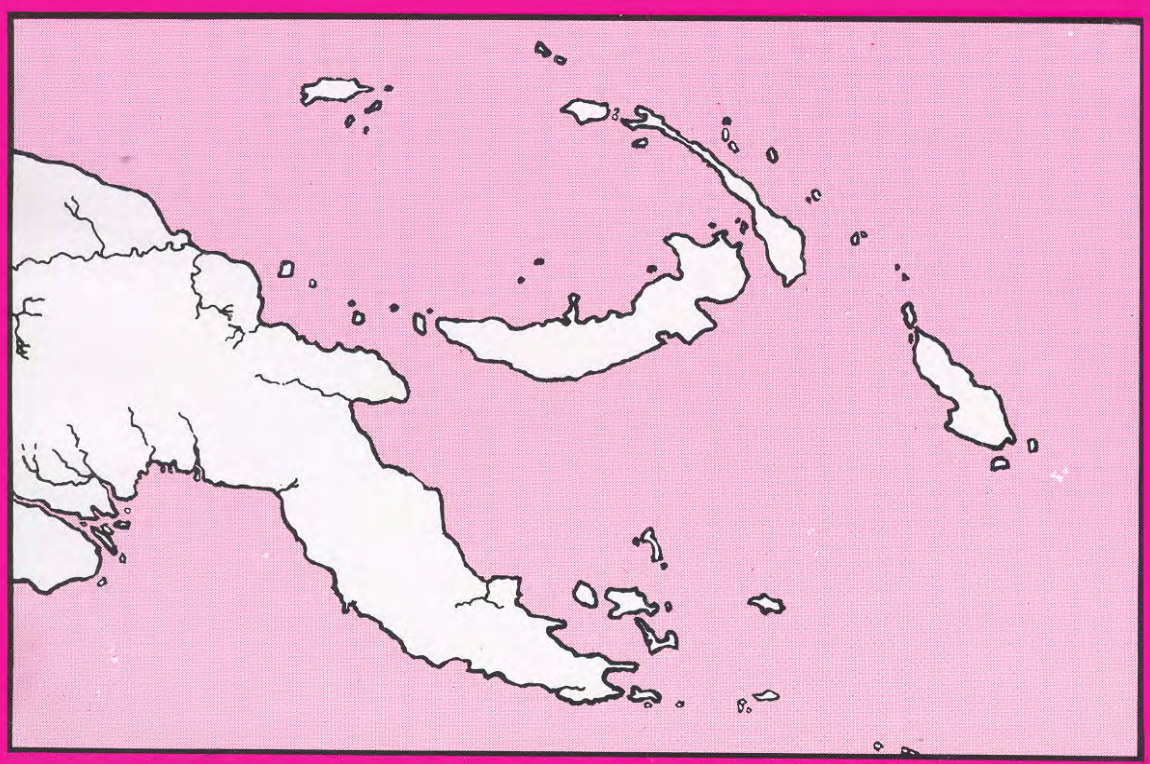

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EDITORIAL

Transmission of HIV from parent to child in Papua New Guinea

In the 1980s when HIV (human immunodeficiency virus) entered Papua New Guinea (PNG) there was great confusion about the disease. Many people thought it was a foreign plague that mainly affected 'sexual deviants' and could not possibly become a problem for PNG, and some thought it was a scourge sent by God as punishment for sin. There was also concern that the HIV epidemic in PNG might follow the course that it had begun to follow in many countries in central Africa. It gradually became accepted that HIV is another sexually transmitted infection (STI), albeit a lethal one with a long incubation period that makes it very tricky to control. It was inevitable that a sexually transmitted disease that kills would become a very emotional issue. The fact that HIV can pass from men to women to infants may make the issues surrounding HIV even more fraught with emotion and guilt.

It is not surprising therefore that HIV transmission to babies attracts a great deal of international and community attention. Even those who take a completely objective view of HIV transmission as a disease feel that it is not fair that 'innocent' babies should become infected inside their mother's womb or simply in the process of breastfeeding.

Whilst everyone agrees that the best way to prevent HIV infection in children is to prevent HIV infection affecting men and women, the focus on secondary prevention – identifying HIV-positive women in antenatal clinics and providing them with antiretrovirals in labour and to the infant soon after birth – persists.

In the current HIV prevalence scenario of about 1% in the general population, 100 of 10000 antenatal mothers will screen positive. If the secondary prevention strategy is carried out with great diligence it has the power to prevent only 9 (12%) to 18 (24%) of the 77 or so potential HIV infections in their infants. Some may be asking how come that there are 77 potential HIV infant infections amongst 100 HIV-positive women when the figures for mother-to-child transmission are usually given as between 30% and 40% – that is, in this case only 30-40 babies – if no prevention of maternal-to-child transmission (PMTCT) is carried out.

Unfortunately more infants become infected than the raw percentage figures would suggest. The figure is inflated by the following factors:

- pregnancy doubles women's susceptibility to receptive transmission of HIV infection (1), and when HIV is spreading amongst young people in a community it is therefore incidence rather than prevalence that determines risk to the fetus; furthermore, a woman may become positive after her first antenatal visit.

- a significant number of infant HIV infections result from new maternal infections late in pregnancy or during breastfeeding.

It is well known that pregnancy and early breastfeeding time is a common risk time for husbands to stray. This is particularly true for young husbands. If a man indulges in unsafe sex outside marriage and gets a new HIV infection and he passes this to his wife during the pregnancy or when she is breastfeeding the virus is much more likely to pass to the baby (2). Infants infected in this way would not be protected by the usual PMTCT program where the blood tests are done at the first visit to the antenatal clinic.

We need to concentrate more on general measures (ie measures which do not rely upon knowing the HIV status of the mother) that lead to less chance of HIV transmission from husband to wife and from mother to baby. Such measures include prevention of all STIs in couples, malaria prevention, promoting exclusive breastfeeding (3) and strengthening family planning services. At the same time these strategies will also result in better reproductive health for all.

Finally 'staying negative' should be a real priority for health promotion in schools,

churches, villages, towns and wherever health workers interact with people, including antenatal clinics. Men need to be encouraged to accompany their wives to antenatal clinics to participate in discussions about safe sex and family planning and to hear the key messages:

- That having sex in pregnancy (and during breastfeeding) is NOT harmful to the fetus/baby (and health workers should even go to the extent of advising more comfortable intercourse positions when having sex in advanced pregnancy, and discussing non-vaginal sex options).
- That unprotected extramarital sex can lead to death of your child, your wife and yourself.
- That family planning gives confidence that there will not be an unintended pregnancy while the new baby is still small. When one asks most PNG women what they will use to prevent pregnancy in the post-partum period, they usually say something like "bai mipela stap tasol" [we will just stay like this]. Women need to be advised that this is dangerous. Probably the husband has not been having much sex during pregnancy, and to prolong post-partum abstinence longer than necessary could tempt any husband more than his ability to resist.

For those who like to seek support from Bible passages St Paul gives good advice to married couples in Corinthians 1, chapter 7, verse 5 "*do not deny yourselves one from the other, except for a short time when you give yourselves to prayer and fasting, but after that come together again, so that Satan cannot tempt you beyond which you can endure".*

Whenever we commence a PMTCT program I believe it should be in conjunction with syphilis detection and treatment (after all syphilis is more prevalent than HIV in our country, can also lead to the death of mothers, fathers and babies and is readily treatable with injections of benzathine penicillin) and with strong 'staying negative' and post-partum family planning programs.

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REFERENCES

1 **Gray RH, Li X, Kigozi G, Serwadda D, Brahmbhatt H, Wabwire-Mangen F, Nalugoda F, Kiddugavu M, Sewankambo N, Quinn TC, Reynolds SJ, Wawer MJ.** Increased risk of incident HIV during pregnancy in Rakai, Uganda: a prospective study. *Lancet* 2005:366:1182-1188.

2 **Wawer MJ, Gray RH, Sewankambo NK, Serwadda D, Li X, Laeyendecker O, Kiwanuka N, Kigozi G, Kiddugavu M, Lutalo T, Nalugoda F, Wabwire-Mangen F, Meehan MP, Quinn TC.** Rates of HIV-1 transmission per coital act, by stage of HIV-1 infection, in Rakai, Uganda. *J Infect Dis* 2005;191:1403-1409.

3 **Iliff PJ, Piwoz EG, Tavengwa NV, Zunguza CD, Marinda ET, Nathoo KJ, Moulton LH, Ward BJ, Humphrey JH, ZVITAMBO Study Group.** Early exclusive breastfeeding reduces the risk of postnatal HIV-1 transmission and increases HIV-free survival. *AIDS* 2005;19:699-708.

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The epidemiology of malaria in the Papua New Guinea highlands: 5. Aseki, Menyamya and Wau-Bulolo, Morobe Province

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SUMMARY

Although not strictly a highlands province, Morobe encompasses large highlands areas, the most important being Aseki, Menyamya and Wau-Bulolo. A series of rapid malaria surveys conducted in both the wet and dry seasons found malaria to be clearly endemic in areas below 1400 m in Menyamya and Wau-Bulolo, with overall prevalence rates in the wet season (25.5%, range: 9.1%-39.2%) greatly exceeding those in the dry season (8.3%, range: 2.4%-22.8%; p <0.001). In the wet season surveys *Plasmodium falciparum* was the clearly predominant species, accounting for 63% of all infections. *P. vivax* increased in frequency in the dry season (from 27% to 46%, p <0.001), while *P. falciparum* and *P. malariae* decreased. In line with past surveys a low prevalence of malaria was found in the Aseki area. Malaria was found to be the main source of febrile illness in the wet season with at least 60% of measured or reported fever associated with parasitaemia. Other causes of febrile illness dominated in the dry. In villages with parasite prevalence rates <20% mean haemoglobin levels and prevalence of severe anaemia were strongly correlated with overall parasite prevalence. In addition concurrent malarial infections were associated with a strong reduction of individual haemoglobin levels (–1.2 g/dl) and there was increased risk of moderate-to-severe anaemia with concurrent malaria. Malarial infections are thus the most significant cause of febrile illness and anaemia in the highlands fringe populations in Morobe. As a consequence all villages below 1500-1600 m in Morobe Province should be included in malaria control activities.

Introduction

Morobe Province, although not strictly a highlands province, encompasses two major mountain ranges – the continuation of the central highlands spine to form the Owen Stanley Range and in the north of the province the Sarawaget Range. Overall, 62% of villages in Morobe are situated at altitudes above 600 m (34% >1200 m, 28% 600-1200 m) and can thus be considered as highland or highland fringe areas, according to the Papua New Guinea (PNG) National Mapping Bureau. With the exception of the hinterland of the Finschhafen coast most villages in the Sarawaget Range are situated in steep, narrow valleys. In the central mountain range, however, there are large intermontane valleys with sizable populations in the Wau-Bulolo, Garaina and to a lesser extent the Aseki-Menyamya areas. The Morobe highland areas are also of substantial economic importance with agricultural cash crops such as coffee and gold mining in the Wau and Bulolo area.

Most highland areas in Morobe have moderate to high rainfall (2000-3000 mm) with moderate seasonality, according to the PNG Resource Information System (PNGRIS) database (1). In parts of Menyamya and Wau-Bulolo, the valleys are considerably drier with annual rainfall of 1500-2000 mm with similar seasonality. The Aseki area in contrast has very high rainfall (4000-5000 mm) with no clear seasonal

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variation.

Of all the highland areas in Morobe, the malaria situation in the Wau-Bulolo and Aseki-Menyamya areas has received the most attention because of the very high incidence of hyperreactive malarious splenomegaly (HMS) – formerly known as tropical splenomegaly syndrome – in these areas (2). This condition is characterized by massively enlarged spleen in adolescents and adults and is thought to be caused by an overproduction of IgM in response to recurrent malaria infections (3). There is familial and ethnic clustering of the disease indicating a genetic basis (4), with the Watut and Tauri-Menyamya areas having some of the highest prevalence rates in the world (2). The presence of this condition has important consequences for landowners from affected communities as it prevents employment in major economic projects such as the Wau gold mines. Control of malaria in the area is thus not only of great health but also of economic benefit to local people.

Early surveys carried out in 1966-1967 in 3 villages in the Watut area showed monthly malaria prevalence rates (PR) fluctuating between 1.9% and 27.9% with *Plasmodium vivax* the most common parasite, accounting for 51.4% of all infections (2). Enlarged spleens were found in up to 75% of people over the age of 5 years. A second survey in 1982 found similar parasite rates, but species composition had shifted to clear *P. falciparum* dominance with >90% of infections due to this species. Similar results were found in a series of surveys in April 1983 in the Menyamya area with an overall parasite rate of 39% and a spleen rate (SR) of 51% (5). Little indication of local transmission was, however, found in surveys in 1972 and 1979 in Aseki (overall PR 0.8%, SR generally <10%) (5).

Although lowland and coastal areas of Morobe were included in the malaria control program, DDT spraying in highland areas was restricted to the urban and periurban areas around Wau and Bulolo and along the road to Mumeng. Extension into other highland areas was planned for 1976-1977 (6), but due to the scaling down of control in the late 1970s was never fully implemented. Vector control activities in the entire province ceased in the early 1980s.

Since the time of those surveys, no further malaria studies have been conducted in any of the highland areas of Morobe Province and accurate information on the current level of malaria transmission in these areas is thus lacking. In order to document changes in malaria transmission over the last 20 years, we conducted a series of wet and dry season malaria surveys in the Bulolo and Menyamya districts.

Methodology

The surveys were conducted using the sample methodology as applied in earlier highlands surveys (7-9). Within selected villages a household-based sampling strategy was used in order to achieve a sample as representative as possible of the entire village population. From each selected household, every member who could be reached during the stay in the village was included in the survey. If the village had less than 200 inhabitants, complete sampling of the village was attempted.

From each individual of the household, demographic data were recorded, a thick and thin blood film was prepared, the spleen palpated in a lying position and axillary temperature taken. Haemoglobin levels were measured using the Hemocue system (HemoCue AB, Ångelholm, Sweden). Symptomatic individuals were treated as indicated by clinical presentation. A short questionnaire on current symptoms, past malaria episodes, treatment, use of bednets and recent travel was administered to each participant or their guardian.

Giemsa-stained blood films were examined under the microscope for 100 thick-film fields under oil immersion before being declared negative. The parasite species in positive films were identified and densities recorded as the number of parasites per 200 white blood cells (WBC). Densities were converted to the number of parasites per µl of blood assuming 8000 WBC per µl. The slides were read at the Papua New Guinea Institute of Medical Research (PNGIMR) in Goroka and Madang by experienced microscopists. A more detailed description of the survey methodology is found in Mueller et al. (7).

Data entry was done at the PNGIMR in Goroka using a double entry system. Statistical analyses were done using STATA 7.0 (Stata Corp., College Station, TX) and

SPlus (Insightful Corp, Seattle, WA) statistical packages. Chi squared tests and logistic regression analyses were used for categorical variables. Continuous variables were investigated using Student's t-test, linear regression and analysis of variance (ANOVA). Haemoglobin values were adjusted for age and gender effects using regression splines.

Results

A total of 2676 people were surveyed during the 16 surveys in 11 different villages (Table 1; Figure 1). Of all participants 49% were female and 16% were aged <5 years, 17% 5-<10 years, 22% 10-20 years and 45% over 20 years. The surveys included the village of Yatitangwa, situated 6 km south of

TABLE 1

SUMMARY OF RESULTS OF PARASITOLOGY SURVEYS: MENYAMYA AND BULOLO

Village	N	Temperature >37.5ºC (%)	Fever in last 3 days (%)	PR (%)	Parasite species Pf/Pv/Pm/Po	SR (%)	Mean Hb (g/dl)	
							Male	Female
Dry season (August-September 2003)								
Yatitangwa (Gulf)	107	6.5	23.4	10.3	8/13/0/0	0.0	10.8	10.3
Wapa	126	2.4	27.0	2.4	1/2/0/0	2.4	13.1	12.7
Ayewa	129	1.6	15.6	5.4	2/5/0/0	0.8	12.5	11.9
Aseki Station	177	0.6	19.8	5.1	3/6/0/0	0.0	13.3	12.4
Kwayami	201	1.5	9.4	4.0	4/4/0/0	0.0	13.1	12.0
Hengitawa	183	7.7	21.3	5.5	8/3/0/0	0.0	11.5	11.0
Nami Compound	195	4.5	20.0	9.2	7/10/1/0	2.5	12.2	11.6
Pararoa CS	218	2.8	18.8	4.1	7/3/0/0	8.3	13.5	12.6
Patep	250	3.6	12.4	22.8	31/27/1/0	26.8	10.7	10.0
Wet season (May 2004)								
Hengitawa	159	3.1	5.0	28.9	29/14/5/0	20.8	12.2	11.9
Kwayami	160	0.6	4.4	9.4	12/2/1/0	5.6	13.2	12.3
Nami Compound	125	6.3	6.3	27.2	10/17/7/0	14.2	12.7	11.6
Pararoa CS	151	9.3	10.6	37.1	43/22/6/0	17.2	11.2	11.1
Wandumi	186	3.6	7.2	9.1	11/7/2/0	15.9	11.7	11.5
Nauti	51	1.8	3.6	17.7	8/3/0/0	46.4	12.1	11.1
Patep	258	5.0	5.4	39.2	80/18/5/3	23.4	11.3	10.6

N = number surveyed
PR = prevalence rate (of malaria parasitaemia)
Pf = *Plasmodium falciparum*; Pv = *Plasmodium vivax*; Pm = *Plasmodium malariae*; Po = *Plasmodium ovale*
SR = spleen rate
Hb = haemoglobin level
CS = Community School

the Morobe border, in the Kaintiba Local Level Government (LLG) area of Gulf Province (Figure 1). As it is located at a similar altitude and in a similar environment as areas surveyed in Morobe, the data are included in the following in-depth data analyses.

In all surveyed villages malaria infections were detected, with mean prevalence rates ranging from 2.4% in Wapa (Aseki) to 39.2% in Patep (wet season, Mumeng). The overall prevalence rates by village are shown in Figure 2. Significantly higher prevalence rates were observed in the wet season than in the dry season: 25.5% (range 9.1% to 39.2%) vs 8.3% (range 2.4% to 22.8%) ($p < 0.001$) overall and 29.5% vs 9.7% ($p < 0.001$) in villages with surveys in both seasons only. In both seasons the highest prevalence rates were found in Patep village (Table 1). In the dry season there was a significant association between altitude and prevalence ($r = -0.73$, $n = 9$, $p = 0.03$), with 19.1% prevalence in villages <1000 m and 5.5% in villages 1000-1499 m, but only 2.4% in villages above 1500 m. No such association was found in the wet season ($r = -0.10$, $n = 7$, $p = 0.83$).

Infections were strongly age dependent ($\chi^2 = 71.7$, $df = 4$, $p < 0.001$), with infections most prevalent in 2-5 year old children (29.4%), followed by children aged 5-10 years (20.3%), adolescents (10-20 years: 14.1%), infants and toddlers (<2 years: 12.4%) and adults (>20 years: 11.0%). There were no significant changes in the age distribution of slide-positive cases between seasons – likelihood ratio (LR)-test: $\chi^2 = 1.2$, $df = 4$, $p = 0.88$.

All four human malaria species were found during the surveys. The relative frequencies of different species differed significantly between seasons (Fisher exact test: $p < 0.001$). In the wet surveys (May 2004), the

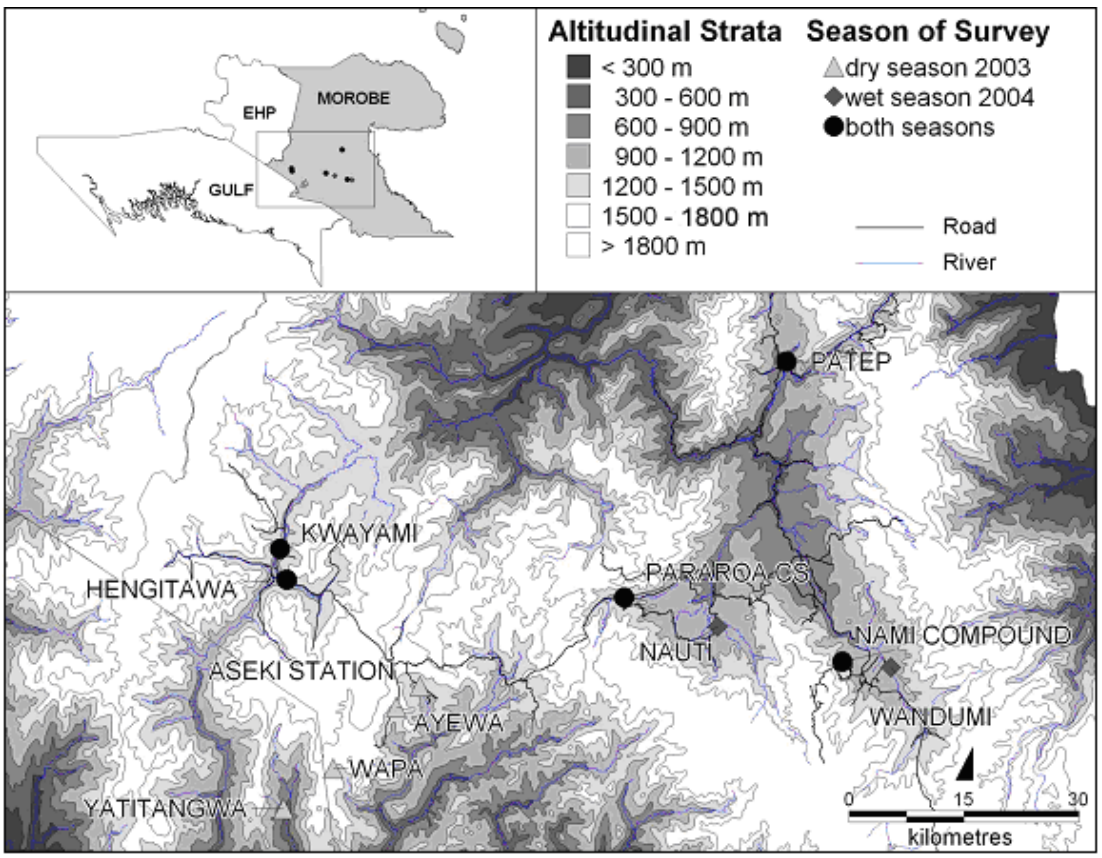

Figure 1. Locations and season of malaria surveys in Morobe Province.

clearly predominant species was *P. falciparum,* which accounted for 63% of infections, followed by *P. vivax* (27%), *P. malariae* (9%) and *P. ovale* (0.3%). In the dry season surveys (August-September 2003), however, *P. vivax* increased in frequency (46%), while *P. falciparum* (52%) and *P. malariae* (1%) decreased. 3.0% of all infections in the dry and 9.4% in the wet season were mixed. The relative frequency of *P. vivax* also increased with altitude (<1000 m: 30%, 1000-1499 m: 41%, ≥1500 m: 67% of all positive cases were infected with *P. vivax*, p = 0.03), while that of *P. falciparum* decreased (<1000 m: 71%, 1000-1499 m: 59%, ≥1500 m: 33%, p = 0.01). Similarly, the prevalence of *P. vivax* among slide-positive cases decreased significantly with increasing age (<5 years: 56%, 5-9 years: 36%, 10-19 years: 29%, ≥20 years: 23%, χ^2 = 30.2, df = 3, p <0.001), while that of *P. falciparum* increased.

The majority of infections were of low density: 59.0% of infections were sparse (<500/µl), 11.7% light (500-999/µl), 23.6% moderate (1000-9999/µl) and 5.6% heavy ((≥10,000/µl), with no significant differences between seasons (χ^2 = 1.7, df = 3, p = 0.65). Densities of *P. falciparum* infections were higher than those of *P. vivax* and *P. malariae* infections (geometric mean 437, 322 and 216/µl, respectively), but the difference did not reach statistical significance (p >0.05). Mixed infections had significantly higher densities than single infections (1199 vs 362/µl, p <0.001). There were highly significant differences in intensity of infection among age groups ($F_{3,400}$ = 18.4, p <0.001). Infections were heaviest in children <5 years and 5-10 years (968 and 488/µl, respectively) and lightest in adolescents and adults (299 and 198/µl).

Spleen rates in the different villages ranged

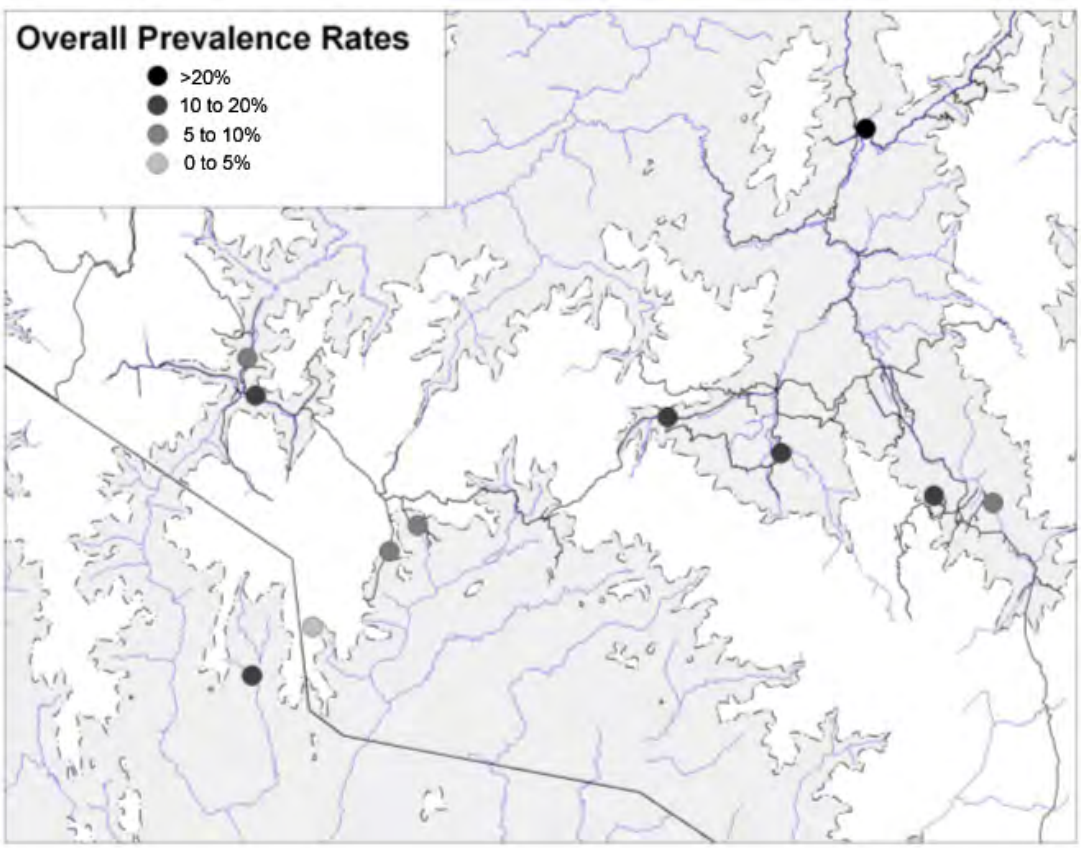

Figure 2. Overall prevalence rates of malaria infections in the 2003-2004 surveys.

from none in higher villages in the dry season surveys to 46% in the wet season in Nauti village. The rate of enlarged spleen was significantly correlated with the parasite prevalence rate overall ($r = 0.62$, $n = 16$, $p = 0.01$). The average size of an enlarged spleen was 1.8 (Hackett's grade). A large spleen was highly significantly associated with a concurrent infection (25.1% vs 8.4%, $p < 0.001$). There was no difference in this association among the different *Plasmodium* species.

In villages with surveys in both seasons, significantly more fevers were reported in the dry season than in the wet season surveys (Table 2: 17.8% vs 6.2%, $p < 0.001$), despite low parasite prevalence rates. Within seasons there were no associations of the prevalence of reported fevers with overall parasite rates ($p > 0.3$). However, prevalence of measured fevers (>37.5°C) was significantly correlated with the parasite rate ($r = 0.50$, $p = 0.05$) with no significant difference in prevalence of measured fevers between seasons (Table 2: 3.4% vs 4.4%, $p = 0.16$). Among all parasite-positive cases only 11.7% had a temperature >37.5°C at the time of the survey; another 6.3% reported fever in the last 3 days. There was no difference in the prevalence of measured fevers among slide-positive people between seasons ($p = 0.61$) but, in parallel with the general trend for reported fevers, parasitaemic people reported febrile symptoms significantly more frequently in the dry season, both overall (28.0% vs 13.3%, $p < 0.001$) and in villages with surveys in both seasons (Table 2: 24.5% vs 12.7%, $p = 0.006$). The occurrence of febrile symptoms was strongly related to the intensity of infection ($\chi^2 = 68.9$, $df = 2$, $p < 0.001$): 69.6%

TABLE 2

DIFFERENCES IN MALARIAL INFECTIONS AND ASSOCIATED MORBIDITY BETWEEN SEASONS IN VILLAGES WITH SURVEYS IN BOTH SEASONS

	Dry 2003 n = 1047	Wet 2004 n = 853	p value
Prevalence rate (%)	9.7	29.5	<0.001
Plasmodium falciparum (%)	53.7	63.5	0.08
Plasmodium vivax (%)	44.3	26.6	<0.001
Plasmodium malariae (%)	1.9	8.8	0.02
Spleen rate (%)	8.6	17.3	<0.001
Mean Hb (g/dl)	11.8	11.7	0.20
δHb with concurrent infection (g/dl)	1.65	1.41	0.38
Proportion of infections with temperature >37.5 (%)	10.8	11.1	0.93
Proportion of infections with fever in last 3 days (%)	24.5	12.7	0.006
Prevalence of temperature >37.5 (%)	3.4	4.4	0.16
Prevalence of reported fevers in last 3 days (%)	17.8	6.2	<0.001
Proportion of temperature >37.5 with parasitaemia (%)	26.8	66.7	<0.001
Proportion of reported fevers with parasitaemia (%)	14.7	60.4	<0.001
Reported 'malaria episode' in last 2 weeks (%)	15.3	33.6	<0.001

Hb = haemoglobin level
δHb = decrease in Hb

of cases with a parasite density ≥10,000/µl reported being febrile, but only 10.7% of those with densities <1000/µl, with no difference between seasons.

The proportion of fevers associated with malaria infections was significantly different between seasons. In the wet 66.7% of measured and 60.4% of reported fever cases had concurrent malarial infections compared to 26.8% and 14.7%, respectively, in the dry season ($p < 0.001$, Table 2).

Mean haemoglobin (Hb) levels in a village (Table 1) were negatively correlated with the prevalence of malarial infections ($r = -0.52$, $n = 16$, $p = 0.038$) and ranged from 10.4 g/dl in Patep (PR 22.8%) to 13.2 g/dl in Pararoa (PR 4.1%). The village mean Hb level decreased by 1.03 g/dl per 10% increase in parasite prevalence rate in surveys with an overall PR <20%. At prevalence rates >20% no significant further decrease was observed (–0.16 g/dl per 10%, $p = 0.97$). Hb values were significantly lower for women than men (11.5 vs 12.2 g/dl, $t_{2698} = 9.0$, $p < 0.001$) and there was a highly significant increase of Hb with increasing age (test for trend: $z = 18.84$, $p < 0.01$). All subsequent analyses of individual Hb levels and parasitaemia were therefore done on age- and sex-adjusted values.

Concurrent plasmodial infection was associated with a significant decrease in Hb of 1.2 g/dl (CI_{95} [1.0-1.4], $p < 0.001$). Infection with *P. falciparum* was associated with a larger drop in Hb than with *P.* vivax (–1.3 vs –0.7 g/dl, $p = 0.007$). The (adjusted) decrease in haemoglobin was strongly dependent on levels of parasitaemia (<500/µl: –0.9, 500-999/µl: –1.5, 1000-9999/µl: –1.3, ≥10,000/µl: –2.2 g/dl, $F_{3,2632} = 4.2$, $p = 0.006$).

With the exception of Yatitangwa village in Gulf Province, which has an exceptionally high prevalence (14.0%) of moderate-to-severe malarial anaemia (SMA) cases (SMA: Hb <8.0 g/dl), the prevalence of SMA was significantly correlated with overall parasite prevalence ($r = 0.65$, $n = 16$, $p = 0.008$). SMA was observed in 2.6% of participants in surveys with PR <10% and in 6.2% in all other surveys. Within the survey, concurrent parasitaemia was a highly significant risk factor for SMA – adjusted odds ratio (AOR) 2.9 (CI_{95} [1.9-4.5], $p < 0.001$). The risk of SMA increased significantly with increasing intensity of infection (<500/µl: AOR = 2.2, 500-999/µl: AOR = 2.3, 1000-9999/µl: AOR = 3.4, ≥10,000/µl: AOR = 11.6; LR-test: $\chi^2 = 9.7$, df = 3, $p = 0.02$).

When people were asked whether they had had malaria in the two weeks prior to the survey between 5% (Pararoa, dry season) and 47% (Hengitawa, wet season) of people reported having had a 'malaria' episode (Table 3). The number of reported malaria episodes was significantly correlated with the prevalence of infection found in a village ($r = 0.60$, $n = 16$, $p = 0.01$). Of all people reporting a malaria episode, however, only 55% went to an aid post/health centre/hospital for treatment. There was no difference in treatment seeking between seasons. Within surveys, a reported malaria episode in the previous two weeks was highly significantly associated with an increased risk of infection (adjusted OR = 1.61, CI_{95} [1.26-2.07], $p < 0.001$), irrespective of whether treatment was sought or not. Of all people who reported previous antimalarial drug use, 25.0% had a positive blood slide in the survey.

The numbers of people in each village sleeping under a bednet varied greatly between surveys, ranging from none in Wapa and Nauti to 90% in Patep (dry season, Table 3). Most of the nets used were non-treated or not re-treated and the reported use of bednets did not significantly alter risk of infection with malaria (adjusted OR = 0.8, CI_{95} [0.6-1.1], $p = 0.19$).

The people in Aseki and Menyamya (Kome and Wapi LLGs) often spend their nights away from the villages, with 49% (range 16-73%) of people reporting regularly sleeping in garden houses. Less people did so in the Watut, Wau and Mumeng villages, in particular in the wet season (Table 3). In these villages, sleeping in a garden house was, however, not associated with a change in risk for malarial infection (adjusted OR = 0.85, CI_{95} [0.63-1.2], $p = 0.29$).

Discussion

The relatively high prevalence of malaria infection, strong age dependence, the high number of asymptomatic infections and considerable prevalence of spleen enlargement all indicate that malaria is clearly endemic in the Wau-Bulolo area. Prevalence rates were, however, found to vary greatly between seasons. In the wet season prevalence rates were comparable to other

TABLE 3

SUMMARY OF VILLAGE CHARACTERISTICS AND MALARIA-RELATED BEHAVIOUR IN MENYAMYA AND BULOLO SURVEY VILLAGES

Village	N	LLG	Altitude (m)	Malaria 'sickness' (%)	Antimalarial use (%)	Bednet use (%)	Slept in garden house (%)
Dry season (August-September 2003)							
Yatitangwa (Gulf)	107	Kaintiba	710	31.1	27.4	17.9	43.0
Wapa	126	Aseki	1620	9.5	3.2	0.0	73.2
Ayewa	129	Aseki	1450	11.1	0.0	0.8	57.0
Aseki Station	177	Aseki	1270	19.8	11.6	10.4	16.4
Kwayami	201	Kome	1180	16.9	14.0	29.9	67.8
Hengitawa	183	Wapi	1160	19.1	14.4	28.7	58.2
Nami	195	Wau	1320	19.1	12.7	42.1	52.6
Pararoa	218	Watut	1210	5.1	2.9	17.9	54.1
Patep	250	Mumeng	740	17.2	21.9	89.6	10.7
Wet season (May 2004)							
Hengitawa	159	Wapi	1160	47.2	87.7	8.8	44.3
Kwayami	160	Kome	1180	17.5	39.3	14.4	31.0
Nami	125	Wau	1320	38.6	55.2	23.2	8.3
Pararoa	151	Watut	1210	40.4	30.7	8.1	26.5
Wandumi	186	Wau	970	27.2	85.0	23.2	0.5
Nauti	51	Watut	980	10.7	56.5	0.0	28.6
Patep	258	Mumeng	740	29.1	62.8	83.5	2.6

N = number surveyed
LLG = Local Level Government

malaria-endemic highland areas at similar altitude (ie, 900-1300 m) in Simbu (9) or Western Highlands Province (7). During the dry season, however, prevalence rates in Morobe Province were much lower, only exceeding 10% in the low-lying village of Patep (740 m). Interestingly the proportion of infection due to *P. vivax*, a parasite that is highly transmissible and prone to relapses from long-lasting liver stages, increased significantly in the dry season. Both overall prevalence rates and seasonal fluctuations in the present surveys are comparable with those found in the 1966-1967 surveys by Crane and Pryor (2) for the Watut area and the wet season prevalence found in Menyamya in 1983 (5), indicating that transmission levels and the seasonality of malaria transmission in the Watut and Menyamya areas have not changed greatly

over the last 20-40 years.

This striking seasonality is most likely linked to the comparatively low annual rainfall (1500-2000 mm) in the surveyed villages in the Wau-Bulolo and Menyamya areas. As in the neighbouring Eastern Highlands, rainfall during the dry season (<100 mm per month) may be insufficient to sustain sufficiently large enough mosquito populations for significant local malaria transmission. At altitudes between 900 and 1300 m, malaria transmission in these areas is thus characterized by moderate to high transmission levels in the wet, but low to very low endemic transmission at the height of the dry season. The drop in transmission levels in the dry season is also reflected in the species composition of malaria infections, with the proportion of infections due to *P. vivax*, which is more easily transmitted and has long-lasting liver stages (10), increasing.

As rainfall is considerable higher in other highland areas in Morobe Province, such strong seasonal fluctuations are likely to be restricted to the Wau-Bulolo and Menyamya areas and possibly some lower-lying areas on the northern side of the Sarawaget Range, where rainfall in the dry season can also drop below 100 mm per month (according to data from PNGRIS).

As the Aseki villages were only surveyed during the dry season, no information on seasonality of transmission is available for this area. However, monthly rainfall in Aseki exceeds 200 mm all year round (from the PNGRIS database) and lack of rainfall is thus unlikely to be a limiting factor for mosquito breeding. Consequently, little seasonality in malaria transmission levels is to be expected. The low prevalence rates in the present surveys confirm the low prevalence of malaria in the Aseki area reported by Crane et al. (5). The low malaria prevalence rates in Aseki villages are also in line with the situation observed in neighbouring valleys in the southern highlands fringe of Eastern Highlands, where the prevalence of malarial infections did not exceed 5% during non-epidemic periods in either wet or dry season (8). The reasons for this low prevalence at altitudes where malaria is endemic in other parts of the highlands are not completely clear, but may be related to the limited availability and/or flushing of mosquito breeding sites in steep terrain with very high rainfall.

With the exception of the survey in Nauti (Watut), the present surveys found substantially lower spleen rates than Crane et al. measured in earlier surveys (2,5). There are several possible explanations for this. On the one hand, there is now a much better awareness among the affected population, local health professionals and major employers, such as Morobe Goldfields, of the danger posed by HMS. While using long-term antimalarial prophylaxis to treat adults with large spleens was rare in 1983 (5), this is now common practice and explains at least part of the reduction in spleen rates. It can, however, not be ruled out that our measurements are underestimations. Measuring spleens in adults, the main sufferers of HMS, is often made difficult by well-developed abdominal muscles and specialized training is required to detect enlarged spleens consistently.

The data on febrile illness indicate that while malaria is the main source of febrile illness in the wet season (ie, the main malaria transmission season), other causes of febrile illness dominate in the dry. There was no difference in the proportion of malaria infections with measured fever between seasons but there was a higher proportion of infections with reported fever in the dry season (see Table 2). The latter may at least partly be due to a generally higher reporting of fevers in the dry season surveys. In addition, concurrent malarial infections were associated with comparable drops in haemoglobin in both seasons. It is thus unlikely that the morbidity associated with malaria infections differs markedly between seasons. The increase in malarial fevers in the wet season is entirely linked to the increase in parasite prevalence (ie, transmission), not due to higher levels of morbidity associated with infections.

The strong association of mean haemoglobin levels and prevalence of severe anaemia in villages with parasite prevalence rates <20%, as well as the strong reduction of individual haemoglobin levels and increased risk of moderate-to-severe anaemia with concurrent malarial infections, indicate that malarial infection is the major cause of anaemia in highlands fringe populations in Morobe. However, the village of Yatitangwa in the Kaintiba area (Gulf Province) shows that other factors, in particular nutrition, may also be important contributors to anaemia. The Kaintiba area

is very marginal with poor soils (according to data from PNGRIS), high levels of malnutrition and low birthweights (11,12), and the low haemoglobin level may thus at least in part be due to bad nutrition or other parasitic infections.

The prevalence (25%) of malaria infections in people who reported previous antimalarial drug use indicates that there may be considerable problems with drug resistance and/or treatment compliance. In recent in vivo drug resistance studies in another highlands fringe community in Karimui, south Simbu, over 20% of children treated with chloroquine plus Fansidar did not successfully clear their malaria parasites, and high levels of molecular markers of drug resistance were found in both *P. falciparum* and *P. vivax* (J. Marfurt, I. Mueller, B. Genton et al., submitted manuscript), indicating that drug resistance may evolve as quickly in areas of moderate as of high endemicity. Improvement of malaria diagnosis and monitoring of treatment success should also be integral parts of a malaria control strategy in the highland areas of Morobe.

Although bednets did not show any association with protection against malaria infections, this does not contraindicate their possible use in malaria control in highland areas in Morobe. Most nets were untreated and use was infrequent. If high coverage of insecticide-treated nets can be achieved they may be an important control tool, particularly in remote areas.

Conclusions and Recommendations

Using data from the present surveys we can define 3 areas with different risks of malaria (Figures 3 and 4).

Malaria moderate to highly endemic and seasonal (<1200-1400 m)

In these areas, malaria is moderate to highly endemic (10-30%) in the wet season. In villages above 900-1100 m, transmission may, however, be markedly reduced in the dry season. Malaria is the main source of febrile illness during times of heightened transmission and is associated with low Hb values. In the dry season malaria is a major source of febrile illness only in villages below 900-1100 m. In the area surveyed, the main valley areas in the Wau-Watut-Mumeng area as well as the lower-lying villages in the Menyamya area fall into this category (Figure 3).

As most villages in the Wau-Watut-Mumeng area have reasonable access, indoor residual spraying (IRS) of long-lasting insecticides is the control method of choice, especially if the program could be coordinated with and supported by major industrial partners such as Morobe Goldfields. Studies in highland areas of Africa (13) showed IRS to be more cost-effective in preventing malaria in highland areas than insecticide-treated nets (ITNs). In order to achieve maximum effectiveness, spraying should be timed to coincide with the start of the rainy season.

The distribution of long-lasting insecticide-treated nets (LLINs) may be an alternative, particularly in remoter areas. However, in order to be fully effective high coverage rates (>80%) need to be achieved. As these nets are effective for 3 years or longer, they are logistically easier to implement and maintain than yearly IRS and are thus preferable in remoter areas with limited access and infrastructure.

Low malaria transmission with potential risk of epidemic outbreaks (1300-1600 m)

In these areas prevalence rates remain <10% all year round. However, in years with exceptionally favourable climate epidemic outbreaks with substantially higher burdens of infections and illness may be possible. In areas with seasonal rainfall patterns, transmission is likely to decrease substantially in the dry season. Outside possible epidemics malaria is not a major source of febrile illness, haemoglobin levels are substantially higher than in moderate to high transmission areas and moderate-to-severe anaemia (Hb <8 g/dl) is rare. This area includes higher areas in Wau-Bulolo and Menyamya as well as the Aseki area (Figure 3).

Although IRS is very suitable to control malaria and prevent epidemic outbreaks in highland areas of low transmission, the logistical challenges to implement and sustain yearly IRS in remoter parts of PNG can be formidable and LLINs may be the better choice. For the Aseki and Menyamya areas, we thus suggest the implementation of LLINs, while in the more accessible Wau-

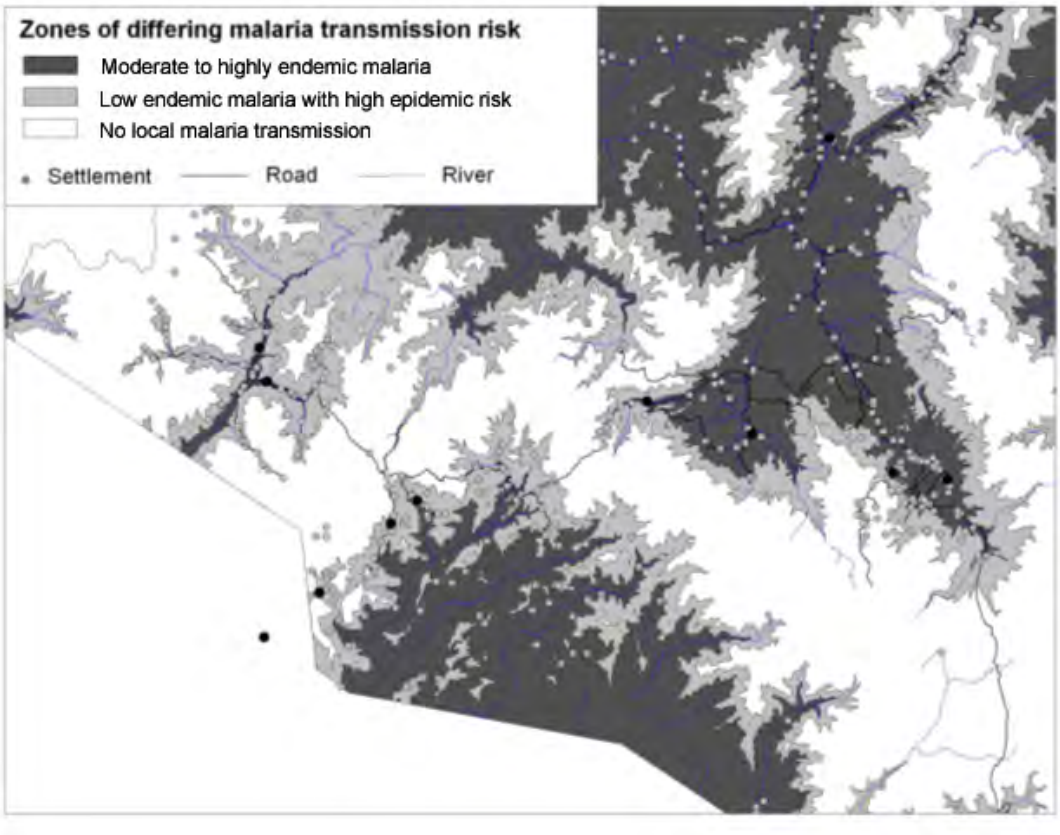

Figure 3. Zones of different malaria risk in Bulolo and Menyamya districts.

Watut-Mumeng areas the intervention chosen for areas moderate to highly endemic for malaria (IRS or LLINs) should be extended to areas of low transmission potential.

Very low or no malaria transmission (areas above 1600 m)

As only one village (Wapa, PR 2.4%) was surveyed in this altitudinal zone, results from surveys at similar altitudes in other highland provinces (7-9) are thus included in conclusions and recommendations.

In villages above 1600 m local transmission is normally restricted to times of exceptional climatic events or villages with lower-lying gardens. Most malaria cases are thus linked to travel to lower-lying areas. Epidemic outbreaks may occur in villages above 1600 m, but may be linked to transmission in lower-lying garden areas (14). In these areas, malaria is not a significant source of illness. Nevertheless most fevers are treated with antimalarials.

Vector control in neighbouring lower-lying areas should prevent malaria transmission from spreading into higher altitudes. Prompt diagnosis and treatment of imported cases in combination with epidemic surveillance and control are thus the main aspects of malaria control in these areas.

Although other highland areas in the province were not included in the present survey, we believe that the above recommendation may also be applied to higher altitude settlements in the Garaina (Waria LLG) and Sarawaget Ranges (Hube, Seko Sialum, Deyamos Yus, Wantoat-Leron, Wain-Erap and Nabak LLGs). Consequently, we would advocate that all villages below 1500-1600 m in Morobe Province should be included in malaria control activities (Figure 4). As most of these areas are rather remote and difficult to access, long-lasting insecticide nets seem the most appropriate control measure. In addition, areas between 1400 and 1700 m should be included in provincial epidemic surveillance and control

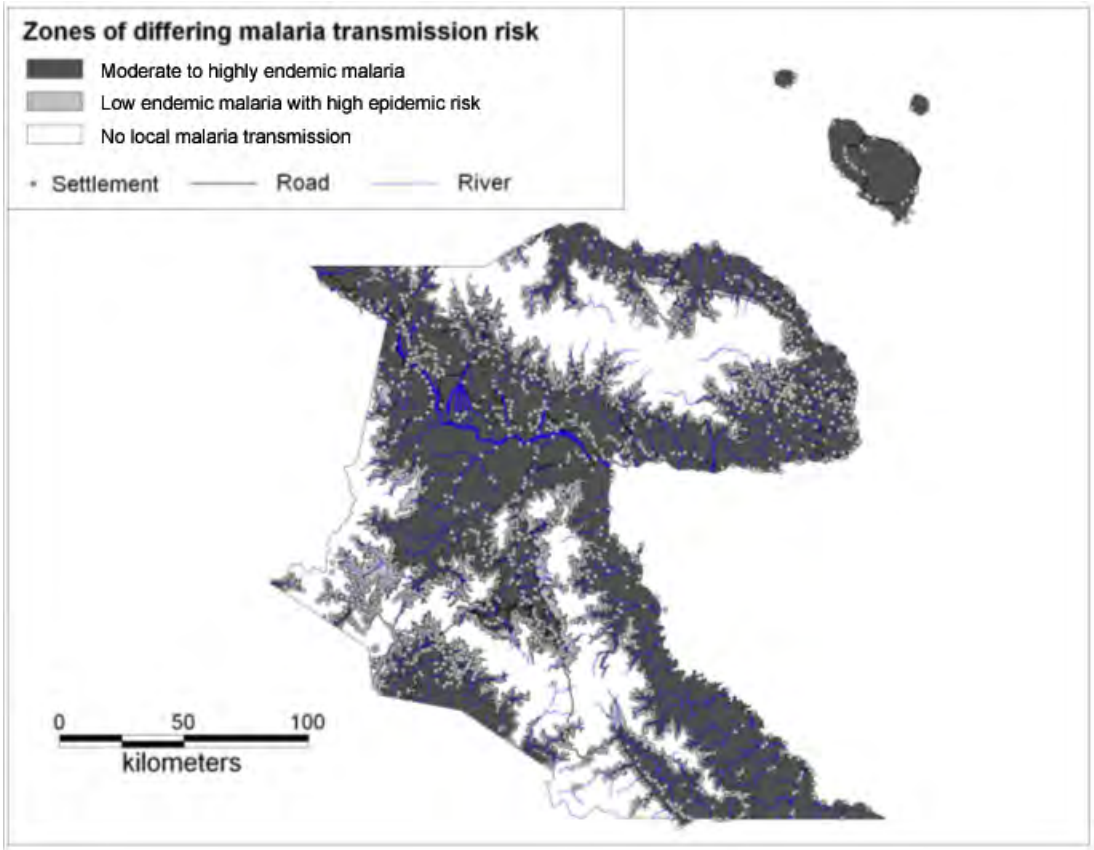

Figure 4. Zones of different malaria risk in Morobe Province.

mechanisms.

REFERENCES

1 **Bellamy JA, McAlpine JR.** Papua New Guinea Inventory of Natural Resources, Population Distribution and Land Use Handbook, 2nd edition. PNGRIS Publication No 6, 1995. Canberra: AusAID, 1996:155p.

2 **Crane GG, Pryor DS.** Malaria and the tropical splenomegaly syndrome in New Guinea. *Trans R Soc Trop Med Hyg* 1971;65:315-324.

3 **Crane GG.** Hyperreactive malarious splenomegaly (tropical splenomegaly syndrome). *Parasitol Today* 1986:2:4-9.

4 **Crane G.** The genetic basis of hyperreactive malarious splenomegaly. *PNG Med J* 1989;32:269-276.

5 **Crane G, Gibson D, Verrall J, Barker-Hudson P, Barker-Hudson BE, Charlwood D, Heywood P.** Malaria and tropical splenomegaly syndrome in the Anga of Morobe Province. *PNG Med J* 1985;28:27-34.

6 **Parkinson AD.** Malaria in Papua New Guinea, 1973. *PNG Med J* 1973;17:8-16.

7 **Mueller I, Taime J, Ivivi R, Yala S, Bjorge S, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 1. Western Highlands Province. *PNG Med J* 2003:46:16-31.

8 **Mueller I, Bjorge S, Poigeno G, Kundi J, Tandrapah T, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 2. Eastern Highlands Province. *PNG Med J* 2003;46:166-179.

9 **Mueller I, Kundi J, Bjorge S, Namuigi P, Saleu G, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 3. Simbu Province. *PNG Med J* 2004:47:159-173.

10 **Gilles HM, Warrell D.** Bruce-Chwatt's Essential Malariology, 4th edition. London: Edward Arnold, 2002.

11 **Mueller I, Betuela I, Hide R.** Regional patterns of birthweights in Papua New Guinea in relation to diet, environment and socio-economic factors. *Ann Hum Biol* 2002;29:74-88.

12 **Mueller I, Vounatsou P, Smith T, Allen BJ.** Subsistence agriculture and child growth in Papua New Guinea. *Ecol Food Nutr* 2001;40:367-395.

13 **Guyatt HL, Kinnear J, Burini M, Snow RW.** A comparative cost analysis of insecticide-treated nets and indoor residual spraying in highland Kenya. *Health Policy Plan* 2002;17:144-153.

14 **Mueller I, Kaiok J, Reeder JC, Cortés A.** The population structure of *Plasmodium falciparum* and *Plasmodium vivax* during an epidemic of malaria in the Eastern Highlands of Papua New Guinea. *Am J Trop Med Hyg* 2002;67:459-464.

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The epidemiology of malaria in the Papua New Guinea highlands: 6. Simbai and Bundi, Madang Province

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SUMMARY

Although predominantly a lowland province, Madang also includes highland areas such as Simbai and Bundi along the northern highland fringe. While the malaria situation in the coastal lowlands has been studied in great detail, the current malaria situation in the highland fringe communities has not been studied in depth since the 1960s. A series of recent malariological surveys found that the malaria situation has changed little over the last 40 years in both Simbai and Bundi. In the Simbai area there is little malaria transmission in villages above 1400 m, with a prevalence rate (PR) of 2.5-4.2%. Below 1400 m, however, there is moderate to high transmission (PR 8.6-24.7%) with surprisingly little difference in prevalence rates between survey villages, despite large differences in altitude. Prevalence rates of malaria infection were low in all Bundi villages (2.5-8.5%) with most infections occurring in adolescents and adults, which indicates limited acquisition of effective immunity to malaria and the possibility that many infections are acquired when travelling to the highly malarious lowlands area. Based on spleen rates the lower Simbai area would be regarded as mesoendemic, and the upper Simbai and Bundi areas as hypoendemic. Only in the lower Simbai area is malaria a major cause of febrile illness. However, in all areas village mean haemoglobin (Hb) levels were highly correlated with the prevalence of malaria infections, while concurrent parasitaemia reduced individual Hb levels by 1.3 g/dl (CI_{95} [1.0-1.5], p <0.001) and significantly increased the risk for moderate-to-severe anaemia (Hb <8 g/dl) (adjusted odds ratio 5.6, CI_{95} [3.6-8.6], p <0.001). Based on the survey results, areas of different malaria epidemiology are delineated and options for control in each area are discussed.

Introduction

Like neighbouring Morobe Province, Madang Province is part of the Momase administrative region of Papua New Guinea (PNG) and is a predominately coastal, lowland province. However, Madang Province also includes highland areas such as Simbai and Bundi along the northern highland fringe, and areas in the Finisterre Range. These areas are characterized by high to very high rainfall (Bundi, Kanainj, Aiome 4000-7000 mm per year, upper Simbai area 3000-3500 mm) with moderate seasonal variation and mostly high relief, as documented in the PNG Resource Information System (PNGRIS) database.

While the malaria situation in coastal Madang province has been studied in depth for over 100 years (1), there is little recent information available about malaria in the highlands and fringe areas. In 1962 a mass blood survey was conducted by the Public Health Department among the Maring people of Simbai (J. Saave personal communication, reported by Buchbinder (2)). These surveys found prevalence rates between 0% and 33% (median 9%) in different Maring villages situated between 800 and 1500 m. There was little relationship between the altitude of main villages and parasite prevalence, probably because all villages have garden areas extending well below 1000 m. Spleen rates averaged 22.5%. There was little

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difference between age groups but lower-lying villages had significantly higher spleen rates than those at higher altitude.

Similar surveys conducted in September 1967 in Bundi found an overall prevalence of 3.4% with *Plasmodium malariae* the predominant species (3). Prevalence rates were higher in infants (<1 year, 6.8%) and toddlers (1-4 years, 5.2%) than older children and adults (2.6% and 2.7%, respectively), indicating at least some degree of local transmission.

The national malaria control program in the 1960s and 1970s covered only a small coastal part of Madang Province near Madang town and, to our knowledge, none of the highland and fringe areas was ever included. Since the control days the malaria situation in coastal Madang areas has been intensively studied by the Papua New Guinea Institute of Medical Research (PNGIMR) (4,5) and major changes such as the rise of high-level chloroquine resistance in *P. falciparum* malaria (6) have been described.

The current malaria situation in the highlands and highland fringe communities, however, has not been studied in depth since the surveys cited above. In order to assess the current level of malaria transmission in these areas we therefore conducted a series of rapid malaria surveys in the Simbai-Aiome and Bundi areas in July and August 2003 (dry season) and in April and May 2004 (late wet season).

Methodology

The surveys were conducted using the sample methodology as applied in earlier highlands surveys (7-9). Within selected villages a household-based sampling strategy was used in order to achieve a sample as representative as possible of the entire village population. From each selected household, every member who could be reached during the stay in the village was included in the survey. If the village had less than 200 inhabitants, complete sampling of the village was attempted.

Demographic data were recorded, a thick and thin blood film was prepared, the spleen palpated in a lying position and axillary temperature taken from each individual of the household. Haemoglobin levels were measured using the Hemocue system (HemoCue AB, Ångelholm, Sweden). Symptomatic individuals were treated as indicated by clinical presentation. A short questionnaire on current symptoms, past malaria episodes, treatment, use of bednets and recent travel was administered to each participant or their guardian.

Giemsa-stained blood films were examined under the microscope for 100 thick-film fields under oil immersion before being declared negative. The parasite species in positive films were identified and densities recorded as the number of parasites per 200 white blood cells (WBC). Densities were converted to the number of parasites per µl of blood assuming 8000 WBC per µl. The slides were read by experienced microscopists at the PNGIMR in Goroka and Madang. A more detailed description of the survey methodology is found in Mueller et al. (7).

Data entry was done at the PNGIMR in Goroka using a double entry system. Statistical analyses were done using STATA 7.0 (Stata Corp., College Station, TX) and SPlus (Insightful Corp, Seattle, WA) statistical packages. Chi squared tests and logistic regression analyses were used for categorical variables. Continuous variables were investigated using Student's t-test, linear regression and analysis of variance (ANOVA). Haemoglobin values were adjusted for age and gender effects using regression splines.

Results

A total of 2847 people were surveyed during the 21 surveys in 13 different villages (Table 1; Figure 1). Of all participants 49% were female and 15% were aged <5 years, 12% 5-<10 years, 22% 10-20 years and 52% over 20 years.

The examination of blood slides revealed malaria infections in all Madang surveys with prevalence rates ranging from 2.5% in Arunk (dry season) to 24.7% in Kanainj (wet season) (Table 1; Figure 2). In the Simbai-Aiome area, overall prevalence was higher in the wet than the dry season (15.8% vs 10.4%, $p = 0.001$). In the Bundi villages, the opposite was found (wet: 4.1%, dry: 6.8%, $p = 0.05$).

The overall prevalence was significantly negatively correlated with altitude ($r = –0.68$,

TABLE 1

SUMMARY OF RESULTS OF PARASITOLOGY SURVEYS IN MADANG HIGHLANDS FRINGE VILLAGES

Village	N	Temperature >37.5ºC (%)	Fever in last 3 days (%)	PR (%)	Parasite species Pf/Pv/Pm/Po	SR (%)	Mean Hb (g/dl)	
							Male	Female
Dry season (July-August 2003)								
Koki	193	0.5	6.7	4.2	1/7/0/0	-	12.5	12.3
Arunk	119	0.8	8.3	2.5	3/0/0/0	-	12.2	12.1
Kanainj	224	3.6	13.8	16.5	18/21/0/0	-	11.0	10.7
Wara Asai	35	0.0	5.7	8.6	1/2/0/0	-	11.6	10.2
Iporats	167	4.8	6.6	9.0	8/7/0/0	-	10.9	10.5
Aiome CS	160	1.9	4.3	16.9	22/4/1/0	-	11.5	10.5
Mendikara –								
Sno Pas	66	1.5	12.1	6.1	3/1/1/0	-	13.6	13.4
Bundikara	104	0.0	15.4	5.8	4/2/0/0	-	12.9	12.7
Bundi Station	199	2.0	16.1	8.5	4/13/1/0	-	12.7	12.0
Karisokara	120	1.7	13.3	5.0	3/3/0/0	-	13.6	12.7
Wet season (April-May 2004)								
Koki	130	2.3	9.2	3.9	1/1/3/0	0.0	13.0	12.4
Arunk	137	7.3	13.4	2.9	4/1/0/0	2.9	12.4	11.8
Kanainj	227	8.3	14.0	24.7	34/14/10/1	28.4	10.9	10.7
Mombasap	103	2.9	6.8	14.6	9/2/4/0	55.3	12.0	11.5
Apanum	146	4.0	6.6	19.7	15/11/9/0	54.0	11.3	10.2
Iporats	124	3.2	16.0	22.6	10/8/14/0	12.8	11.1	10.4
Karisokara	107	0.0	1.0	3.7	4/0/0/0	3.7	13.7	13.1
Mendikara –								
Sno Pas	74	1.4	4.1	2.7	0/1/1/0	4.1	14.0	13.5
Bundikara	151	4.6	9.9	6.0	7/2/0/0	5.9	13.9	12.7
Bundi Station	122	2.5	4.9	2.5	2/1/0/0	17.2	12.7	12.0
Yandra	139	3.6	6.4	4.3	2/4/0/0	2.0	13.5	13.3

N = number surveyed
PR = prevalence rate (of malaria parasitaemia)
Pf = *Plasmodium falciparum*; Pv = *Plasmodium vivax*; Pm = *Plasmodium malariae*; Po = *Plasmodium ovale*
SR = spleen rate
Hb = haemoglobin level
CS = Community School

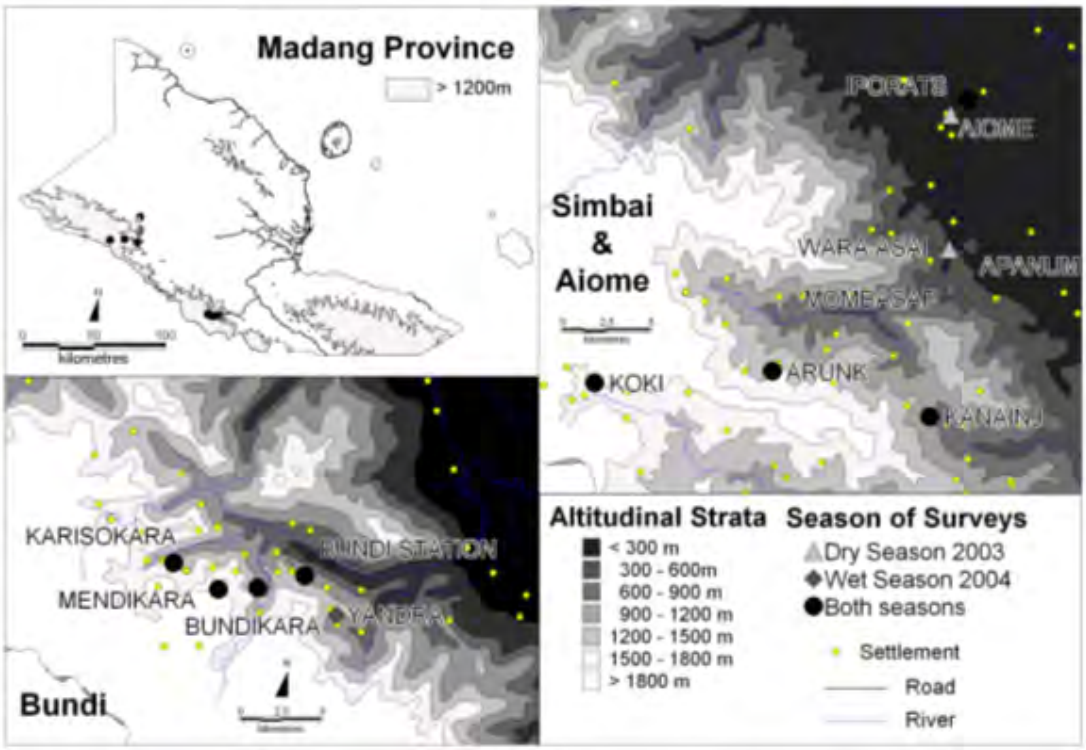

Figure 1. Locations and season of malaria surveys in Bundi and Simbai-Aiome, Madang Province.

n = 21, p <0.001). In Simbai villages malaria was rare in villages above 1400 m (3.5%), but common at lower altitudes (17.7%, p <0.001). There were, however, no significant differences in prevalence rates among Simbai-Aiome villages below 1200 m (χ^2 = 7.5, df = 5, p = 0.18) or all Bundi villages (χ^2 = 1.7, df = 4, p = 0.80) despite large differences in altitude.

Infections were strongly age dependent (χ^2 = 51.7, df = 4, p <0.001). In Simbai-Aiome infections were most prevalent in children aged 5-9 years (21.6%) and 2-4 years (18.8%), followed by adolescents (10-19 years, 18.0%), adults (7.4%), and infants and toddlers (<2 years, 5.3%). In Bundi peak prevalence was observed in the adolescent age group (8.9%), followed by children 2-4 years (5.9%), 5-9 years (4.8%), adults (4.6%) and infants and toddlers (1.8%). In the villages of Mendikara – Sno Pas, Karisokara and Yandra, no infections were seen in children <5 years of age, indicating very low or no local transmission.

All four human malaria species were found during the Simbai-Aiome surveys. The dominant species was *P. falciparum,* which accounted for 51% of infections, while 32% were *P. vivax* and 17% *P. malariae*. *P. ovale* was found only in the wet season survey in Kanainj. 6% of positive blood slides showed mixed infections. In the Bundi surveys similar numbers of *P. falciparum* (51%) and *P. vivax* (47%) were found with *P. malariae* infection being rare (2%).

Most infections were of low density: 49.8% of infections were sparse (<500/µl), 11.5% light (500-999/µl), 31.7% moderate (1000-9999/µl) and only 7.0% were heavy (≥10,000/µl). Infections with *P. falciparum* had significantly higher densities than those with *P. vivax* and *P. malariae* (geometric mean 850 vs 396 and 341/µl, respectively, p = 0.001 and 0.007).

Mixed infection had higher densities than single infections (1544/µl vs 576, p = 0.05). There were highly significant differences in intensity of infection among age groups ($F_{4,282}$ = 8.4, p <0.001). Infections were heaviest for children aged 2-4 years and infants and

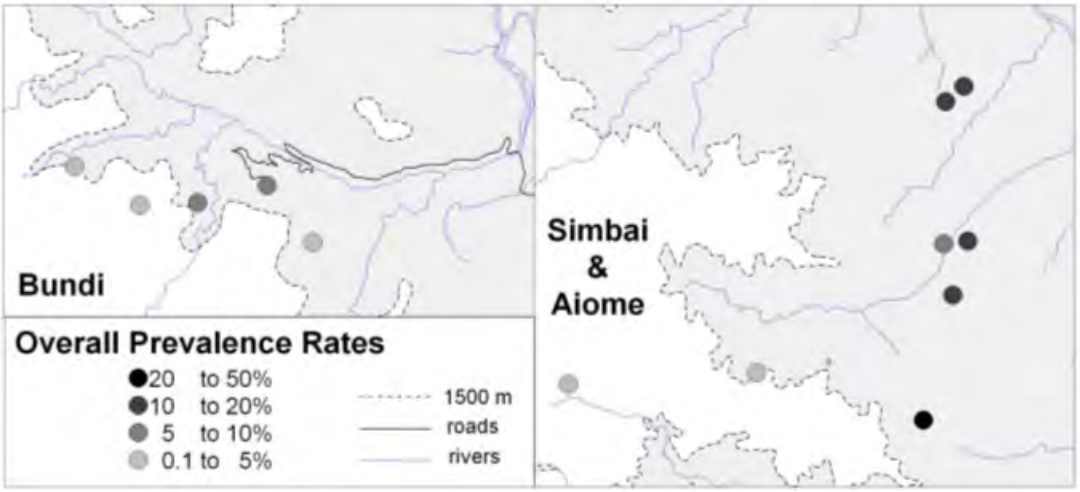

Figure 2. Overall prevalence rates of malaria infections in Bundi and Simbai-Aiome surveys.

toddlers (1893 and 1434/µl, respectively), and lightest in adolescents and adults (666 and 275/µl, respectively). After adjusting for difference in age there were no significant differences in the density of infections between Bundi and Simbai-Aiome ($p = 0.18$). However, parasite densities in the dry season were significantly greater (1002 vs 407/µl, $F_{1,252} = 12.3$, $p < 0.001$).

Spleen rates were assessed only in the wet season surveys. In the different villages they ranged from 0% in Koki to 55% in Momesap (Table 1). The rate of enlarged spleen was highly correlated with the parasite prevalence rate overall ($r = 0.64$, $n = 11$, $p = 0.03$). Enlarged spleens were most common in the 2-5 year olds (25.3%), while infants (10.3%) and adults (14.3%) had the lowest spleen rates. The average size of an enlarged spleen was 1.9 (Hackett's grade). A large spleen was highly significantly associated with a concurrent infection (27.3% vs 8.2%, $p < 0.001$). There was no difference in this association among the different *Plasmodium* species.

The overall prevalence rates of measured but not reported fever (Table 1) were significantly correlated with the prevalence of malaria parasites in a population ($r = 0.44$, $n = 21$, $p = 0.046$ and $r = 0.29$, $p = 0.20$, respectively). Among the parasite-positive cases only 8.4% had a temperature >37.5°C at the time of the survey; another 9.8% reported fever in the last 3 days. There was no difference in the prevalence of simple malarial morbidity among slide-positive people between season or area. However, the occurrence of febrile symptoms was strongly affected by the intensity of infection ($\chi^2 = 24.0$, $df = 4$, $p < 0.001$) with 50% of cases with a parasite density ≥10,000/µl reporting being febrile, but only 9% and 23% of those with densities <500/µl and 500-9999/µl. Parasite-positive cases accounted for 32.3% of all fevers at the time of survey and 25.7% of reported fever in the last 3 days overall in Simbai-Aiome, but only for 13.0% ($p = 0.08$) and 7.6% ($p < 0.001$), respectively, in Bundi. There was, however, no difference between seasons.

Mean haemoglobin levels in a village (Table 1) were highly negatively correlated with the prevalence of malarial infections ($r = -0.75$, $n = 21$, $p < 0.001$) and ranged from 13.8 g/dl in Mendikara – Sno Pas (wet, prevalence rate 2.7%) to 10.6 g/dl in Iporats (dry, prevalence rate 9.0%). The village mean haemoglobin level was found to decrease by 1.1 g/dl per 10% increase in parasite prevalence rate. Haemoglobin values were significantly lower for women than men (11.7 vs 12.3 g/dl, $t = 7.7$, $p < 0.001$) and there was a highly significant increase of haemoglobin with increasing age (test for trend: $z = 21.6$, $p < 0.01$). All subsequent analyses of haemoglobin levels and parasitaemia were therefore done on age- and sex-adjusted values.

Concurrent plasmodial infection was associated with a significant decrease in

haemoglobin (Hb) of 1.3 g/dl (CI_{95} [1.0-1.5], p <0.001). Infections with *P. falciparum* were associated with a larger drop in Hb than with *P. vivax* (–1.5 vs –0.8 g/dl, p <0.001). The (adjusted) decrease in haemoglobin was strongly dependent on levels of parasitaemia (<500/µl: –0.5, 500-999/µl: –1.5, 1000-9999/µl: –1.9, ≥10,000/µl: –2.1 g/dl, $F_{3,251}$ = 9.2, p <0.001). In Bundi, but not in Simbai-Aiome, haemoglobin levels were lower (–0.3 g/dl, p <0.001) in the dry than in the wet season.

The prevalence of moderate-to-severe malarial anaemia (SMA) (SMA, Hb <8 g/dl) was significantly correlated with overall parasite prevalence (r = 0.61, n = 21, p = 0.004). Within a survey, concurrent parasitaemia was a highly significant risk factor for SMA – adjusted odds ratio (AOR) 5.6 (CI_{95} [3.6-8.6], p <0.001). The risk of SMA increased significantly with increasing intensity of infection (<500/µl: AOR = 2.1, 500-999/µl: AOR = 9.7, 1000-9999/µl: AOR = 11.2, ≥10,000/µl: AOR = 10.1; likelihood ratio (LR)-test: χ^2 = 19.0, df = 3, p <0.001).

Between 3% (Koki, dry season) and 32% (Bundi, wet season) of people reported having had a 'malaria' episode in the two weeks before the survey (Table 2). Significantly more people in Bundi reported a recent 'malaria' episode than in the Simbai and Aiome villages (18.5% vs 14.0%, p = 0.001). The number of reported malaria episodes was highly correlated with the prevalence of infection found in a village in the Simbai-Aiome but not in the Bundi surveys (r = 0.73, n = 12, p = 0.007 and r = 0.22, n = 9, p = 0.56). Of all people reporting a malaria episode only 61% went to an aid post/health centre/hospital for treatment. There was no difference in treatment seeking between seasons, but people in Bundi were more likely to go for treatment than those in Simbai-Aiome (66.7% vs 56.1%, p = 0.02). The majority of people visiting a health centre with a complaint of 'malaria' were treated with antimalarials (72.5% of cases with available drug information). There was no significant difference in the prevalence of concurrent parasitaemia in people reporting a recent 'malaria' episode, but the risk of infection was considerably lower in people that reported recent treatment (adjusted OR 0.56, CI_{95} [0.31-1.02], p = 0.059).

With the exception of the villages near Aiome, the number of people in each village sleeping under a bednet was low (overall 23.6%, range 0-89.5%) (Table 2). Within villages, those people in a village who did use bednets had the same likelihood of infection with malaria (adjusted OR = 1.0, CI_{95} [0.6-1.5]) as those who did not.

People in Bundi used garden houses (away from their main villages) significantly more often than those in Simbai-Aiome (15.3% vs 8.3%, p <0.001). The proportion of people reporting regularly sleeping in garden houses was not significantly associated with parasite prevalence in a village (r = –0.31, n = 21, p = 0.17). In addition, among people in a village sleeping in a garden house was not associated with a significant change in the risk of malarial infection (adjusted OR = 0.68, CI_{95} [0.41-1.12], p = 0.13).

Discussion

The present surveys confirm the low endemicity of local malaria transmission in Bundi reported in the 1967 survey (3). Although there was little association of prevalence rates with altitude, infections in young children were only found at Bundi Station and Bundikara, which are at lower altitudes. In addition, the relatively high age at peak prevalence indicates that mobile age groups such as adolescents are at highest risk of infection, while local transmission is too low for the acquisition of effective immunity to malaria. Together, all this indicates that there is little or no local transmission in villages above 1600 m and most infections there are acquired when travelling either to Bundi Station or to the highly malarious Ramu Valley or Madang areas.

Current overall prevalence rates are slightly higher than those observed in 1967 (5.3% vs. 3.4%) (3). However, there has been a major shift in age distribution of malaria cases from infants and toddlers to older age groups. The difference in age distribution is probably linked to the fact that in 1967 there was no road link to Bundi and thus there was much less movement between Bundi and the Ramu and Madang areas. Infections in the 1967 surveys were thus probably mostly locally acquired or the result of chronic infections resulting from occasional trips to the Ramu Valley (thus explaining the high proportion of *P. malariae,* which can lead to long-lasting, chronic infections (10), in the 1967 surveys).

TABLE 2

SUMMARY TABLE WITH VILLAGE CHARACTERISTICS AND MALARIA-RELATED BEHAVIOUR IN MADANG HIGHLANDS FRINGE VILLAGES

Village	N	LLG	Altitude	Malaria 'sickness' (%)	Antimalarial use (%)	Bednet use (%)	Slept in garden house (%)
Dry season (July-August 2003)							
Koki	193	Simbai	1780	3.1	1.5	3.6	16.5
Arunk	119	Simbai	1480	8.3	2.5	1.7	4.2
Kanainj	224	Usino	1120	19.6	8.5	4.0	11.1
Wara Asai	35	Arabaka	300	17.1	8.6	55.9	20.6
Iporats	167	Arabaka	90	9.1	7.3	80.0	9.0
Aiome CS	160	Arabaka	80	6.3	3.8	81.9	7.5
Mendikara – Sno Pas	66	Bundi	2120	6.1	1.5	0.0	40.9
Bundikara	104	Bundi	1500	20.4	13.6	1.0	23.3
Bundi Station	199	Bundi	1420	27.1	23.1	42.6	12.1
Karisokara	120	Bundi	1680	10.0	7.5	9.2	15.0
Wet season (April-May 2004)							
Koki	130	Simbai	1780	6.9	3.1	0.0	3.1
Arunk	137	Simbai	1480	12.3	10.9	0.0	6.6
Kanainj	227	Usino	1140	24.5	14.4	2.2	4.5
Mombasap	103	Simbai	620	11.7	4.9	12.0	5.9
Apanum	146	Arabaka	240	15.8	5.9	73.8	14.1
Iporats	124	Arabaka	80	31.2	21.6	89.5	0.0
Karisokara	107	Bundi	1680	7.3	16.5	4.6	17.4
Mendikara – Sno Pas	74	Bundi	2120	9.5	9.5	0.0	33.3
Bundikara	151	Bundi	1500	26.3	15.8	1.3	10.6
Bundi Station	122	Bundi	1420	31.5	33.6	27.3	5.0
Yandra	139	Bundi	1200	11.7	15.9	0.7	5.4

N = number
LLG = Local Level Government
CS = Community School

The prevalence rates in the Bundi area are similar to those observed along the southern fringe in Eastern Highlands Province (Simbari, Marawaka) (8), but lower than those found at similar altitude in other highland and fringe areas (9). The low endemicity in Bundi may be related to the high rainfall and steep terrain, which may lead to regular flushing of potential breeding sites thereby impeding efficient mosquito breeding.

In the Simbai area there is also little malaria transmission in villages above 1400 m. Below 1400 m, however, there is moderate to high transmission with surprisingly little difference in prevalence rates between survey villages, despite large differences in altitude. The prevalence rates in the Simbai villages (ie, Koki, Arunk and Kanainj) are similar to those found (at similar altitude) in the neighbouring Maring area in 1964 (2), indicating that transmission intensity may not have changed greatly over the last 40 years.

The lack of a clear association of prevalence with altitude in villages below 1400 m may be linked to a series of factors. On the one hand, the high coverage of insecticide-treated bednets (ITNs) in the Aiome area, following a recent distribution program, may have significantly reduced transmission in these villages. In African trials, high ITN coverage resulted in a reduction of 50% in the incidence of symptomatic malaria and 13% in prevalence (11). Wara Asai and Mombasap, on the other hand, are small isolated communities and such communities tend to have lower prevalence rates than large, less isolated ones. Last but not least, the altitude for Kanainj main village might only very loosely correspond to the altitudinal range of gardens and hunting areas. In the neighbouring Maring area, garden and hunting areas regularly extend down to 300-600 m altitude (2).

As spleen rates are highly correlated with the overall prevalence of infections in endemic situations, but not during epidemics, and also vary less between seasons, they are generally considered to be a robust measure of malarial endemicity (12). In the current surveys, spleen rates were highly correlated with parasite prevalence rates at the population level, and with concurrent infections at the individual level. Spleen rates in children aged <5 years in the Simbai villages were similar to those reported in the 1964 Maring surveys (2) (28.6% vs 24%), again indicating that malaria transmission has not changed substantially in 40 years in the Simbai area. Based on spleen rates the lower Simbai area would be regarded as mesoendemic, and the upper Simbai and Bundi areas as hypoendemic.

The patterns of simple malaria morbidity (ie, fever + presence of parasites) reveal two interesting facts. Firstly, at low transmission intensities (ie, the upper Simbai and Bundi villages) very few observed and reported fevers are linked to malarial infections. However, even in these areas almost every episode of fever is treated with antimalarials (see below). At higher transmission levels (Kanainj and Aiome villages) malaria is a major cause of febrile illness. It is interesting to note that even in higher villages many infections are of low density and asymptomatic. As has been observed in highly endemic areas (13) the presence of febrile symptoms is strongly dependent on parasite density and only with densities exceeding 10,000 parasites/µl were a majority of infections symptomatic.

Anaemia is a more chronic complication of malarial infections. Recent studies have found that haemoglobin levels in African children were more tightly linked to the average parasitaemia over the preceding 3 months than with concurrent infection (14). It is therefore not surprising that, as in other highlands populations (7-9), in Bundi and Simbai villages mean haemoglobin levels are strongly linked with the prevalence rate, while concurrent parasitaemia reduced Hb levels by 1.3 g/dl. The sharp decrease in individual haemoglobin and risk of severe malarial anaemia with increasing density of infection are further indications of the importance of malaria in the aetiology of anaemia in these populations.

Although Bundi had lower prevalence rates and a significantly lower proportion of measured or reported fevers with concurrent malarial infections, a higher proportion of people reported having recently suffered from ‘malaria’. In addition, a higher number of ‘malaria’ sufferers reported going for treatment. This indicates that as elsewhere in the PNG highlands (7-9) in people’s perception the term ‘malaria’ is synonymous with any febrile illness. At rural health centres, where microscopy is lacking, most

cases of 'malaria'-type febrile illness are treated routinely with antimalarials, and over-treatment with antimalarials is a serious problem that leads both to higher drug costs and the risk of increasing drug resistance.

Despite the lack of association of reported bednet use with concurrent parasitaemia, the fact that the Aiome villages, where an ITN distribution program had occurred just before the dry season surveys, had similar prevalence rates as the much higher-lying village of Kanainj might well be due to the difference in bednet use.

Conclusions and Recommendations

Although other highland areas in the province were not included in the present surveys, we believe that recommendations derived from the present surveys may also be applied to higher-altitude settlements in the Finisterre Range. Using data from the present and past surveys in coastal Madang we can thus define 3 malaria-endemic areas with different risks of malaria in Madang Province (Figures 3 and 4).

Moderate to highly endemic (<1200 m)

The great majority of villages (87%) in Madang Province are situated in areas with moderate to highly endemic malaria (prevalence 10-30+%). In these areas spleen rates are high and malaria is a major cause of febrile illness, haemoglobin levels are comparatively low and severe anaemia is common. All areas situated below 1200 m fall within this category (Figure 4).

Distribution of insecticide-treated mosquito nets (ITNs) is the preferred control measure for these areas. In areas of high malarial transmission in some African countries ITNs have been shown to reduce malarial morbidity by 50% (11). However, in order to be effective a high coverage (>80%) needs to be achieved and regular re-treatment with insecticide is required. Given the remoteness of many parts of the province, this seems too difficult to achieve. Consequently, long-lasting insecticide-treated nets (LLINs) that

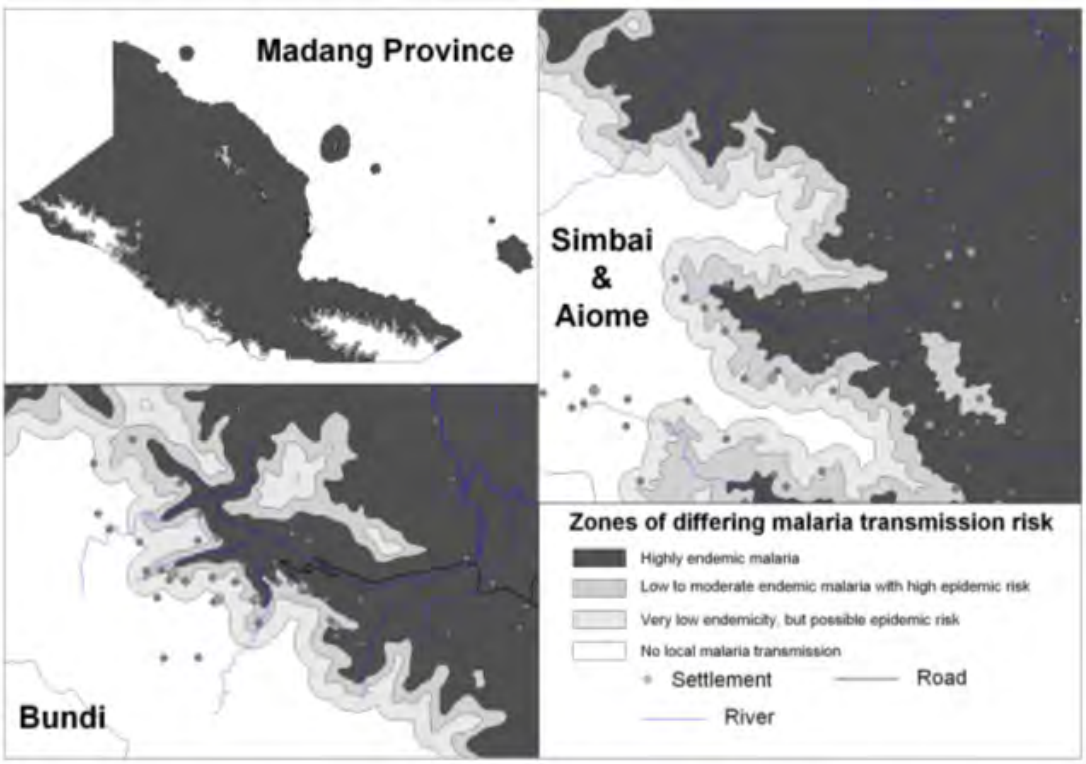

Figure 3. Zones of different malaria risk in Bundi and Simbai-Aiome areas, Madang Province.

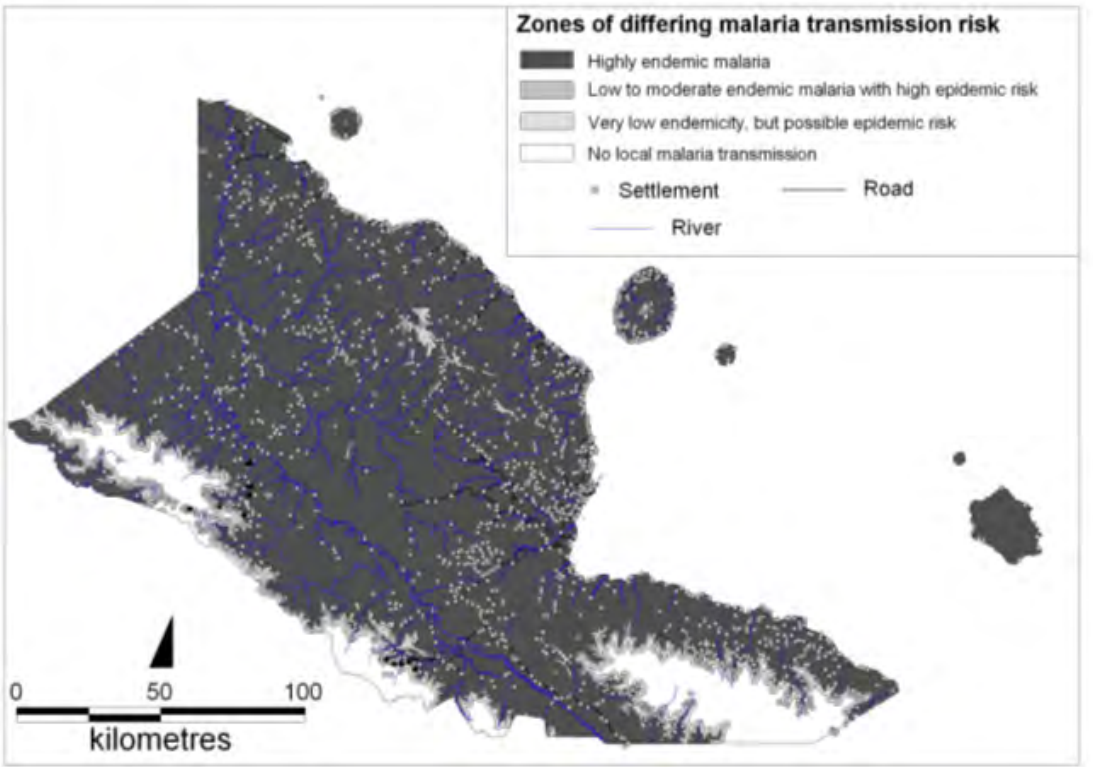

Figure 4. Zones of different malaria risk in Madang Province.

do not need re-treatment are preferable.

Low transmission potential with potential risk of epidemic outbreaks (1200-1600 m)

At intermediate altitudes (1200-1600 m), transmission is generally low (PR <5-10%). Although no epidemic outbreaks were detected in the present surveys, based on knowledge from other highland areas (7-9) it is likely that these areas have the potential for epidemic outbreaks. Importation of malaria episodes acquired by travelling to neighbouring highly endemic areas or to lower-lying garden and hunting grounds may be an important source of malaria infection and illness in these areas. Outside possible epidemics malaria is not likely to be a major source of febrile illness. 5.6% of Madang villages are situated in this area. An additional 3.1% of villages are at altitudes of 1600-1700 m and, depending on local circumstances, may also fall in this risk category.

The remoteness of all areas at that altitude rules out the use of indoor residual spraying and thus the distribution of long-lasting insecticide-impregnated bednets may be the preferred method of control. In addition, people should be educated about the risk of acquiring malaria infections when travelling to the lowlands and local health centres should be strengthened to efficiently diagnose and treat both locally transmitted and imported malaria cases.

Very low transmission or non-transmission areas (above 1600-1700 m)

Very few villages in Madang (2.6%) are situated at altitudes that preclude stable local malaria transmission. In these areas prevalence rates are low (<5%) and most malaria cases are likely to result from travel to lower-altitude malarious areas. Consequently, malaria is only a very minor source of febrile illness and most reported 'malaria' fevers are non-malarial in origin.

Malaria control in these areas should be based on the prompt treatment of imported cases, rather than vector control. While control measures in neighbouring lower-lying

areas should prevent possible malaria epidemics from spreading into higher areas, epidemic surveillance and, if necessary, control (using indoor residual spraying and mass drug administration) should nevertheless be part of malaria control plans for these areas.

REFERENCES

1 **Müller I, Bockarie M, Alpers M, Smith T.** The epidemiology of malaria in Papua New Guinea. *Trends Parasitol* 2003;19:253-259.

2 **Buchbinder G.** Maring microadaptation: a study of demographic, nutritional, genetic and phenotypic variation in a Highland New Guinea population. PhD Thesis, Columbia University, New York, 1973.

3 **Malcolm LA.** Growth and Development in New Guinea - a Study of the Bundi People of the Madang District. Institute of Human Biology (now Papua New Guinea Institute of Medical Research) Monograph No 1. Goroka: Institute of Human Biology, 1970:105p.

4 **Cattani JA, Moir JS, Gibson FD, Ginny M, Paino J, Davidson W, Alpers MP.** Small-area variations in the epidemiology of malaria in Madang Province. *PNG Med J* 1986;29:11-17.

5 **Cattani JA, Tulloch JL, Vrbova H, Jolley D, Gibson FD, Moir JS, Heywood PF, Alpers MP, Stevenson A, Clancy R.** The epidemiology of malaria in a population surrounding Madang, Papua New Guinea. *Am J Trop Med Hyg* 1986;35:3-15.

6 **Al-Yaman F, Genton B, Mokela D, Narara A, Raiko A, Alpers MP.** Resistance of *Plasmodium falciparum* malaria to amodiaquine, chloroquine and quinine in the Madang Province of Papua New Guinea, 1990-1993. *PNG Med J* 1996;39:16-22.

7 **Mueller I, Taime J, Ivivi R, Yala S, Bjorge S, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 1. Western Highlands Province. *PNG Med J* 2003;46:16-31.

8 **Mueller I, Bjorge S, Poigeno G, Kundi J, Tandrapah T, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 2. Eastern Highlands Province. *PNG Med J* 2003;46:166-179.

9 **Mueller I, Kundi J, Bjorge S, Namuigi P, Saleu G, Riley ID, Reeder JC.** The epidemiology of malaria in the Papua New Guinea highlands: 3. Simbu Province. *PNG Med J* 2004;47:159-173.

10 **Gilles HM, Warrell D.** Bruce-Chwatt's Essential Malariology, 4th edition. London: Edward Arnold, 2002.

11 **Lengeler C.** Insecticide-treated bed nets and curtains for preventing malaria. *Cochrane Database Syst Rev* 2004;2:D000363.

12 **Metselaar D, Van Thiel PH.** Classification of malaria. *Trop Geogr Med* 1959;11:157-161.

13 **Smith T, Genton B, Baea K, Gibson N, Taime J, Narara A, Al-Yaman F, Beck HP, Hii J, Alpers M.** Relationships between *Plasmodium falciparum* infection and morbidity in a highly endemic area. *Parasitology* 1994;109:539-549.

14 **McElroy PD, ter Kuile FO, Lal AA, Bloland PB, Hawley WA, Oloo AJ, Monto AS, Meshnick SR, Nahlen BL.** Effect of *Plasmodium falciparum* parasitemia density on hemoglobin concentrations among full-term, normal birth weight children in western Kenya, IV. The Asembo Bay Cohort Project. *Am J Trop Med Hyg* 2000;62:504-512.

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Chlamydia trachomatis infection and distribution of serovars in the Eastern Highlands Province, Papua New Guinea

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SUMMARY

We have used nested polymerase chain reaction (PCR) and the PCR-based endonuclease digestion method to genotype *Chlamydia trachomatis* serovars in 460 infected individuals from the Eastern Highlands Province of Papua New Guinea. Our study groups comprised women who presented in labour to the Goroka Base Hospital, their newborn infants, symptomatic children who presented to the hospital's Outpatients Department and men and women from 15 randomly selected villages in the Asaro Valley. In this analysis, the major outer membrane protein (MOMP) gene, *omp*1, of *C. trachomatis* was amplified using DNA obtained from the endocervix of women, urine from men, and both the eye and nasopharynx of children. Amplified DNAs were digested concurrently using *Alu*I and a combination of *Eco*RI, *Hinf*I and *Hpa*II restriction enzymes. The mixtures were separated on electrophoretic gels and the respective serovars designated on the basis of resolved digested DNA patterns. Our results, which were confirmed also by *omp*1 sequence data, show serovars D, E, F, G, H and L3 to be present in the studied communities. The overall relative frequencies of these serovars were 30%, 21%, 25%, 1%, 20% and 2% respectively, with serovars D, E, F and H accounting for 97% of these infections. Double infections among these principal serovars were also detected in all our study groups but at a low overall frequency of 3%. Serovar D was the major agent involved in the aetiology of chlamydial infection in both children and adults though serovar F was the most frequent in newborn infants. Serovar H was relatively less frequent in symptomatic children. No trachoma-related serovars were detected, confirming the rarity of this disease in Papua New Guinea. In contrast, although clinical cases of lymphogranuloma venereum have not been described in the country, the detection of serovar L3 in this study suggests that it may occur. However, the association

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of L3 also with childhood infection indicates that it may be causing the same pathology as the serovars D-K that are associated with non-ulcerative sexually transmitted infections.

Introduction

Chlamydia trachomatis is a major cause of the severe morbidity resulting from sexually transmitted infections (STIs) worldwide (1). It has been recognized as the leading cause of bacterial STIs in the United States (2) and other developed countries (3,4). The prevalence and impact on disease burden attributed to this pathogen is expected to be greater in resource-poor settings, such as among ethnic minorities (5,6) and developing countries (7), given their high background of STIs.

This pathogen presents a spectrum of clinical disease that is similar to that caused by gonococci in infected hosts. However, unlike the more acute disease from the more highly transmissible *Neisseria gonorrhoeae*, transmission of *C. trachomatis* occurs at an observably low rate (8) and symptoms when present are less apparent. Ironically, due to its reported chronicity and fulminating character (5,9,10), progressive infections by this organism in both the ocular and genitourinary tract can result in life-threatening conditions. This appears to be immunologically mediated. The immunopathological responses elicited against the chlamydia heat-shock protein (11,12) and other antigens, in particular the major outer membrane protein (MOMP) (12-15), cause scarring and pathological changes in the superficial epithelia and columnar cells leading to disease. In infected women, the subsequent tissue destruction, fibrosis and scarring of the columnar endothelial linings in the upper genitourinary tract can result in chronic abdominal pain, pelvic inflammatory disease (PID) and ectopic pregnancies (16). Both female and male adults may become sterile following occlusion of the reproductive tract (12) and children born to infected women can develop neonatal conjunctivitis and pneumonia (17), with the latter group being predisposed to pulmonary complications later in life (18). The potential of this happening is said to increase over time with persisting and recurrent infections (12,15). Moreover, recurrent infections can also result in reactive arthritis and, in pregnant women, can lead to premature rupture of the amniotic membranes and preterm births (19,20).

Disease-causing strains of *C. trachomatis* have been broadly categorized into 18 major serological variants or serovars, now determined by genotyping of the major outer membrane protein gene. These include serovars A, B, Ba, C, D, Da, E, F, G, H, I, Ia, J, K, L1, L2, L2a and L3. Traditionally, serovars A to C have been associated with trachoma, serovars D to K with non-ulcerative STIs and serovars L1 to L3 with lymphogranuloma venereum (LGV). Whether or not pathogenicity varies between serovars is not clearly established because it is usually difficult to grow this organism for such analysis. However, what is known is that both T-cell and humoral responses elicited against chlamydial infection appear to be serovar specific (21), and therefore not broadly protective. This suggests that reinfection with a different serovar may enhance hypersensitive immunity thereby contributing to progressive disease.

Preferential serovar infection in certain hosts and diseases (22) and association of some serovars with specific clinical manifestations (23), or their influence in confounding clinical diagnosis in heterogeneous STI infections (24), further support the concept of serovar-specific disease associations. New insights into this field show, however, that this is not always the case. The detection of trachoma-related variants, serovars B and Ba, in the genitourinary tract (25-27) is a case in point. Indeed, both serological relatedness studies and recent nucleotide and deduced amino acid analyses (28) define these serovars as belonging to the B serocomplex, which also includes serovars D, E, L1 and L2. The other agent of LGV, serovar L3, is found to be more related to the C serocomplex group which comprises serovars A, C, H, I, J and K, with serovars F and G making up the third serogroup. Regional variation is another factor to be considered and apparent disease associations with some serovars may really be a consequence of their geographic

distributions.

Studies to date have shown *C. trachomatis* to be highly prevalent in Papua New Guinea (PNG) (29-33). However, no data have been available about which serovars are prevalent in the country. The identification of *C. trachomatis* serovars constitutes an essential step towards greater understanding of the epidemiology of chlamydial infections. While the detection of chlamydial STIs implies that the relevant serovars are present, no epidemiological or clinical data are available on trachoma-related serovars in the country. Both trachoma and LGV have been assumed to be rare, or even non-existent, but this has not been rigorously documented. As a first step in determining the distribution of *C. trachomatis* serovars in PNG, we have tested previously collected samples in the Eastern Highlands Province (EHP) to establish the serovars found in the study population groups there.

Materials and Methods

Study populations

Samples that were determined previously to be positive for *C. trachomatis* by direct fluorescent antibody test (DFA) (Behring Diagnostics) and polymerase chain reaction (PCR) were used. The samples represented a cross-section of population groups studied in 1991 and between June 1994 and March 1998. They comprised first-void urine from men and endocervical swabs from women living in 15 randomly selected villages of the Asaro Valley in EHP (30, 31), endocervical swabs from women who presented in their first stage of labour for delivery at the Goroka Base Hospital (GBH) and both eye swabs and nasopharyngeal aspirates from the newborn infants of these women (32). Eye swabs collected previously from children who presented with eye discharge to the Outpatients Department of GBH (33) were also tested. Ethical approval for these studies was obtained from the Medical Research Advisory Committee of Papua New Guinea.

Polymerase chain reaction detection

A 1200 base-pair (bp) gene segment, *omp*1, spanning almost the entire region of *C. trachomatis* MOMP, was amplified from crudely extracted DNA using forward primer FLS (5'-CTC TTG AAA TCG GTA TTA GTA TTT GCC GCT-3') and reverse primer FLA (5'-TTA GAA GCG GAA TTG TGC ATT TAC GTG AGC-3') (34). PCR was done in a volume of 50 µl using 5 µl of the crude DNA lysate, 50 mM KCl, 50 mM Tris (pH 9.0), 1.5 mM $MgCl_2$, 0.1% Triton X-100, 0.2 mM dATP, dCTP, dGTP and dTTP, 0.5 µM of each primer and 0.04 units of Taq DNA polymerase. Samples were subjected to 30 cycles of denaturation at 94°C for 1 minute, annealing at 60°C for 1 minute and extension at 72°C for 1 minute, with final extension at 72°C for 7 minutes. A 1 µl aliquot of this primary PCR product was reamplified using species-specific internal primers CT7 (5'-TGA CTT TGT TTT CGA CCG TGT TTT-3') and CT6 (5'-TTT TCT AGA TTT CAT CTT GTT CAA T/(C)TG-3') as previously described by Gaydos and co-workers (35). Amplified products were resolved by electrophoresis on 1% agarose gel and stained with 0.01 µl/ml ethidium bromide solution for confirmation.

Restriction endonuclease digestion

Ten microlitre aliquots of CT7/CT6 PCR products were digested concurrently with *Alu*I and a combination of *Eco*RI, *Hinf*I and *Hpa*II (Promega) restriction endonucleases. Samples were incubated at 37°C overnight for 16 hours in appropriate restriction buffers. 8 microlitres of the digested DNA mixtures were resolved separately on 12% polyacrylamide gels at 10.5 volts/cm for 2.5 hours at room temperature. The restricted DNA fragments were detected by silver staining.

Sequencing

The dideoxy chain termination method was performed independently to confirm the initial results of *C. trachomatis* genotypes obtained by endonuclease digestion. PCR products of primers FL8/FL3 and N2/N4 (M. Ward et al., Southampton University, UK), which span the variable segments II and IV of *omp*1 respectively, were used as templates for this analysis.

Data analysis

Data analysis was carried out using Stata 8.0 (Stata Corporation) and Epi Info 6.04b (Centers for Disease Control and Prevention, USA). Associations of serovars and their variables were analyzed using contingency tables and their statistical significance

determined using the Pearson chi-squared test. Where appropriate, the Mantel-Haenszel chi-squared test was performed to test for interaction with possible cofactors.

Results

Samples from 460 individuals were successfully tested, including 195 women who presented in labour at Goroka Base Hospital and were subsequently delivered (median age of 23 years), 113 of their newborn infants (median age of 56 days) and 121 individuals from the 15 villages in the Asaro Valley, of whom 19 were men (median age of 26.5 years) and 102 women (median age of 24 years). The majority of the women who presented to the hospital for delivery came from the Asaro Valley but not necessarily from the same villages as the community-based women recruited there. Results were also available for 31 children (median age of 13 days) who presented previously with eye discharges at the same hospital.

Serovars D, E, F, G, H and L3 were detected in the study population groups. Serovars D, E, F and H were the predominant types detected and accounted for 97% of infections overall. Their respective frequencies were 30%, 21%, 25% and 20% (Table 1). Serovars L3 and G were consistently detected at low relative frequencies in all the study groups, accounting overall for 2% and 1% respectively.

The respective distributions for the studied population groups were, however, observed to be variable (Table 1). Except among the newborn infants and the village-based men, serovar D was the most common serovar detected: 27% in the women who presented in labour, 35% in the symptomatic children and 40% in the women recruited from the villages. Serovar E was the commonest type detected in the village-based men and serovar F in the newborn infants, accounting for 42% and 39% respectively. Serovar F was also determined to be the second most common type detected in the women in labour and the village-based men, with relative frequencies of 26% and 32% respectively.

Only 8% of women in the general population were found to be infected with serovar F while the relative frequencies were higher in the infants and the mothers (39% and 26% respectively). The observed difference in the distribution of serovar F between the two groups of women was significant, even when corrected for the year of study ($\chi^2 = 13.34$, 1df, $p < 0.001$); our results showed that *C. trachomatis*-positive pregnant women were significantly more likely to be

TABLE 1

DISTRIBUTION OF *CHLAMYDIA TRACHOMATIS* SEROVARS DETECTED IN THE STUDIED POPULATION GROUPS

Serovar	Groups studied					
	Women in labour	Newborn infants	Children with eye discharge	Village-based women	Village-based men	Overall
	N=195 %	N=113 %	N=31 %	N=102 %	N=19 %	N=460 %
D	27.2	26.5	35.5	40.2	26.3	30.4
E	21.0	14.2	32.3	22.5	42.1	21.4
F	25.6	38.9	16.2	7.8	31.6	24.6
G	1.0	0.9	0.0	2.0	0.0	1.1
H	22.6	17.7	6.5	27.5	0.0	20.3
L3	2.6	1.8	9.7	0.0	0.0	2.2

infected with serovar F than women in the general population who were *C. trachomatis* positive (odds ratio = 5.0, 95% confidence interval = 1.81–14.29, p <0.001). The proportion of pregnant women infected with serovar F also increased significantly with age (by age group <20 years, 20-25 years and >25 years) (χ^2 = 11.59, 2df, p = 0.003).

Among newborn infants, of the total of 77 infections in the eye, 9 were caused by serovar H and 1 by serovar L3. Similarly, of 96 infections in the nasopharynx, 19 were caused by serovar H and 2 by serovar L3. There was no significant difference in the relative frequency of serovars with respect to samples taken from the eye and the nasopharynx in this group of newborn infants. Serovar L3 was detected at an unexpectedly higher frequency in the children who presented with eye discharges (10%), contrary to its low frequency rates observed in the other groups (Table 1).

Double serovar infections were detected in the population groups studied but at a low frequency rate of 3% overall. 4 individuals in the study groups had mixed infection with serovars DF, 3 with serovars DH, 2 with serovars EF and 1 individual each with mixed serovars DE, EH and FH.

Distributions of the predominant serovars detected in the women who were seen at the hospital during the study period are shown in Figure 1. Except in 1995 where all the four major serovars showed rates between 20% and 30%, their respective rates were markedly variable in the study period. Significant differences over the study period per serovar were noted only in the distribution of serovar E (χ^2 = 10.24, 4df, p = 0.04) and serovar F (χ^2 = 9.39, 4df, p = 0.05). All the community-based women were enrolled in 1995. The corresponding frequencies of the respective serovars in the latter group are reported in Table 1.

The restricted DNA fragments generated by the panel of restriction enzymes that were used in the study showed a typical DNA profile for each serovar (Figures 2 and 3). In all cases, serovars D, E, F and G, represented respectively in lanes 2, 3, 4 and 5 of Figure 2,

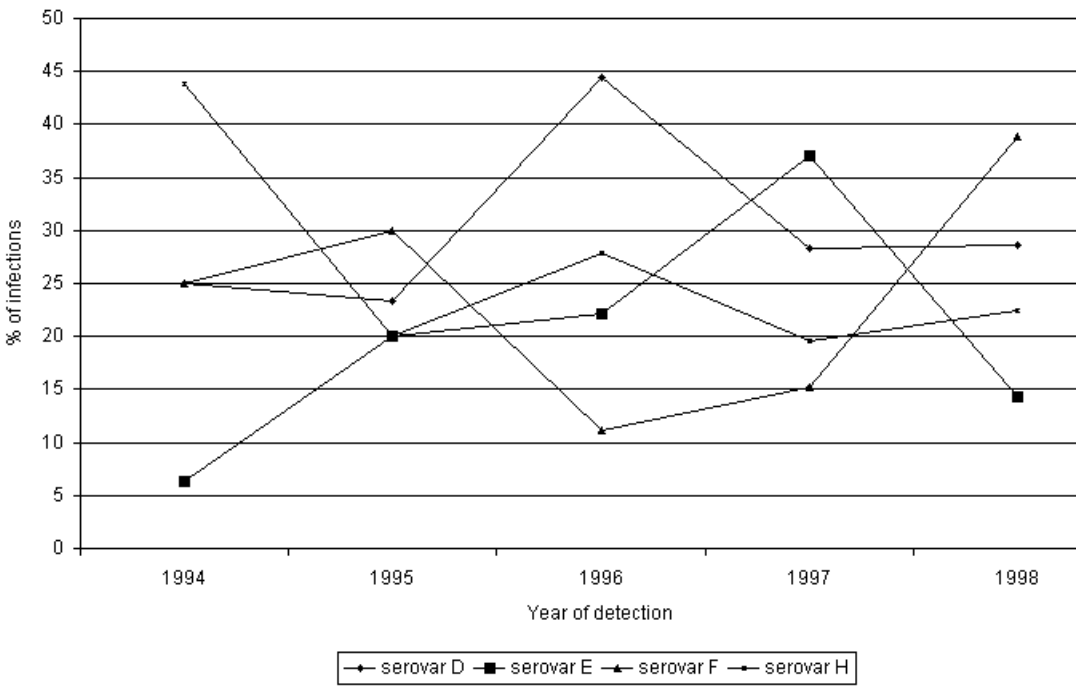

Figure 1. Distribution of predominant serovars detected in women in labour positive for *C. trachomatis* by year, 1994-1998.

were readily distinguishable by *Alu*I digestion. Serovars H and L3 (lanes 6 and 10 of Figure 2) are known to share the same restriction site for this enzyme as serovars A, C, I and J (35), and were detected separately by multiple digestions with *Eco*RI, *Hinf*I and *Hpa*II (Figure 3). Potential variants of serovars H and L3 are also shown in lanes 9 and 7 respectively of Figure 2. The multiple endonuclease restriction patterns for these variants were also noted to be distinct as depicted in Figure 3. Minor shifts in restricted DNA bands also suggested that variants of serovars D and E were present (results not shown). In all these cases, the restricted DNA fragments of multiple digestions maintained the variability of their respective prototype bands. One unresolved profile suggested an infection with more than two serovars to be present, or alternatively the presence of a new variant, but this could not be confirmed due to unsuccessful reamplification attempts.

Our RFLP (restriction fragment length polymorphism) genotype results were found to be consistent overall with the sequencing data (Table 2). No sequencing data were available for serovars G and L3 but the restricted DNA banding profile obtained for these serovars in the study appeared to be consistent with previously published patterns (35).

The PCR-based genotyping adopted in this study was found to be efficient in designating the serovars that are present in the studied population groups. However, although amplification of *C. trachomatis* with primers CT7/CT6 had been shown to be successful in the study of Gaydos et al. (35), some positive samples determined previously by *C. trachomatis* species-specific PCR with N2/N4 primers in our laboratory failed to amplify in the current study. This failure continued despite several reamplification attempts using

Figure 2. Restricted DNA patterns of *C. trachomatis* serovars using *Alu*I restriction endonuclease.
Lane 1, 1 kilobase (kb) DNA ladder; lane 2, serovar D; lane 3, serovar E; lane 4, serovar F; lane 5, serovar G; lane 6, serovar H; lane 7, variant of serovar L3; lane 8, 1 kb DNA ladder; lane 9, variant of serovar H; lane 10, serovar L3.

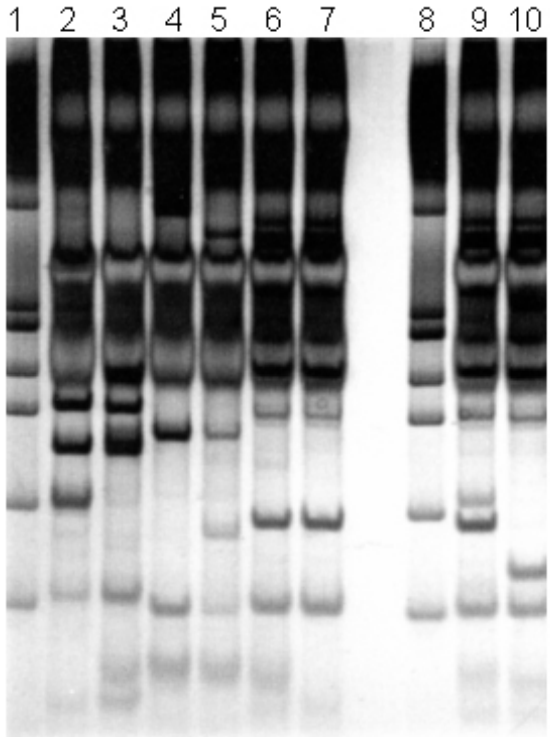

Figure 3. Restricted DNA patterns of *C. trachomatis* serovars using *Eco*RI, *Hinf*I and *Hpa*II restriction endonucleases.
Lane 1, 1 kb DNA ladder; lane 2, serovar D; lane 3, serovar E; lane 4, serovar F; lane 5, serovar G; lane 6, serovar H; lane 7, variant of serovar L3; lane 8, 1 kb DNA ladder; lane 9, variant of serovar H; lane 10, serovar L3.

phenol-chloroform-treated *C. trachomatis* DNA extracts.

Discussion

This is the first report of serovars of *C. trachomatis* identified from Papua New Guineans, including both asymptomatic and symptomatic individuals. Serovars D, E, F and H were the predominant types found among children and adults in our study communities. Serovar D was the most common type detected in the study groups, except among the newborn infants and the village men, where serovars F and E predominated (Table 1). Serovars D, E and F accounted for 76% of chlamydial infections in the studied groups, slightly more than the 60-70% reported previously among urogenital chlamydia in The Netherlands and Sweden (25,36), and also elsewhere (3,37-40). They appear to be the clinically important serovars in the study population as suggested by the 84% (26/31) carriage rate detected among the symptomatic children.

The high relative frequency of serovar H determined in our study is exceptional as much lower frequencies of this serovar have been reported in the earlier mentioned studies (3, 25, 36-40). Nevertheless, the observed frequencies in the present study are consistent with a high urogenital prevalence that has been reported twice before: among STI patients in The Netherlands and also from individuals who presented to sexually transmitted disease and other related adult consultation clinics in Lisbon (26, 41). In Lisbon, women who were over 25 years old had a significantly higher prevalence of serovar H than younger women or men. Results of the present study show that serovar H was not found in men though it was frequently detected in women and children (Table 1). It was found to colonize both the eye and the nasopharynx in children. A lower frequency noted among symptomatic children shows, however, that it might not be as

TABLE 2

CHLAMYDIA TRACHOMATIS SEROVARS DETERMINED BY RESTRICTION ENDONUCLEASE DIGESTION AND SEQUENCING ANALYSIS USING PRIMERS FL8/FL3 AND N2/N4

Sample	PCR-RFLP	VS2 (FL8/FL3)	VS4 (N2/N4)
1	D	D	D
2	E	E	E
3	F	F	F or G
4	Ha	H	H-K
5	Hb	L3	H-K
7	D+H mixed	D+H mixed	D+H-K mixed
8	F+H mixed	H	H-K

PCR-RFLP = polymerase chain reaction – restriction fragment length polymorphism

important clinically as serovars D, E and F in children. Put together, serovars D, E, F and H caused 97% of all symptomatic and asymptomatic chlamydial infections in the study population groups, including mixed infections, with serovars G and L3 accounting for the rest.

Observed differences in the relative frequencies of serovars between the women in labour and their newborn infants (Table 1) are not unexpected as the samples drawn from the respective population groups were not paired. The two distributions are not, however, significantly different ($\chi^2 = 7.01$, 5df, $p = 0.22$). An investigation of maternal-infant transmission of *C. trachomatis* is being carried out and will be reported separately. On the other hand, the difference observed for *C. trachomatis* serovar F infection between the non-pregnant community-based women in this study and those who presented for delivery at the hospital (Table 1) is noteworthy, implying host preference. Women who were pregnant and older were significantly more likely to be infected with this serovar ($p = 0.003$) than the non-pregnant women drawn from the villages, or women who were pregnant but younger. Results of earlier studies in Japan, however, suggest that there the distributions of serovars E, F and D are similar in these groups of women (27,37). The variable distribution observed for the respective serovars shows that differences in time and place, as well as age and parity, can influence serovar distribution. To resolve these questions of host and disease associations in PNG communities would require a specifically designed study.

We have previously (42) reported that, despite an apparent absence of clinical LGV in PNG communities, the presence of serovar L3 as confirmed in this study shows that the potential for this disease does exist. The epidemiology of LGV is poorly defined. Unlike infections with serovars A-K, which are largely confined to mucosal columnar epithelial surfaces of the genital tract and the eye, the so-called LGV serovars primarily infect macrophages and monocytes. They pass through the epithelial surfaces to access the regional lymph nodes from where they can also cause disseminated infections. However, since this infection cannot be distinguished clinically from other causes of genital ulcerative disease associated with bubo formation, it is quite possible that some LGV may have been misdiagnosed in the past as syphilis, chancroid, herpes or donovanosis in PNG communities. A relatively higher frequency of serovar L3 seen among children presenting with eye discharge (10%) in this study (Table 1), compared to its low genital prevalence and the apparent absence of clinical LGV, suggests that serovar L3 may be causing the same spectrum of pathology associated with non-ulcerative STI serovars D-K in this population. Although this has not been shown to occur

elsewhere, the observed clinical association in this study is consistent with the established similarity of serovar L3 to the other members of the C serocomplex group at the molecular level, namely serovars H, I, J and K (28).

Mixed serovar infections were also detected but found to be uncommon (3%) in the studied population groups. An earlier study (43) showed that mixed serovar infections can be expected in instances where a history of multiple sex partners is present. In the present study, double infections involved the common types. The combinations DE, DF, DH, EF, EH and FH were detected, with mixed serovars DF, DH and EF determined to be more common (4, 3 and 2 individuals respectively). Detection of one unresolved case with multiple atypical DNA bands suggested the possibility of an infection involving more than two serovars. Both this and the presence of new variants have been suggested to occur elsewhere (44), but appear to be rare, as we have also found. We were not able to resolve this particular issue in our study due to failed reamplification attempts.

We did not detect serovars A-C associated with trachoma in this study. This potentially eye-blinding disease has not been rigorously documented in PNG in the past, although it has been looked for clinically (45) and described in population surveys (46); furthermore, D.M. Graham (1982, unpublished data) has reported having detected antibodies to this group of serovars in the ocular samples she obtained. Established genetic and antigenic similarities between serovar Ba and serovars D-K (47) and its association with genital infection implicate it as the most likely causative agent. In view of these observations, a more extensive study would be warranted to confirm the presence or absence of serovar Ba as well as those of serovars A, B and C in PNG.

Infections with these serovars are treatable. Standard treatment with a combination of amoxycillin, probenecid, Augmentin and doxycycline, followed by six days of doxycycline, though broadly efficacious against *C. trachomatis* as well as *N. gonorrhoeae*, was not, however, found to be effective in reducing rates of STIs, probably because of the low patient compliance observed in PNG communities. This regimen has now been replaced with a single oral dose of azithromycin (1 g stat), for syndromic treatment of both *C. trachomatis* and *N. gonorrhoeae,* which should result in an improved outcome. The apparent lack of broad immunological protection induced by serovars of *C. trachomatis* and increased potential for infections with alternative types leading to subsequent immunopathology emphasize the need for unwavering adherence to the recommended treatment algorithms.

In all, we have shown in this study that the same major serovars of *C. trachomatis* responsible for urogenital infections detected in populations elsewhere are present and circulating in communities in EHP. Our results show that beside serovars D, E and F, serovar H can be a common pathogen of the genitourinary tract and can colonize the eyes and nasopharynx of children as well. While a more rigorous study would be required to verify the presence of variants, or host and disease associations with specific serovars, the overall serovar distributions in the population groups of our study generally conform to previously published global patterns.

The PCR-based endonuclease digestion method employed in the study may be sensitive and specific and holds promise for rapid genotyping of *C. trachomatis* serovars as demonstrated in the overall agreement of its results with the sequencing data and its capacity to distinguish between the detected serovars and possible genovariants. Despite this, our continued failure to amplify some samples, even after they were purified, shows that there is a need to further optimize its application in our setting. On the basis of continued PCR failure under optimized conditions, we now think that this may be due to an error in the design of the reverse primer (primer CT6) used in the study: instead of incorporating an analogue for bases 'T/C' in the primer to accommodate reported variability at this site (position 1177) in the *C. trachomatis* genome (35, 48), only base 'T' was incorporated. This is crucial as the exact complementary sequence at the 3'-hydroxyl terminal is critical for primer binding, template extension and a successful outcome during PCR. Correcting this should result in a better outcome in future studies.

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REFERENCES

1 **Alder MW, Meheus A.** Epidemiology and control. In: Weatherall DJ, Ledingham JGG, Warrell DA, eds. Oxford Textbook of Medicine, 3rd edition. Oxford: Oxford University Press, 1996:3345-3348.

2 **Centers for Disease Control and Prevention.** Recommendations for the prevention and management of *Chlamydia trachomatis* infections. *MMWR Morb Mortal Wkly Rep* 1993;42:12.

3 **Moncan T, Eb F, Orfila J.** Monoclonal antibodies in serovar determination of 53 *Chlamydia trachomatis* isolates from Amiens, France. *Res Microbiol* 1990;141:695-701.

4 **Frost EH, Deslandes S, Veilleux S, Bourgaux-Ramoisy D.** Typing *Chlamydia trachomatis* by detection of restriction fragment length polymorphism in the gene encoding the major outer membrane protein. *J Infect Dis* 1991;163:1103-1107.

5 **Schachter J, Stoner E, Moncada J**. Screening for chlamydial infections in women attending family planning clinics. *West J Med* 1983;138:375-379.

6 **Moran JS, Aral SO, Jenkins WC, Peterman TA, Alexander ER.** The impact of sexually transmitted diseases on minority populations. *Public Health Rep* 1989;104:560-565.

7 **Brunham RC, Embree JE.** Sexually transmitted diseases: current and future dimensions of the problems in the third world. In: Germain A, Holmes KK, Piot P, Wasserheit JN, eds. Reproductive Tract Infections: Global Impact and Priorities for Women's Reproductive Health. New York: Plenum Press, 1992:35-58.

8 **Brunham RC, Plummer FA.** A general model of sexually transmitted disease epidemiology and its implications for control. *Med Clin North Am* 1990;74:1339-1352.

9 **Stamm WE, Cole B.** Asymptomatic *Chlamydia trachomatis* urethritis in men. *Sex Transm Dis* 1986;13:163-165.

10 **Zelin JM, Robinson AJ, Ridgway GL, Allason-Jones E, Williams P.** Chlamydial urethritis in heterosexual men attending a genitourinary medicine clinic: prevalence, symptoms, condom usage and partner change. *Int J STD AIDS* 1995;6:27-30.

11 **Horner PJ, Cain D, McClure M, Thomas BJ, Gilroy C, Ali M, Weber JN, Taylor-Robinson D.** Association of antibodies to the *Chlamydia trachomatis* heat-shock protein 60 kD with chronic nongonococcal urethritis. *Clin Infect Dis* 1997;24:653-660.

12 **Stephens RS.** Challenge of *Chlamydia* research. *Infect Agents Dis* 1992;1:279-293.

13 **Zhong GM, Brunham RC.** Antigenic determinants of chlamydial major outer membrane protein resolved at a single amino acid level. *Infect Immun* 1991;59:1141-1147.

14 **Newhall WJ, Batteiger VB, Jones RB.** Analysis of the human serological response to proteins of *Chlamydia trachomatis*. *Infect Immun* 1982;38:1181-1189.

15 **Barbosa-Cesnik CT, Gerbase A, Heymann D.** STD vaccines - an overview. *Genitourin Med* 1997;73:336-342.

16 **Cates W Jr, Wasserheit JN.** Genital chlamydial infections: epidemiology and reproductive sequelae. *Am J Obstet Gynecol* 1991;164:1771-1781.

17 **Preece PM, Anderson JM, Thompson RG.** *Chlamydia trachomatis* infections in infants: a prospective study. *Arch Dis Child* 1989;64:525-529.

18 **Weiss SG, Newcomb RW, Beem MO.** Pulmonary assessment of children after chlamydial pneumonia of infancy. *J Pediatr* 1986;108:659-664.

19 **Gravett MG, Nelson HP, DeRouen T, Critchlow C, Eschenbach DA, Holmes KK.** Independent associations of bacterial vaginosis and *Chlamydia trachomatis* with adverse pregnancy outcome. *JAMA* 1986;256:1899-1903.

20 **Martin DH, Koutsky L, Eschenbach DA, Daling JR, Alexander ER, Benedetti JK, Holmes KK.** Prematurity and perinatal mortality in pregnancies complicated by maternal *Chlamydia trachomatis* infections. *JAMA* 1982;247:1585-1588.

21 **Arno JN, Xie C, Jones RB, Van Der Pol B.** Identification of T cells that respond to serovar-specific regions of the *Chlamydia trachomatis* major outer membrane protein in persons with serovar E infection. *J Infect Dis* 1998;178:1713-1718.

22 **Workowski KA, Suchland RJ, Pettinger MB, Stamm WE**. Association of genital infection with specific *Chlamydia trachomatis* serovars and race. *J Infect Dis* 1992;166:1445-1449.

23 **Morré SA, Rozendaal L, van Valkengoed IG, Boeke AJ, van Voorst Vader PC, Schirm J, de Blok S, van Den Hoek JA, van Doornum GJJ, Meijer CJLM, van den Brule AJC.** Urogenital *Chlamydia trachomatis* serovars in men and women with a symptomatic or asymptomatic infection: an association with clinical manifestations? *J Clin Microbiol* 2000;38:2292-2296.

24 **Stamm WE, Stevens CE, Suchland R, Holmes KK, Eschenbach DA, Pettinger M, Wölner-Hanssen P.** Association of infecting serovar with clinical manifestations in *Chlamydia trachomatis* genital infections in women. In: Bowie WR, Caldwell HD, Jones RP, Mårdh PA, Ridgway GL, Schachter J, Stamm WE, Ward ME, eds. Chlamydial Infections. Proceedings of the Seventh International Symposium on Human Chlamydial Infections, Harrison Hot Springs, Canada. Cambridge: Cambridge University Press, 1990:303-306.

25 **Wagenvoort JHT, Suchland RJ, Stamm WE.** Serovar distribution of urogenital *Chlamydia trachomatis* strains in The Netherlands. *Genitourin Med* 1988;64:159-161.

26 **Borrego MJ, Gomes JP, Lefebvre JF, Eb F, Orfila J, Catry MA.** Genotyping of Portuguese *Chlamydia trachomatis* urogenital isolates. *Genitourin Med* 1997;73:561-563.

27 **Ikehata M, Numazaki K, Chiba S.** Analysis of *Chlamydia trachomatis* serovars in endocervical specimens derived from pregnant Japanese women. *FEMS Immunol Med Microbiol* 2000;27:35-41.

28 **Yuan Y, Zhang YX, Watkins NG, Caldwell HD.** Nucleotide and deduced amino acid sequences for the four variable domains of the major outer membrane proteins of the 15 *Chlamydia trachomatis* serovars. *Infect Immun* 1989;57:1040-1049.

29 **Hudson BJ, van der Meijden WI, Lupiwa T, Howard P, Tabua T, Tapsall JW, Phillips EA, Lennox VA, Backhouse JL, Pyakalyia T.** A survey of sexually transmitted diseases in five STD clinics in Papua New Guinea. *PNG Med J* 1994;37:152-160.

30 **Passey M, Mgone CS, Lupiwa S, Suve N, Lupiwa T, Clegg A, Alpers MP.** Community based study of sexually transmitted diseases in rural women in the highlands of Papua New Guinea: prevalence and risk factors. *Sex Transm Infect* 1998;74:120-127.

31 **Tiwara S, Passey M, Clegg A, Mgone C, Lupiwa S, Suve N, Lupiwa T**. High prevalence of trichomonal vaginitis and chlamydial cervicitis among a rural population in the highlands of Papua New Guinea. *PNG Med J* 1996;39:234-238.

32 **Mgone CS, Lehmann D, Passey M, Ekwempu C, Lupiwa T, Suarkia D, Kave H, Yoannes M, Wal T, Saleu G, Namuigi P, Kum E, Kakazo M, Alpers MP.** Perinatal transmission of *Chlamydia trachomatis* in the Eastern Highlands of Papua New Guinea. Abstract A43 in Program and Abstracts of the Thirty-third Annual Symposium of the Medical Society of Papua New Guinea, Goroka, 2-5 Sep 1997:50.

33 **Lehmann D, Sanders RC, Marjen B, Rongap A, Tschäppeler H, Lamont AC, Hendry GMA, Wai'in P, Saleu G, Namuigi P, Kakazo M, Lupiwa S, Lewis DJ, Alpers MP.** High rates of *Chlamydia trachomatis* infections in young Papua New Guinean infants. *Pediatr Infect Dis J* 1999;18:S62-S69.

34 **Hayes LJ, Bailey RL, Mabey DCW, Clarke IN, Pickett MA, Watt PJ, Ward ME.** Genotyping of *Chlamydia trachomatis* from a trachoma-endemic village in The Gambia by a nested polymerase chain reaction: identification of strain variants. *J Infect Dis* 1992;166:1173-1177.

35 **Gaydos CA, Bobo L, Welsh L, Hook EW 3rd, Viscidi R, Quinn TC.** Gene typing of *Chlamydia trachomatis* by polymerase chain reaction and restriction endonuclease digestion. *Sex Transm Dis* 1992;19:303-308.

36 **Persson K.** Epidemiology of serovars of *Chlamydia trachomatis*. In: Bowie WR, Caldwell HD, Jones RP, Mårdh PA, Ridgway GL, Schachter J, Stamm WE, Ward ME, eds. Chlamydial Infections. Proceedings of the Seventh International Symposium on Human Chlamydial Infections, Harrison Hot Springs, Canada. Cambridge: Cambridge University Press, 1990:559-562.

37 **Takeuchi M, Sugase M, Shiga S, Hagiwara T.** Serovar distributions of cervical *Chlamydia trachomatis* isolated in Nagano Prefecture. [Jap] *Kansenshogaku Zasshi* 2000;74:37-42.

38 **Saikku P, Wang SP.** *Chlamydia trachomatis* immunotypes in Finland. *Acta Pathol Microbiol Immunol Scand [B]* 1987;95:131-334.

39 **Kuo CC, Wang SP, Holmes KK, Grayston JT.** Immunotypes of *Chlamydia trachomatis* isolates in Seattle, Washington. *Infect Immun* 1983;1:865-868.

40 **Brunham RC, Kimani J, Bwayo J, Maitha G, Maclean I, Yang C, Shen C, Roman S, Nagelkerke NJD, Cheang M, Plummer FA.** The epidemiology of *Chlamydia trachomatis* within a sexually transmitted disease core group. *J Infect Dis* 1996;173:950-956.

41 **Morré SA, Ossewaarde JM, Lan J, van Doornum GJJ, Walboomers JMM, MacLaren DM, Meijer CJLM, van den Brule AJC.** Serotyping and genotyping of genital *Chlamydia trachomatis* isolates reveal variants of serovars Ba, G, and J as confirmed by *omp*1 nucleotide sequence analysis. *J Clin Microbiol* 1998;36:345-351.

42 **Suarkia D, Lupiwa T, Mgone CS, Passey M, Lehmann D, Alpers MP.** Lymphogranuloma venereum (LGV) *Chlamydia* serovar L3 in the highlands of Papua New Guinea. Abstract in Programme and Abstracts of the Thirty-fourth Annual Symposium of the Medical Society of Papua New Guinea, Port Moresby, 7-11 Sep 1998:64-65.

43 **Barnes RC, Suchland RJ, Wang SP, Kuo CC, Stamm WE.** Detection of multiple serovars of *Chlamydia trachomatis* in genital infections. *J Infect Dis* 1985;152:985-989.

44 **Morré SA, Moes R, van Valkengoed I, Boeke JP, van Eijk JTM, Meijer CJLM, van den Brule AJC.** Genotyping of *Chlamydia trachomatis* in urine specimens will facilitate large epidemiological studies. *J Clin Microbiol* 1998;36:3077-3078.

45 **Parsons GA.** An ocular survey of community school children in Madang Province. *PNG Med J* 1982;25:151-154.

46 **Mann I, Loschdorfer J.** Ophthalmic Survey of the Territories of Papua and New Guinea. Port Moresby: Department of Public Health, 1955:53p.

47 **Dean D, Schachter J, Dawson CR, Stephens RS.** Comparison of the major outer membrane protein variant sequence regions of the B/Ba isolates: a molecular epidemiology approach to *Chlamydia trachomatis* infections. *J Infect Dis* 1992;166:383-392.

48 **Stephens RS, Mullenbach G, Sanchez-Pescador R, Agabian N.** Sequence analysis of the major outer membrane protein gene from *Chlamydia trachomatis* serovar L2. *J Bacteriol* 1986;168:1277-1282.

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Urinary tract infection in infants and young children presenting with fever without a focus in Port Moresby

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SUMMARY

A prospective study was undertaken to document the importance of urinary tract infection (UTI) as a cause of fever without a focus (FWF) in children less than 3 years of age presenting to the Children's Outpatients Department (COPD) of Port Moresby General Hospital (PMGH). 98 children, 55 males and 43 females, with a median age of 17 months and an interquartile range of 5-31.25 months, were recruited. In addition to a history and physical examination each child had a full blood count, a malaria parasite smear, and a urine sample (obtained by clean catch or midstream methods) for dipstick testing, microscopy and culture. Blood culture was performed where practicable. Lumbar puncture and cerebrospinal fluid (CSF) examination were done only if clinically indicated. UTI was diagnosed on urine culture in 9 of the 98 children. Both urinary nitrite and leukocyte esterase tests were sensitive (89%) and specific (96%). Other causes of FWF were classified as non-specific viral infection (31 children), lower respiratory tract infection (11), malaria (7), meningitis (4), bacteraemia (1 neonate) and other or unknown causes. The finding of UTI in 9% of the children is consistent with data from other tropical countries. Checking for urinary tract infection, which can be done using non-invasive methods of urine collection, is an important part of the investigation of infants and children with FWF.

Introduction

It is widely recognized that urinary infection in infancy and childhood is frequently associated with underlying abnormalities of the renal tract such as vesicoureteric reflux, that recurrent urinary tract infection (UTI) may result in renal scarring and chronic pyelonephritis and that early diagnosis of UTI enables preventive measures to be taken with potential benefit to the quality and duration of life for affected children (1-3). UTI may be asymptomatic, or may present in infants and young children with non-specific symptoms and signs, such as irritability and fever. Western studies consistently report that UTI accounts for 3-6% of children younger than 2 years of age presenting with fever without a focus (FWF) (4), and collection and examination of urine is recognized as an important part of the investigation of the young febrile child (5-7). Much less information is available from the tropical world, but studies from Africa suggest that the prevalence of UTI in this group of children may be higher, with rates of 9% and 11.4% reported in two studies (8,9).

The diagnosis of UTI in children presents problems. In addition to the non-specific presentations, diagnosis depends on obtaining a sample of urine. There is an understandable tendency among clinicians to regard the obtaining of a urine sample from a small child as being either too difficult, when it involves catheterization or suprapubic aspiration, or relatively easy, when performed by using a urine collection bag. Unfortunately the latter technique produces contaminated urine in a very high proportion of samples

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and is thus highly inaccurate (10).

We have been aware that the diagnosis of UTI is made infrequently in our setting and have felt that this is most likely to be due to inadequate diagnosis. It is certainly easy to overlook the possibility of UTI in infants and young children. We therefore attempted to find out how frequently this occurs and took children presenting with FWF as a starting point.

The primary aim of the study was to determine the prevalence of UTI as a cause of FWF in Papua New Guinean children. A secondary aim was to assess the feasibility of using the non-invasive clean catch method of urine collection. In addition the study aimed to document the causes of FWF. The study was approved by the Papua New Guinea (PNG) Medical Research Advisory Committee, the School of Medicine and Health Sciences Research Committee and the hospital administration.

Subjects and Methods

This prospective study was carried out in the Children's Outpatients Department (COPD), Port Moresby General Hospital (PMGH) over a 20-week period in 2003. It followed a pilot feasibility study to determine the frequency of FWF and age distribution of the affected children. Inclusion criteria were an age of less than 36 months, an axillary temperature of >37.2°C, the absence of a focus elicited by history and physical examination and no antibiotic treatment in the previous week. Children meeting these criteria were referred by the nursing and medical staff to the main investigator (CMM). After informed verbal consent, a further history, physical examination and investigation were performed, and further management determined.

All children had a full blood examination (FBE), including haemoglobin and white blood count (WBC), a blood smear for malaria parasites, and urine testing, with dipstick for leukocyte esterase and nitrite, microscopy and culture. Blood cultures were taken when practicable, depending on availability of blood culture bottles and amount of blood collected. A lumbar puncture and cerebrospinal fluid (CSF) examination were done only if clinically indicated.

Urine was collected non-invasively, by midstream collection in the older cooperative children and by clean catch in the younger children. For midstream collection the child's external genitalia, particularly the urethral orifice, were washed with clean water before voiding. In the clean catch method the child was placed on a flat surface, the legs abducted, and the genitalia cleaned with water. Light taps were then applied over the suprapubic area while a sterile urine-collecting container was held in place ready to collect the urine. Urine, once obtained, was tested by dipstick for leukocyte esterase and nitrite (Bayer Multistix SG10) and taken immediately to the microbiology laboratory where microscopy and culture using standard culture methods were carried out. A pure growth with a colony count of $>10^5$ organisms/ml was taken as the 'gold standard' of diagnosis of a urinary infection.

Study children who returned to the clinic within the next two days with additional symptoms were classified accordingly.

A diagnosis of non-specific viral infection was made if no cause was apparent after investigation, if there was a lymphocyte predominance on white cell count (WCC) and if the child did not return with additional symptoms. Diagnoses of lower respiratory tract infection and gastroenteritis were made on the basis of the subsequent development of suggestive clinical symptoms and signs.

Data were entered into Epi Info 6 and SPSS 10.0 software programs for analysis. Sensitivity and specificity were calculated for the various urine tests.

Results

The median age of the 98 children (55 males and 43 females) was 17 months (interquartile range 5-31.25 months). 58 children had been febrile for less than 24 hours and fever was described as persistent in 64 children.

Urine was collected by midstream in 56 and by clean catch technique in 42 children. A diagnosis of UTI was made on the basis of culture in 9 children, 5 boys and 4 girls. *Escherichia coli* was cultured from 5, *Klebsiella* species from 3 and *Proteus* from 1. There was no growth from 74 samples and non-significant growth from 15. The results of urine dipstick and microscopy tests, and their respective sensitivities and

specificities, are shown in Table 1. The leukocyte esterase test was positive at all grades in 25 samples, weakly to strongly positive (1+ to 3+) in 12, including 8 of the 9 samples which had culture-positive UTI, and negative in 73 samples. It was recorded as 'trace' for 13 samples and these were regarded as negatives for the purpose of specificity calculations. Nitrite was also positive in 12, including 8 of the 9 with UTI, and negative in 86 samples. 7 of the culture-positive samples were recorded as positive on both dipstick tests. Both leukocyte esterase and nitrite tests had high sensitivity (89%) and specificity (96%) although the specificity of the leukocyte esterase test fell if the trace results were taken as false positives. Combining the two tests (a positive on either one or both) increased the sensitivity to 100% but reduced specificity to 92%.

Urine microscopy suggested UTI in 4 of the 9 culture-positive samples, was not suggestive in 3 and was uncertain in 2. It was suggestive of sterile urine in 79 samples, uncertain in 10, and suggestive in 9. Its sensitivity was therefore 57% and its specificity was 95% if the uncertain samples were regarded as negative. With the inclusion of uncertain samples as positive, sensitivity, though still low, improved to 67% whilst specificity fell to 85% (Table 1).

The clean catch technique compared favourably with the midstream technique (Table 2). Clean catch samples had a lower rate of non-significant growth and resulted in fewer false positive dipstick tests than midstream samples, though differences were not statistically significant ($p = 0.096$ Yates corrected chi squared test for non-significant growth).

All 9 children with UTI had an axillary temperature >38°C (range 38.1-39.2). 3 children with UTI had a peripheral WBC $>25x10^9$/l, 1 a count of $18x10^9$/l and only 2 a count of $10x10^9$/l or less. Clinical signs and symptoms (irritability, diarrhoea, vomiting and abdominal pain) were not predictive of UTI.

7 children were diagnosed with malaria on

TABLE 1

DIPSTICK AND MICROSCOPY RESULTS IN RELATION TO POSITIVE URINE CULTURE

Test	Positive*	Negative	Trace or uncertain	Sensitivity (%)	Specificity (%)
Leukocyte esterase					
UTI	8	1		89	96#
Other	4	72	13		
Nitrite					
UTI	8	1		89	96
Other	4	85			
Urine microscopy					
UTI	4	3	2	57**	95##
Other	5	76	8		

*1+ to 3+ for leukocyte esterase test strip
#81% if trace results included as false positives
**67% if uncertain results included as positives
##85% if uncertain results included as positives
UTI = urinary tract infection

TABLE 2

COMPARISON OF CLEAN CATCH WITH MIDSTREAM URINE COLLECTING METHODS

Collecting method (Number)	UTI by 'gold standard' Number (%)	Non-significant growth Number (%)	False +ve leukocyte esterase Number (%)	False +ve nitrite Number (%)
Clean catch (42)	5 (11.9)	3 (7.1)	1 (2.4)	1 (2.4)
Midstream (56)	4 (7.1)	12 (21.4)	3 (5.4)	3 (5.4)

UTI = urinary tract infection

the basis of a positive blood slide with no other detectable cause of fever. The blood slide was also reported positive in 2 of the children with UTI.

Lumbar puncture was performed in 37 children. *Haemophilus influenzae* was isolated from one sample and *Streptococcus pneumoniae* from another, whilst microscopy suggested meningitis in two more. Only 1 of 42 blood cultures taken had a growth – of *Staphylococcus aureus*.

The diagnostic assessment is shown in Table 3. 31 children were diagnosed with probable non-specific viral infection, 11 with lower respiratory tract infection, 9 with urinary tract infection, 7 with malaria, 4 with meningitis, 1 with bacteraemia, 21 with other diagnoses and in 14 others the diagnosis was unclear.

Discussion

Of the 98 study children 9 (9.2%) had urine culture results complying with our 'gold standard' for diagnosis of UTI. This is similar to that reported recently from Nigeria and rather higher than that of 3-6% reported from the western literature (4,8). Reasons for the higher prevalence are speculative but would include the effect of poor nutrition, which is associated with a higher prevalence of UTI than is found in well-nourished infants (11,12). Our predominance of males is unusual, but our numbers are small, and the study sample was skewed toward males.

We were aware that the generally accepted methods of urine collection in infants and young children are suprapubic aspiration or urinary catheterization. Both these procedures are painful and invasive, and in unpractised hands have the propensity for causing harm. They are impractical in the vast majority of settings in which febrile children present in the developing world. Bag urine collection has been widely used to collect urine from infants and young children, but even when used appropriately has a very low specificity and is not recommended (10). We purposely therefore decided to use the clean catch method of collection and, in older cooperative children, the midstream urine method. In some cases a second attempt was required to obtain a sample but the techniques were successful in every patient. Although numbers were relatively small, the quality of the clean catch samples was at least as good as that of the midstream samples, with a lower contamination rate (as defined by non-significant growth on culture), and a lower rate of false positive dipstick tests. A recently published study comparing contamination rates in urine collected by clean catch, cotton wool pad and urine bag from children less than 3 years of age in Brazil reported a significantly lower contamination rate in the clean catch samples than the other methods (13). The contamination rate for clean catch urine in our study (7.1%) was lower than that reported from Brazil (14.7%). A recent study from Scotland comparing the clean catch with suprapubic aspiration as the gold standard technique reported sensitivity and specificity of 88.9% and 95% for clean catch. Furthermore the same study indicated a success rate of only 13/21 for suprapubic aspiration without ultrasound guidance (14).

TABLE 3

CLASSIFICATION OF CAUSES OF FEVER WITHOUT A FOCUS

Diagnosis	Number (%)
Non-specific viral infection	31 (31.6)
Lower respiratory tract infection	11 (11.2)
Urinary tract infection	9 (9.2)
Malaria	7 (7.1)
Meningitis	4 (4.1)
Bacteraemia	1 (1)
Others	21 (21.5)
Unknown	14 (14.3)
Total	98 (100)

We are therefore confident that our results are highly, if not absolutely reliable. It would seem that at least in the setting of a general paediatric outpatients or even a consultation clinic, the clean catch method is the best option for urinary collection in young children. It depends on the initiation of the voiding reflex by percussion or gentle pressure waves. In this study the children were lying flat, and gentle taps were applied over the suprapubic area. A variation of the method is to hold the child vertical in the 'standing' position and to either gently percuss or to 'bounce' thumb or finger over the suprapubic region. Clean catch urine collection is easy and requires minimal equipment, but may require a little patience.

Urinary dipstick testing with both leukocyte esterase (using 1+ to 3+ as positive) and nitrite dipsticks gave sensitivities of 89% and specificities of 96%. Positive predictive values for the individual tests were 67%, but when combined (using either a positive leukocyte esterase or a positive nitrite test or both) the positive predictive value decreased to 53%. Whilst these results should be interpreted with caution in view of the small numbers involved, they are consistent with other published data (10). Unfortunately the test sticks are relatively expensive and unlikely to be widely available.

Urine microscopy had low sensitivity so that reliance solely on this test would result in missed diagnosis in a significant proportion of children. Inclusion of the uncertain microscopy results as positive increased sensitivity to 67% but at the expense of reducing specificity to 85%, which would result in over-treatment.

Clinical signs of urinary infection in children are generally non-specific, although the recent Nigerian study of febrile infants and children reported an association with abdominal pain and tenderness (8). Our data did not support an association, but were limited and incomplete.

In any area where malaria occurs this should obviously be considered as a major cause of FWF. In our study 7 children were diagnosed to have malaria as the cause of their fever on the basis of a positive blood slide and no other findings. However, the blood slide was also positive in 2 of the children with urinary tract infection. Had a specific search for UTI not been made this diagnosis would have been missed. This reinforces the point that in a situation where malaria is common, additional diagnoses may be present.

In our study, only 1 (2%) of the 42 blood

cultures taken was positive. This was from a neonate and grew *Staph. aureus.* The prevalence and spectrum of bacteraemia and bacterial infection in febrile children is highly dependent on the age, the temperature and other characteristics of the population studied (5). Bacteraemia has been reported in 5% of febrile infants less than 3 months of age and in 4% of children 3-36 months with rectal temperature of ≥38°C (15,16). Our low prevalence may be at least partly explained by the broad selection criteria. By contrast a recent Nigerian study reported bacteraemia in 38% of children, selected on the basis of age 1-12 months, with a rectal temperature ≥38°C and a negative history of antibiotic use, attending the teaching hospital Emergency Ward (17).

The possibility of meningitis was considered on clinical grounds and lumbar puncture performed on 37 children, the diagnosis being confirmed in 4 (11%) (one *Strep. pneumoniae*, one *H. influenzae* and two based on the microscopy of the CSF). This finding reinforces the need to consider the possibility of meningitis in any child presenting with FWF.

Our study can be criticized on the basis that many of the diagnoses were presumptive. However, it was performed in the context of a routine children's outpatient and laboratory service. With the exception of the leukocyte esterase and nitrite tests, the investigations employed in the study were those routinely performed by the PMGH laboratory service. We did not therefore have access to tests that would confirm viral infection, the most frequent presumed cause in our sample. The diagnosis of non-specific viral infection may therefore be queried. However, the diagnosis was made on the basis of the self-limited nature of the illness and all cases so classified had lymphocyte predominance on WCC. Children classified as 'others' included 8 who developed diarrhoea and 5 who were somewhat unsatisfactorily classified as having failure to thrive. It is at least possible that some of these children may have had HIV infection or tuberculosis. There remains the group of 14 children classified as having an 'unknown' cause, in whom there were no clues and no positive laboratory results.

In view of the association of UTI with underlying functional or anatomical abnormality of the renal tract and the long-term consequences of renal scarring, imaging of the urinary system in young children with UTI is recommended routine practice (10). Regrettably only 3 of the 9 children in our study with UTI had an ultrasound scan assessment of their renal system done. The scans were normal.

In conclusion, urinary tract infection was found in 9% of the children studied. It should therefore be routinely considered as a possible diagnosis in children with unexplained fever. The clean catch technique is a practical way of obtaining urine in young children in both hospital and clinic settings.

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REFERENCES

1 **Winberg J, Bergstrom T, Jacobsson B.** Morbidity, age and sex distribution, recurrences and renal scarring in symptomatic urinary tract infection in childhood. *Kidney Int Suppl* 1975;8:S101-S106.
2 **Jodal U.** The natural history of bacteriuria in childhood. *Infect Dis Clin North Am* 1987;1:713-729.
3 **Winberg J, Bollgren I, Kallenius G, Mollby R, Svenson SB.** Clinical pyelonephritis and focal renal scarring. A selected review of pathogenesis, prevention and prognosis. *Pediatr Clin North Am* 1982;29:801-814.
4 **Hoberman A, Chao HP, Keller DM, Hickey R, Davis HW, Ellis D.** Prevalence of urinary tract infection in febrile infants. *J Pediatr* 1993;123:17-23.
5 **Jaskiewitz JA.** Fever without localizing signs in infants and children. In: Rudolph CD, Rudolph AM, Hostetter MK, Lister G, Siegel NJ, eds. Rudolph's Pediatrics, 21st edition. New York: McGraw-Hill, 2003:889-892.
6 **Baraff LJ.** Management of fever without source in infants and children. *Ann Emerg Med* 2000;36:602-614.
7 **Browne GJ, Currow K, Rainbow J.** Practical approach to the febrile child in the emergency department. *Emerg Med* 2001;13:426-435.
8 **Musa-Aisien AS, Ibadin OM, Ukoh G, Akpede GO.** Prevalence and antimicrobial sensitivity pattern in urinary tract infection in febrile under-5s at a children's emergency unit in Nigeria. *Ann Trop Paediatr* 2003;23:39-45.
9 **Akpede GO, Abiodun PO, Sykes RM.** Acute fevers of unknown origin in young children in the tropics. *J Pediatr* 1993;122:79-81.
10 **American Academy of Pediatrics. Committee on Quality Improvement Subcommittee on Urinary Tract Infection.** Practice parameter: the diagnosis, treatment, and evaluation of the initial urinary tract infection in febrile infants and young children. *Pediatrics* 1999;103:843-852.
11 **Rabasa AI, Shattima D.** Urinary tract infection in

severely malnourished children at the University of Maiduguri Teaching Hospital. *J Trop Pediatr* 2002;48:359-361.

12 **Morton RE, Lawande RH.** Frequency and clinical features of urinary tract infection in paediatric out-patients in Nigeria. *Ann Trop Paediatr* 1982;2:113-117.

13 **Alam MT, Coulter JB, Pacheco J, Correia JB, Ribeiro MG, Coelho MF, Bunn JE.** Comparison of urine contamination rates using three different methods of collection: clean-catch, cotton wool pad and urine bag. *Ann Trop Paediatr* 2005;25;29-34.

14 **Ramage IJ, Chapman JP, Hollman AS, Elabassi M, McColl JH, Beattie TJ.** Accuracy of clean-catch urine collection in infancy. *J Pediatr* 1999;135:765-767.

15 **McGowen JE Jr, Bratton L, Klein JO, Finland M.** Bacteremia in febrile children seen in a "walk-in" pediatric clinic. *N Engl J Med* 1973;288:1309-1312.

16 **Powell KR.** Fever without a focus. Chapter 167. In: Nelson WE, Behrman RE, Kleigman RM, Arvin AM, eds. Nelson Textbook of Pediatrics, 15th edition. Philadelphia: WB Saunders, 1996:698-704.

17 **Ayoola OO, Adeyemo AA, Osinusi K.** Predictors of bacteraemia among febrile infants in Ibadan, Nigeria. *J Health Popul Nutr* 2002;20:223-229.

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The use of seatbelts in Port Moresby 12 years after the seatbelt legislation in Papua New Guinea

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SUMMARY

The aim of this study was to assess the rate of seatbelt use by drivers and front-seat passengers in Port Moresby, 12 years after the seatbelt legislation in 1993. Before the legislation, the rate of seatbelt usage was only 13.3% for drivers and 11.4% for front-seat passengers. Use of seatbelts was assessed by observers at the main city roundabout. 50% of male drivers, 78% of female drivers, 49% of Papua New Guinean drivers and 69% of expatriate drivers wore seatbelts. Among the young drivers (teenagers aged <20 years) 55% wore seatbelts. Of the front-seat passengers, 37% of males and 58% of females wore seatbelts. Female drivers and female front-seat passengers were more likely to wear seatbelts than males (OR 2.55 [95% CI 1.53-4.23] and 2.34 [95% CI 1.32-4.14]). The front-seat passengers were more likely to be wearing seatbelts if the drivers wore theirs (OR 2.70 [95% CI 1.60-4.55]). Proportionately more drivers and front-seat passengers were wearing seatbelts than during the pre-legislation period, but more seatbelt education and awareness is needed because of the increasing number of road traffic accidents in Papua New Guinea.

Introduction

It was realized in the early 1980s that road traffic accidents (RTAs) were a major public health problem (1) and that severe injuries and deaths can be prevented by the use of seatbelts. A survey conducted in Port Moresby found the rate of seatbelt use was 13.3% and 11.4% for drivers and front-seat passengers (FSP) respectively (2). Another survey in 1983 showed that the most dangerous positions in a moving vehicle are the open backs of utilities and the front seats (driver and passenger) (3).

Studies in the USA, United Kingdom and Australia have shown that the wearing of seatbelts reduced fatal and severe injuries by 35% (4,5).

The legislation for the compulsory wearing of seatbelts was passed in the National Parliament in 1993, and under this legislation car manufacturers were required to have seatbelts fitted in all vehicles. This was in response to the rising death toll and severe injuries from road traffic accidents in Papua New Guinea (PNG).

In 2000 there were 3635 RTAs, a 16% reduction from 4346 throughout PNG in 1999, with 211 fatalities and 277 people severely injured (6). This represents a financial cost of more than K200 million annually. The National Road Safety Council (NRSC) was established by Act of Parliament in 1997 and given the task of "reducing the carnage on our roads" (6).

Port Moresby with a population of nearly 400,000 is the capital city of Papua New Guinea. In 2002, there were 31,012 motor vehicles and 13,986 drivers registered in the city (7). In the same year, there were 1370 road traffic accidents (some with fatal consequence) and 4200 traffic infringement fines (including failure to wear a seatbelt) issued (8).

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The aim of this study was to assess the rate of seatbelt usage in a sample of drivers and front-seat passengers in Port Moresby, 12 years after the introduction of the legislation for compulsory wearing of seatbelts.

Methodology

This was a cross-sectional study conducted in Port Moresby, during the lunch hour between 12.00 and 1.00 pm for 2 days on 5-6 December 2005.

The Boroko roundabout was chosen as the study site. The locations of the observers were carefully selected to avoid the possibility of counting the same vehicle twice. When the vehicles go around the roundabout, at lower speed, the researchers were able to observe the vehicles and whether or not the drivers and front-seat passengers were wearing seatbelts. An attempt was made to classify drivers as adults or 'teenagers' – based on their appearance. Each vehicle selected was observed by 3 observers without visual aids such as binoculars. The study site was not advertised before the conduct of the study.

The first group of researchers observed vehicles coming from Waigani (west) and turning eastward into the Boroko shopping area. The second group on the other side of the Boroko roundabout observed vehicles driving along Hubert Murray Highway (northbound) from Taurama and turning westbound into Wards Road to Waigani.

Data obtained were entered on to the prepared survey forms. Cars, open-back utilities and small buses up to 15-seater buses were included in the study. Public motor vehicles (PMVs) and large trucks were excluded. The data were entered into Epi Info 6.04 and analyzed.

Results

Drivers

Of the 284 drivers, the majority (88%) were Papua New Guineans (Table 1). Among the drivers, there were 20 'teenagers' and by gender, only 6% of drivers were females.

Nearly half (49%) of the national and 69% of the expatriate drivers were wearing seatbelts. The percentage of female drivers

TABLE 1

WEARING OF SEATBELTS BY DRIVERS

Driver	Wearing of seatbelt				Total N = 284
	Yes		No		
	Number	%	Number	%	
Nationality					
National	122	49.0	127	51.0	249
Expatriate	24	68.6	11	31.4	35
Gender					
Male	132	49.6	134	50.4	266
Female	14	77.8	4	22.2	18
Age group					
Teenager	11	55.0	9	45.0	20
Adult	135	51.1	129	48.9	264

wearing seatbelts was higher (78%) than for males (50%) (OR 2.55, 95% CI 1.53-4.23). When the drivers were categorized into teenagers and adults, it was observed that 51% of adults (including expatriates) and 55% of teenage drivers were wearing seatbelts. All teenage drivers were Papua New Guineans.

Front-seat passengers

Overall, 43% of FSPs wore seatbelts (Table 2). The majority (73%) of FSPs were males but only 37% of them wore seatbelts. Of the 76 female FSPs, 58% wore seatbelts. Females were more likely than males to wear seatbelts (OR 2.34, 95% CI 1.32-4.14).

By categorizing FSP into 3 different age groups, it was observed that 45% of children, 49% of teenagers and 59% of adults were not wearing seatbelts.

Wearing of seatbelts by drivers and front-seat passengers

It was observed that when the drivers wore seatbelts, it was more likely that their FSPs also wore seatbelts. This association was statistically significant (OR 2.70, 95% CI 1.60-4.55) (Table 3).

Wearing of seatbelts by nationality

It was of interest to observe that when the driver was an expatriate, the FSP was more likely to be wearing a seatbelt. This association was statistically significant (OR 2.95, 95% CI 1.33-6.63) (Table 4).

Discussion

In the 10 years from 1968 to 1978, the rate of fatal road accidents rose by 400% and the majority of drivers involved in RTAs had blood alcohol levels above 80 mg/100ml (9-11). Reducing drunk driving and use of seatbelts could have prevented many of these fatalities and injuries. In 1980, the Police Commissioner advocated the use of breath analysis apparatus to reduce drunk driving in order to reduce RTAs (12). This policy has yet to be implemented.

In recent years, there have been frequent police road blocks particularly during the long weekends and Christmas and New Year holidays. At these road blocks, the police and officers from the National Road Safety Council check the vehicle's registration, its roadworthiness and the driver's driving licence. They often remind the drivers and passengers to wear seatbelts and on occasions issue fines for not wearing them.

There are a number of limitations to the present study. Observations were of

TABLE 2

WEARING OF SEATBELTS BY FRONT-SEAT PASSENGERS

Front-seat passenger	Wearing of seatbelt				Total N = 284
	Yes		No		
	Number	%	Number	%	
Gender					
Male	77	37.0	131	63.0	208
Female	44	57.9	32	42.1	76
Age group					
Child	6	54.5	5	45.5	11
Teenager	20	51.3	19	48.7	39
Adult	95	40.6	139	59.4	234

TABLE 3

WEARING OF SEATBELTS BY DRIVERS AND FRONT-SEAT PASSENGERS

Driver wearing seatbelt	Front-seat passenger		Total
	Yes	No	
Yes	79	67	146
No	42	96	138
Total	121	163	284

Odds ratio = 2.70; 95% confidence interval 1.60-4.55; p <0.001

TABLE 4

WEARING OF SEATBELTS BY DRIVER'S NATIONALITY AND FRONT-SEAT PASSENGERS

Nationality of driver	Front-seat passenger wearing seatbelt		Total
	Yes	No	
Expatriate	23	12	35
National	98	151	249
Total	121	163	284

Odds ratio = 2.95; 95% confidence interval 1.33-6.63; p = 0.003

necessity made on the occupants of moving vehicles and were therefore brief. The assessment of age of the occupants was highly subjective and the study sample was small. Nevertheless, the study does provide a 'snap-shot' of the rate of seatbelt usage in Port Moresby.

In this study, 12 years after the introduction of the seatbelt legislation, the rate of wearing of seatbelts by Papua New Guinean drivers has increased to 49% compared to the pre-legislation rate of 13.3% (2). However, this is still low compared to 76-77% in developed countries such as England, France or Germany (13).

More expatriates and female drivers wore seatbelts than Papua New Guinean male drivers. It was encouraging to observe that 55% of teenage drivers wore seatbelts.

Overall, 43% of front-seat passengers wore seatbelts. This is better than the pre-legislation statistic of 11.4% (2). The majority (73%) of the FSPs were males but only 37% of them wore seatbelts. Like the drivers, more female FSPs than males wore seatbelts. Of major concern is that many children (45%) and teenagers (49%) in the front seats were not wearing seatbelts. This was related to the driver's own seatbelt use behaviour, and awareness of the importance of seatbelts. There was a significant association (p <0.001) between the driver and front-seat passenger wearing seatbelts.

There was also a strong association (OR = 2.95, p = 0.003) between the FSP wearing a seatbelt and the driver being an expatriate.

Evaluative studies in the United Kingdom and elsewhere have shown that wearing of seatbelts by drivers and FSPs reduces both deaths and injuries (14-16).

A 2002 survey of 50 drivers by Chief Sergeant Joseph Joe (Traffic Police) in Port Moresby found that 78% obtained their driving licences without going to a formal driving school. Of major concern were the 30% of drivers who only observed others driving and still managed to get their driving licences. They were able to drive a vehicle without learning the road safety rules (17).

The National Government must support the NRSC and the schools to educate our young people about road safety. Our young people need to be educated about the importance of wearing seatbelts. The entry points for this educational intervention would be the schools, the community and the driving schools.

The National Road Safety Council is actively involved in many aspects of road safety in this country. One of the Council's projects is the Community Road Safety Education Program. The Council conducts community education through the media, eg, Drive Time and 'Talk Back' radio (NBC) programs that provide road safety tips such as wearing of seatbelts to listeners (6).

Conclusion

The legislation has had some impact with a higher percentage (49%) of Papua New Guinean drivers and 43% of FSPs wearing a seatbelt. A FSP is more likely to wear a seatbelt if the driver wears one. Further improvements will require government support for road safety education and awareness in the community and schools on the importance of seatbelt usage.

It would be good to have a follow-up study in future on the knowledge, attitude and behaviour regarding seatbelt usage of both drivers and front-seat passengers.

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REFERENCES

1 **Sinha SN.** Seat belts, road safety, society and surgeons. *PNG Med J* 1983;26:7-8.
2 **Lourie JA.** Use of seat-belts in Port Moresby. *PNG Med J* 1982;25:214-218.
3 **Lourie JA, Sinha SN.** Port Moresby road traffic accident survey. *PNG Med J* 1983;26:186-189.
4 **SenGupta SK, Patil NG, Law G.** Seat-belt messages and the law? *PNG Med J* 1989;32:199-202.
5 **Thomas J.** Road traffic accidents before and after seatbelt legislation – a study in a district general hospital. *J R Soc Med* 1990;83:79-81.
6 **National Road Safety Council.** National Road Safety Council Annual Report 2000. Papua New Guinea National Road Safety Council, Port Moresby, 2000.
7 **Papua New Guinea Department of Transport.** Papua New Guinea Department of Transport (Motor Traffic Registry) Report 2002. Papua New Guinea Department of Transport, Port Moresby, 2002.
8 **Papua New Guinea Department of Police.** Papua New Guinea Department of Police (Traffic) Report 2002. Papua New Guinea Department of Police, Port Moresby, 2002.
9 **Wyatt GB.** The epidemiology of road accidents in Papua New Guinea. *PNG Med J* 1980;23:60-65.
10 **Watters DAK, Dyke T.** Trauma in Papua New Guinea: what do we know and where do we go? *PNG Med J* 1996;39:121-125.
11 **Papua New Guinea Department of Transport.** Road Traffic Seminar. Proceedings of the Second National Road Safety Seminar, Lae, May 1994.
12 **Bouraga P.** Road traffic accidents and the role of the police. *PNG Med J* 1980;23:59.
13 **Steptoe A, Wardle J, Fuller R, Davidsdottir S, Davou B, Justo J.** Seatbelt use, attitudes, and changes in legislation: an international study. *Am J Prev Med* 2002;23:254-259.
14 **Newman RJ, Jones IS.** A prospective study of 413 consecutive car occupants with chest injuries. *J Trauma* 1984;24:129-135.
15 **McCarthy M.** The benefit of seat belt legislation in the United Kingdom. *J Epidemiol Community Health* 1989;43:218-222.
16 **Ward NJ, Okpala E.** Analysis of 47 road traffic accident admissions to BMH Shaibah. *J R Army Med Corps* 2005;151:37-40.
17 **Joe JP.** A case study on road safety in Port Moresby, NCD. Papua New Guinea Institute of Public Administration, Port Moresby, 2002.

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Large-vessel injuries of the neck admitted to Chongqing Emergency Medical Centre, China and Port Moresby General Hospital, Papua New Guinea, 1996-2006

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SUMMARY

Stab wounds are the main cause of large-vessel injuries in the neck and they have a fairly good prognosis when the patient receives expeditious and appropriate management. The objective of this study is to present the experience of managing patients with large-vessel injuries in the neck. A retrospective study was carried out involving 22 cases with vessel injuries in the neck over the past 10 years. Stab wounds accounted for 77% of this series (17 of 22 patients). All 22 cases were treated operatively. The overall survival rate was 95%. One patient died on the operating table because of torrential haemorrhage into the chest. Complications included one thrombosis of the right subclavian artery, and five cases of haemothorax, all of which resolved. Successful salvage of patients with major vessel injuries in the neck depends on familiarity with the anatomy, accurate and timely clinical diagnosis and expedient surgical intervention. Patients with haemodynamic instability, rapidly expanding cervical haematomas or uncontrollable bleeding require immediate operative intervention, forgoing any diagnostic study. Stable patients may undergo radiological studies to detect occult injuries that may result in late morbidity such as false aneurysms and arteriovenous fistulae.

Introduction

Trauma, both blunt and penetrating, is extremely common in China and Papua New Guinea. As a result, trauma to major vessels, in particular arteries, is a common clinical occurrence. Vascular injury of the neck is more fatal because of the difficulty with examining and operatively exposing these areas compared to other regions of the body. The high density of vascular, neurological and visceral structures warrants suspicion of injury and thorough appreciation of cervical anatomy. There are three anatomical zones of the neck that are important in the management paradigm of penetrating cervical injuries (Figure 1). Zone one is between the sternal notch and the cricoid cartilage; zone two is between the cricoid cartilage and the angles of the mandible; and zone three is above the angles of the mandible (1,2). Zone one and zone three injuries are more difficult to expose and more complicated to manage. Zone two injuries are exposed easily and operative intervention is usually chosen. The diagnosis of vascular injury was proven by surgical exploration in most cases. In some patients whose haemodynamics were stable, preoperative diagnosis was obtained through angiography, Doppler studies and computed tomography (CT) scan.

The management protocol has remained the same for the past 10 years: that is, if the diagnosis of vascular injury of the neck is obvious or highly suspicious, and located at zone two, the patient should be promptly transferred to the operating room and a vascular exploration performed. In our patients limited preoperative intravascular replenishment was carried out, and preoperative exploration of the wound tract was avoided.

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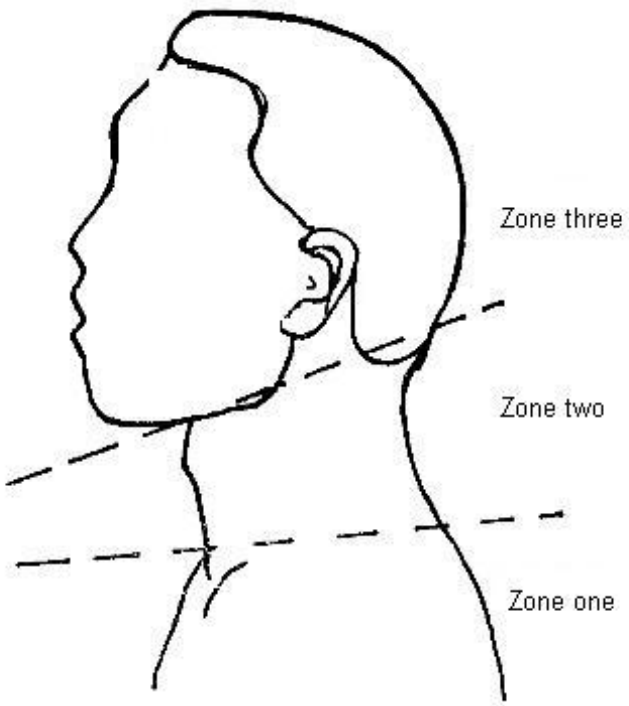

Figure 1. The three zones of the neck.

Materials and Methods

This was a retrospective study of 22 cases (20 patients from Chongqing Emergency Medical Centre, China and 2 patients from Port Moresby General Hospital) with vascular injury of the neck treated during a 10-year period between April 1996 and October 2006. All these cases were operated on by the authors. Information was reviewed regarding the patients' sex; age; the type of instrument inflicting the wounds; duration of time before admission; vital signs; clinical type on admission; preoperative evaluation; resuscitative treatment in the emergency department, which included diagnostic modalities and efficacy of the preoperative treatment; time taken to get the patient to the operating room; the location and zone of the vascular wound; the amount of blood transfused during the operation; associated organic injuries; the surgical therapy; and the ultimate outcome (uneventful recovery, complications or death).

Results

In this series of 22 patients, there were 17 males and 4 females (1 not recorded), with ages ranging from 16 to 52 years (average 28 years). 17 patients (77%) sustained stab wounds, 2 had gunshot wounds, 1 had an iatrogenic injury, 1 had a motor vehicle crash and 1 was gored by a bull (Table 1).

In addition to the injuries to the vessels, injuries to the trachea were seen in 2 patients, the brachial plexus in 2 patients and the oesophagus in 2 patients. The wound was located in zone two in 14 cases, zone one in 7 cases and zone three in 1 case (Table 2). The length of the vascular wounds was from 0.5 cm to 8 cm. 6 patients had an associated haemothorax and/or pneumothorax, 1 had diaphragmatic injuries and 2 had associated injuries of abdominal viscera. 16 patients (73%) were in hypovolaemic shock. The amount of blood loss was over 4000 ml in 3 patients, and it was 1500-4000 ml in the

TABLE 1

CAUSES OF VASCULAR INJURY WITH THE VESSELS INJURED

Vessel injured	Cause of injury				
	Stab	Gunshot	Iatrogenic	MVA	Gored
Common carotid artery	4	1	-	-	-
Innominate vein	2	-	-	-	-
Jugular vein	3	-	-	-	-
Internal carotid artery	1	1	-	-	-
External carotid artery	6	-	-	-	-
Vertebral artery	-	-	1	-	-
Subclavian artery	5	-	-	1	1
Subclavian vein	4	-	1	-	1
Arteriovenous fistula and false aneurysm	1*	-	-	-	1

Some patients had more than one injury
MVA = motor vehicle accident
*Shown in Figure 2

TABLE 2

OPERATIVE MANOEUVRES FOR VASCULAR INJURIES WITH THE NUMBER OF CASES AND ZONE OF THE INJURIES

Operative manoeuvre	Cases	Zone of the injury
Repair	16	Two (10 cases) or one (6 cases)
Vein graft	2	Two or one
External-internal carotid transposition	1	Two
Ligation	2	Two
Selective angiography and embolization	1	Three

remainder (average 1765 ml). 3 patients had airway tamponade and 4 had mediastinal emphysema.

All 19 cases who presented acutely were explored as an emergency. The 2 cases with an arteriovenous fistula and false aneurysm of the cervical vessels (Figure 2) had exploration performed at 3 and 6 months after injury. The operative manoeuvres employed for the vascular injuries with the number of cases and zone of the injuries are shown in Table 2. 1 patient died on the operating table because of torrential haemorrhage into the chest. The overall case fatality rate was 5% (1/22). Complications included 1 thrombosis of the right subclavian artery and 5 with haemothorax, all of which resolved.

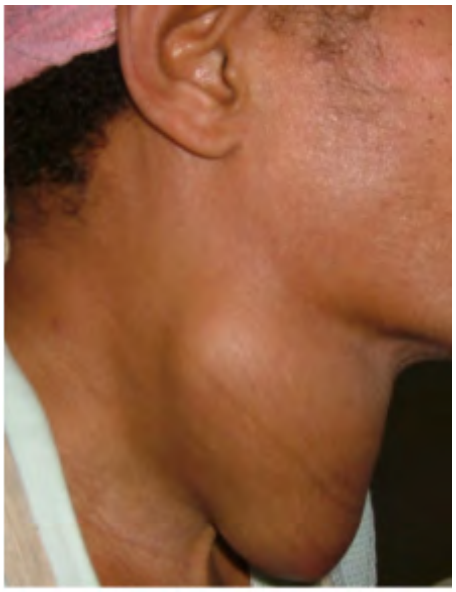

Figure 2. False aneurysm in the neck. A case from Port Moresby General Hospital.

Discussion

Penetrating injuries of the neck are generally considered difficult to evaluate and manage. The neck is a small anatomical area with a dense concentration of vital structures, many of which are not easily accessible to clinical examination, and their surgical exposure may be technically difficult. Cervical vascular injuries occur more commonly after penetrating trauma, and approximately 25% of penetrating neck injuries result in a vascular injury (3).

Successful salvage of patients with major vessel injuries in the neck depends on familiarity with the anatomy, accurate and timely clinical diagnosis and expedient surgical intervention. Early recognition of vessel injuries in the neck is the precondition of success (4). The outcome is better if repair is done early (5). Thus any patient with a possible vascular injury should be urgently explored, especially where arteriography is not available (6). In those patients, delay caused either by excessive moving of the patient or time-consuming diagnostic procedures can be lethal. The diagnosis should depend mainly on the signs (haemodynamic lability, rapidly expanding cervical haematomas or uncontrollable bleeding). Further knowledge of some factors in relation to the injury should include the shape and length of the weapon and the site, size, depth and direction of the wound.

Operative exposure of zone one injuries may necessitate a supraclavicular incision (Figure 3), with removal of the head of the clavicle or a 'trapdoor' or 'book' thoracotomy. Zone one and zone three injuries are more difficult to expose and more complicated to manage. Zone two injuries are exposed easily and operative intervention is usually chosen (1).

Patients with haemodynamic instability, rapidly expanding cervical haematomas or uncontrollable bleeding require immediate operative intervention. Emergency operative exploration was performed in 19 of the patients in this study, with good recovery after surgery. In the patients with stable haemodynamics, however, angiography and Doppler studies are very useful and should be performed. Angiography was performed and correctly diagnosed vessel injuries in the neck in 6 of the patients in this study. Special attention should be given to delayed haematoma of the neck, as occurred in 2 patients in this series, with good recovery after surgery.

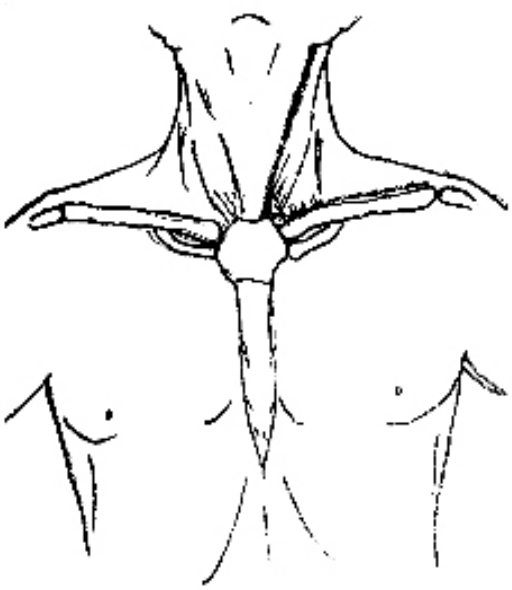

Figure 3. Standard neck supraclavicular incision shown above the left clavicle.

Imaging evaluation of patients suspected to have arterial injuries of the neck has been of recent interest and there is now a growing experience with the use of non-invasive imaging techniques for the assessment of vascular injuries. CT angiography is increasingly being used to evaluate trauma patients in a stable condition who are at risk for vascular injuries.

A correct policy for preoperative management is important. In most of the patients with vessel injuries of the neck arriving at the hospital alive, the bleeding might have ceased or have been relieved as a result of spontaneous factors, eg, hypotension, clot formation or the natural tamponade effect provided by the deep cervical fascial matrix. Therefore we believe that it is not wise to break this balance before surgical exploration. From the experience of the present study, three principles of preoperative management can be suggested.

Firstly, excessive transfusion should be avoided. Massive intravascular volume expansion is not only of no help, but it may also provoke life-threatening recurrent haemorrhage from the vascular wound because of the rise in vascular pressure and the dislodgement of the clot already formed. Preferably, an adequate amount of blood is given only after bleeding from the vascular wound has been surgically checked. However, a limited infusion of crystalloid solution and moderate volume of blood may if necessary be given before controlling the vascular wound in order to ensure that the blood pressure is not less than 80 mmHg.

Secondly, wounds should not be probed, cannulated or locally explored because these manoeuvres can dislodge a clot and lead to uncontrolled haemorrhage or embolism. A weapon that remains in the wound should not be removed until the start of the operative exploration (7).

Thirdly, intercostal tube drainage is usually used as a routine preoperative procedure for zone one of neck trauma under positive pressure to prevent tension pneumothorax.

Uncontrollable exsanguinations may occur after decompression of the haematoma. It is possible to provide a relatively avascular field by finding and blocking the vascular wound with digital pressure throughout the skin preparation and until direct vascular control is achieved. The bleeding is arrested by finger

pressure.

Delayed or missed diagnosis and treatment are far more harmful than a negative surgical exploration. Therefore, when a high index of suspicion for vessel injuries in the neck exists, immediate operative exploration should be performed to avoid disastrous outcome.

Conclusions

From the experience of this study, early diagnosis and prompt exploration are the fundamental factors affecting patient outcome after large-vessel injuries in the neck. When a high index of suspicion for large-vessel injuries in the neck exists, immediate exploration should be performed, because delayed or missed diagnosis and treatment are far more harmful than a negative surgical exploration. To manage these patients preoperatively, excessive transfusion should be avoided before controlling the bleeding from the vessel distal and proximal to the vascular trauma, and preoperative exploration of the wound tract is not advocated.

REFERENCES

1 **Britt L, Peyser M.** Penetrating and blunt neck trauma. In: Mattox KL, Feliciano DV, Moore EE, eds. Trauma, 4th edition. New York: McGraw-Hill, 1999:437-450.

2 **Perry MO.** Injuries of the brachiocephalic vessels. In: Rutherford RB, ed. Vascular Surgery, 4th edition. Philadelphia: WB Saunders, 1995:705-712.

3 **Asensio JA, Valenziano CP, Falcone RE, Grosh JD.** Management of penetrating neck injuries. The controversy surrounding zone II injuries. *Surg Clin North Am* 1991;71:267-296.

4 **Gao JM, Gao YH, Wei GB, Liu GL, Tian XY, Hu P, Li CH.** Penetrating cardiac wounds: principles for surgical management. *World J Surg* 2004;28:1025-1029.

5 **Mathew PK, Kainge T, Kaupa F, Barua R.** Management of vascular trauma in a provincial hospital. *PNG Med J* 1996;39:126-128.

6 **Haina T, Ponifasio P, Jacob OJ, Watters DAK.** Traumatic false aneurysms in Port Moresby. *PNG Med J* 1999;42:77-83.

7 **Maxwell RA.** Penetrating neck injury. In: Peitzman AB, Rhodes M, Schwab CW, Yealy DM, Fabian TC, eds. The Trauma Manual, 2nd edition. Philadelphia: Lippincott Williams & Wilkins, 2002:189-197.

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Body size of Papua New Guineans: a comparison of the body mass index of adults in selected urban and rural areas of Papua New Guinea

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SUMMARY

This is a cross-sectional study conducted in Port Moresby and 3 rural areas of Papua New Guinea from 1999 to 2002. These areas were selected because of their specific characteristics such as modernity, geographical location and remoteness. The aim of the study was to compare the body mass index (BMI) of selected urban and rural populations. When age was standardized, in urban and periurban populations, the mean BMI increased with age to about 40 years, plateaued and then decreased in older age. The BMI was higher in Port Moresby than in the other study areas: many people in Port Moresby were overweight (40%) and obese (21%), and by gender, 26% of females and 16% of males were obese. In Manus, the prevalence of overweight and obesity was 36% and 18% respectively. In both Port Moresby and Manus, more women than men were obese. Obesity was not a problem in rural areas of Strickland and Central Province. In rural Central Province 52% of subjects had a BMI <20 kg/m^2. Obesity is becoming a public health problem in the urban areas. The high prevalence of overweight and obesity corresponds with the high intake of refined carbohydrates and fatty foods in urban and periurban areas. It will be necessary to carry out health awareness and education on the risk factors associated with obesity in the urban and periurban areas and promote healthy environments: healthy foods should be available and affordable, and the accessibility and safety of exercise and walking tracks must be supported by the community and government agencies.

Introduction

This study was part of a larger study of the prevalence of diabetes mellitus in the urban and rural areas of Papua New Guinea. This paper reports on the comparison of body mass index (BMI) of subjects in Port Moresby and three rural areas of Papua New Guinea (PNG). The objective of this study was to compare the body mass index of Papua New Guinean adults living in urban and rural areas.

In Papua New Guinea, overweight or obesity is not seen as a health problem and is even admired. We expect our government leaders, politicians and senior executives to look 'big' (overweight or obese) because it is viewed as a sign of good health and wealth. This view is also shared by our other Pacific neighbours and many parts of Africa (1,2).

BMI is calculated as weight in kilograms divided by height in metres squared (kg/m^2). The BMI indicates the nutritional status of the community. Based on western standard BMI criteria, a value of 20.0 – 25.0 kg/m^2 is considered to be normal, 25.1 – 29.9 kg/m^2 signifies the subject is overweight and a value of 30 kg/m^2 or more signifies obesity (3,4).

Methodology

Study sites

This is a cross-sectional study conducted in Port Moresby, the Mt Obree area in the foothills of the Owen Stanley Ranges in Central Province, the Upper Strickland River area between Southern Highlands and West Sepik Provinces, and Balopa District in Manus Province from 1999 to 2002. The study sites were purposely selected because

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of their specific characteristics.

Port Moresby

Port Moresby is the most developed and urbanized centre and the capital city of Papua New Guinea. People living in Port Moresby come from all over PNG. There are many modern supermarkets easily accessible by the city residents and periurban villagers.

Upper Strickland River area

Upper Strickland River area is a very remote area along the Strickland River separating Southern Highlands and West Sepik Province. There are very few government services and infrastructure and no cash crops in the area but villagers living near the Strickland River have received some benefits (royalties) from the Porgera Mine.

The villages surveyed were Egali, Bulago, Aluni and Yokona in the Southern Highlands and Bimin, Kunanap, Dupan, Gawa and Sisimin in the West Sepik Province. Two villages (Egali and Bimin) are accessible by plane but the rest only by helicopter or walking.

Mt Obree, Central Province

Mt Obree is a very remote area without any economic activity and no cash crops. The area is situated in the southern foothills of the Owen Stanley Ranges in Central Province. It is accessible only by plane or a 3-4 days' walk (local persons) through rugged mountainous terrain to Sogeri or Kwikila government stations, which are about 45 and 90 kilometres respectively from Port Moresby. Of the 10 villages, 9 were surveyed.

Balopa Islands, Manus Province

The islands of Balopa on the south coast of Manus Island were selected because of their apparent 'remoteness' but with some periurban characteristics. The islands are about 3-4 hours away by open motorized boat ride from Lorengau, the provincial town. They have cash crops such as coconut and cocoa and also sell marine products such as fish, shells and various types of sea cucumber. The islanders would come into town in the morning to sell their produce in the market, do their shopping and return to their village in the evening. Six villages on the islands were surveyed.

Sampling

The subjects were informed about the study well in advance and individuals volunteered to participate in the survey. Pregnant women and subjects aged less than 20 years old were excluded from the study.

Data collection methods

The weights were measured using a standard portable bathroom foot scale. The subjects were weighed without heavy clothing or shoes. The weighing scale was regularly calibrated using a 5.0 kg metal weight. In Port Moresby and Manus, the height was measured using the standard 'pull down' height-measuring tape. The subject would stand barefooted directly under the tape, which was pulled down to touch the top of the head. In rural Strickland and Mt Obree, height was measured using the height-measuring stand. Subjects stood against the metal stand with the measuring plate lowered to touch the top of the head; the reading was recorded to the nearest centimetre.

Results

Age

The subjects in Manus were older with a mean age of 50 and 47 years for males and females respectively. In Port Moresby, mean age was similar for males and females at 36 years. Among the females, Strickland had the lowest mean age of 34 years (Table 1).

Weight

Overall, the mean and median weight and interquartile range of Port Moresby subjects were 71.9 kg, 72 kg and 62-80 kg respectively. In Manus, the mean, median and interquartile range were 65.5 kg, 65 kg and 57-74 kg respectively. Subjects in the Strickland area had a mean, median and interquartile range of 51.2 kg, 50 kg and 47-56 kg respectively. Central recorded the lowest mean, median and interquartile range of 49.6 kg, 50 kg and 45-55 kg respectively.

Subjects in Port Moresby had the highest mean weight of 74 kg and 69 kg for males and females respectively whilst Central had the lowest mean weight of 53 kg and 47 kg

TABLE 1

MEANS OF SELECTED CHARACTERISTICS BY GENDER AND STUDY SITE

	Study site			
	Port Moresby	**Manus**	**Central**	**Strickland**
Males				
Total	367	116	149	72
Age (years)	35.7 (35.0)* sd=10.6	49.9 (46.0)* sd=17.1	37.4 (32.0)* sd=13.9	46.8 (31.0)* sd=15.7
Height (cm)	165.9	161.7	162.4	150.7
Weight (kg)	74.4	68.1	53.1	52.7
BMI (kg/m^2)	26.5 sd=4.1	25.5 sd=4.4	19.6 sd=2.1	22.7 sd=3.1
Females				
Total	356	204	176	51
Age (years)	35.6 (35.0)* sd=9.3	46.7 (50.0)* sd=15.4	35.2 (32.0)* sd=11.9	33.8 (46.0)* sd=9.1
Height (cm)	156.5	152.9	155.9	145.7
Weight (kg)	69.4	63.9	46.6	49.2
BMI (kg/m^2)	27.8 sd=4.9	26.7 sd=5.3	18.7 sd=2.4	22.9 sd=2.9

*median age
sd=standard deviation
BMI=body mass index

for men and women respectively (Table 1).

Central had the lowest mean and median BMI. There was no obesity observed in rural Central. The highest BMI was recorded in Port Moresby. In rural Strickland, the mean BMI was the same for males (22.7 kg/m^2) and females (22.9 kg/m^2) (Table 1).

Body mass index and gender

In Port Moresby, obesity among males and females was 16% and 26%, and in Manus 13% and 20% respectively (Table 2).

Among the Port Moresby subjects, 5% of females and 4% of males had a BMI <20 kg/m^2 while in rural Central 44% of males and 59% of females had a BMI <20 kg/m^2 (Table 2). BMI less than 20 kg/m^2 is considered to be underweight by the World Health Organization (WHO) (5,6).

Body mass index and age group

Men

In Port Moresby and Manus the mean body mass index of men increased with age up to

TABLE 2

BODY MASS INDEX OF MALES AND FEMALES BY STUDY SITE

	Port Moresby n=723		Manus n=320		Central n=325		Strickland n=123		Total n=1491
Gender									
Males	367		116		149		72		704
Females	356		204		176		51		787
Body mass index									
Males									
<20	13	3.5%	4	3.4%	66	44.3%	6	8.3%	89
20.0-25.0	149	40.6%	61	52.6%	81	54.4%	56	77.8%	347
25.1-29.9	146	39.8%	36	31.0%	2	1.3%	9	12.5%	193
30+	59	16.1%	15	12.9%	0	0	1	1.4%	75
Females									
<20	18	5.1%	10	4.9%	104	59.1%	6	11.8%	138
20.0-25.0	100	28.1%	74	36.3%	72	40.9%	38	74.5%	284
25.1-29.9	145	40.7%	79	38.7%	0	0	6	11.8%	230
30+	93	26.1%	41	20.1%	0	0	1	2.0%	135

about 40 years, plateaued and then decreased with age. In all study sites, BMI decreased in older age (Figure 1). The mean BMI was greater than 20 kg/m^2 in all age groups in Port Moresby, Manus and Strickland; in the rural Central area, except for the 25-34 year age group, it was less than 20 kg/m^2 (Figure 1).

Women

The body mass indexes of women in Manus and Port Moresby were very similar in all age groups. In these sites, the mean BMI of just under 25 kg/m^2 at 20-24 years of age increased to about 28 kg/m^2, then plateaued until 55-64 years of age, after which it declined to 23 kg/m^2. In Strickland the mean BMI of women was highest at 25 kg/m^2 in the 20-24 year age group and decreased with age to 19 kg/m^2 in the group aged 65 years and above (Figure 2). Central women had the lowest mean BMI, which was less than 20 kg/m^2 in all age groups and decreased to 14.5 kg/m^2 at the age of 65 years and above. In all study sites, BMI decreased in older age.

Nutritional status of subjects by study site

Based on the WHO standard of normal BMI of 20-25 kg/m^2 (5,6) 67% women and 56% of men in Port Moresby were overweight or obese. In Manus, 59% of women and 44% of men were overweight or obese. Overweight or obesity was not a problem in the Central or Strickland areas (Table 3).

Prevalence of obesity by age group and study site

In Port Moresby the prevalence of obesity was high at 29% in the age group of 35-44 years and 15% in the older age group of 55

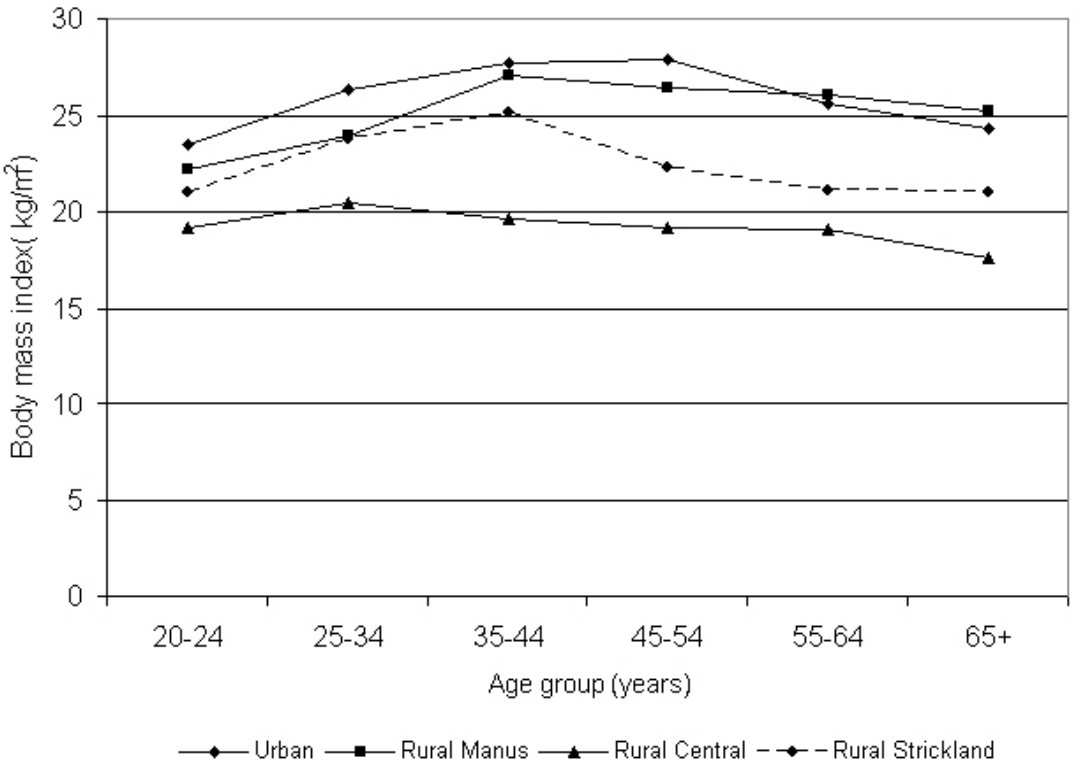

Figure 1. Mean body mass index of males by age group and study site.

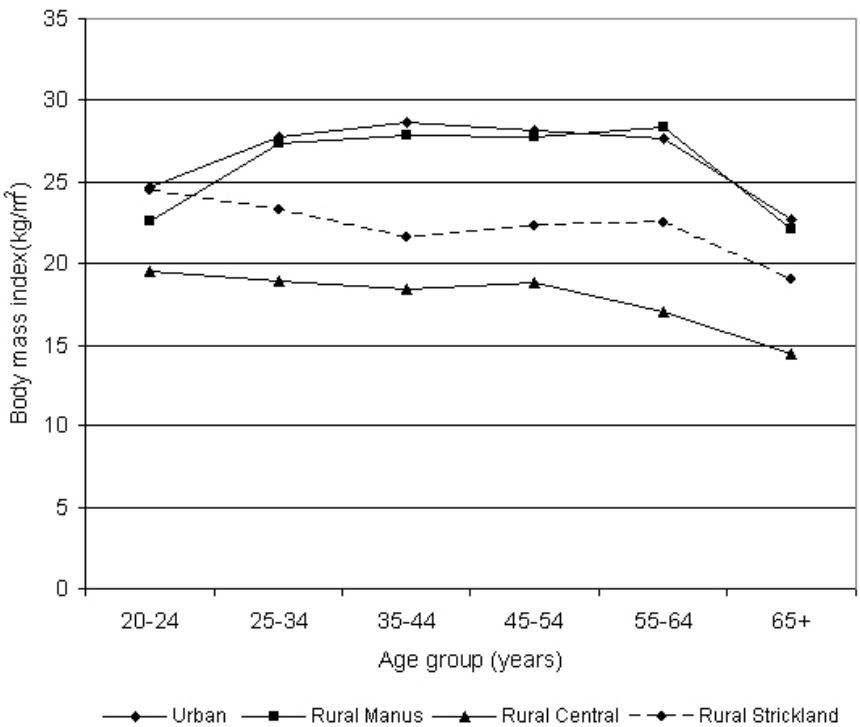

Figure 2. Mean body mass index of females by age group and study site.

TABLE 3

NUTRITIONAL STATUS OF ALL SUBJECTS BY STUDY SITE

	Port Moresby	Manus	Central	Strickland	Total
Males	n=367	n=116	n=149	n=72	704
Normal weight	162 (44%)	65 (56%)	147 (99%)	62 (86%)	436
Overweight	146 (40%)	36 (31%)	2 (1%)	9 (13%)	193
Obese	59 (16%)	15 (13%)	0	1 (1%)	75
Females	n=356	n=204	n=176	n=51	787
Normal weight	118 (33%)	84 (41%)	176 (100%)	44 (86%)	422
Overweight	145 (41%)	79 (39%)	0	6 (12%)	230
Obese	93 (26%)	41 (20%)	0	1 (2%)	135

years and over. In Manus, the prevalence of obesity was 22% in the 25-34 year age group and 16% in the group aged 55 years and above (Figure 3). In the Strickland, less than 4% of subjects in the 25-34 and 35-44 year age groups were obese. There was no obesity observed in rural Central (Figure 3).

Discussion

Body mass index

WHO accepts a body mass index of 20-25 kg/m^2 as normal, 25.1-29.9 kg/m^2 as overweight and 30 kg/m^2 or more as obese. The normal and desirable body mass index is considered to be 20-25 kg/m^2 (5-7). In this study, 61% of the urban subjects were overweight or obese and 21% were obese.

The mean BMI was higher in urban Port Moresby and Manus subjects (males and females) than in Strickland and Central. It was surprising to see that the mean BMI of rural Manus was similar to that in Port Moresby. This is because, unlike in the other two study sites, the people in rural Manus have easy access to western-type foods, particularly refined carbohydrates and fatty foods. This is significant, even though their diet may not be the same in terms of quantity, frequency or variety as the diet of those living in Port Moresby, who have easy access to shops and supermarkets.

In rural Central Province, the majority of subjects (52%) had a BMI between 18 and 20 kg/m^2, and no obesity was seen. This is because the people in rural Central still live on a more traditional diet and are physically more active. According to the WHO standards, a BMI less than 20 kg/m^2 indicates undernutrition, but the people there look healthy and well.

When age was standardized, the mean BMI of males increased with age (except in Central) to about 40 years, plateaued in Port Moresby and Manus, and then decreased (Figure 1); in females only those in Manus and Port Moresby had BMI patterns similar to the males. In general, the BMI of both males and females decreased in older age. This finding is similar to the Kitava study done in Milne Bay where BMI decreased in older age groups (8).

Traditional diet

The people of rural Central and Strickland areas could not be more rural than they are at the present time. Their diet is very traditional, with very little western-type food.

A traditional diet is mainly vegetarian, low in fat and protein. Crops such as cassava,

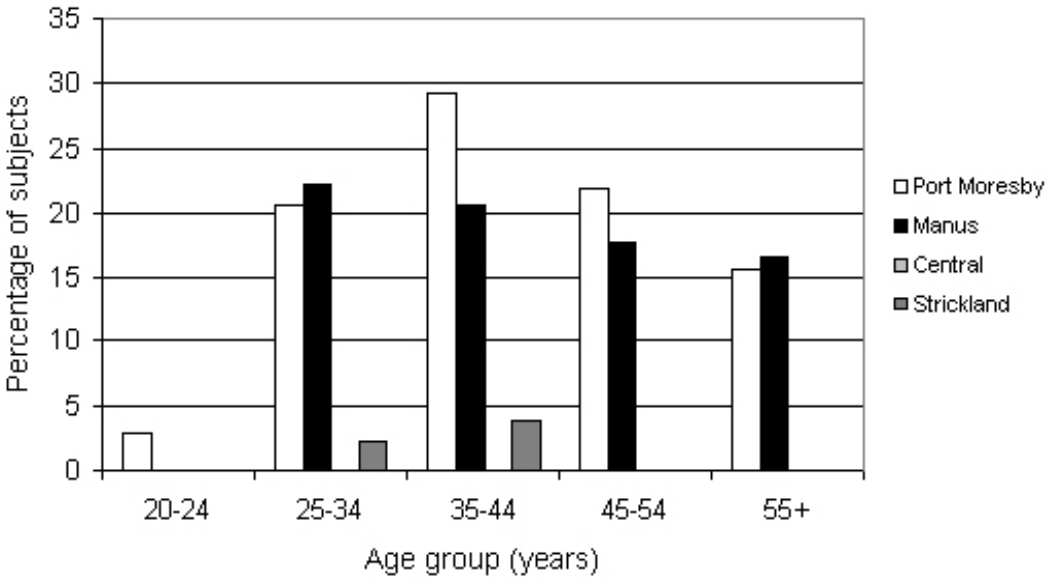

Figure 3. Prevalence of obesity by age group and study site.

sweet potato, taro, banana and yams are staple food and are supplemented by green leafy vegetables, river fish, pork and sometimes game (wildlife) meat and chicken. In rural Manus, reef fish and other seafood such as prawns and shellfish are the main source of protein supplementing their traditional diet.

Traditional Papua New Guinean foods are bulky and most Papua New Guineans from childhood are used to eating large volumes of food; when this habit is continued in the urban setting with western-type food, their fat, carbohydrate, protein and salt intake will increase significantly, leading to obesity.

Urban diet

In Port Moresby, many Papua New Guineans eat refined carbohydrates, such as rice, bread and scones, and a variety of meat products including lamb flaps. Food choice leans more towards western-type food. Fast-food outlets are always busy selling fried takeaway food, a wide variety of soft drinks and sweets, which are favoured by a lot of people. Although there are healthy alternatives such as whole-grain carbohydrates (eg, oats), these are not easily affordable.

Although traditional types of food such as sweet potato, banana, taro, yams, cassava, sago and various green vegetables can be bought from the local markets in the city, many Papua New Guineans in the urban centres like to eat western-type food. Taufa and Benjamin found that in the dining hall of a mining company, the queue for the western food was very long while the local food queue was very short and at times non-existent (9).

Overweight and obesity

Obesity is more common in males and females in urban Port Moresby and the periurban population of Manus. In the Strickland area, only one male and female were obese. No case of obesity was recorded in rural Central (Table 3) (10).

Being overweight or obese increases with age to about 55 years and then begins to decline with advancing age. The high prevalence of overweight and obesity in Port Moresby and to some extent in Manus indicates the high intake of refined carbohydrates and fatty foods in these two populations (10).

The rural populations of Strickland and Central live on a traditional diet and in geographical locations where walking and physical activity are part of their daily routine. They are more physically active than the

subjects in Port Moresby and Manus.

Implications of obesity for Papua New Guineans in urban areas

Obesity has been shown to be associated with hypertension, ischaemic heart disease and diabetes (2). The high prevalence of overweight and obesity in Port Moresby and Manus corresponds well to the high prevalence of diabetes mellitus in these two study sites. Among the diabetes patients attending the diabetes clinic in Port Moresby, the majority (65%) of patients were overweight or obese at the time of diagnosis as is illustrated by their high body mass index (11).

Even highlanders, who are non-Austronesian and not at risk of diabetes in their traditional settings, are now developing diabetes in the urban centres (10). This shows the problems that Papua New Guineans living in urban centres are facing, with an increased likelihood of getting a non-communicable disease such as diabetes mellitus or cardiovascular disease.

Obesity has long been implicated as a very important risk factor in type 2 diabetes mellitus (12,13) because it can induce resistance to the action of insulin. It can do this by reducing the number of insulin receptors on the target cells and/or decreasing the glucose transport through post-receptor changes (14,15).

Type 2 diabetes is common among populations in the Pacific and Aboriginal Australians who are more obese and physically less active (12,16,17). In the Torres Strait Islanders, obesity has been shown to be associated with diabetes mellitus type 2 (18-20).

Dietary change: a balanced healthy diet

It is not easy to change people's eating habits. The urban and periurban communities need to be informed about the dangers of overeating and obesity. Over 90% of patients attending the diabetes clinic in Port Moresby had been living mainly on western, imported food and lacked physical activity (10).

Community awareness about a balanced healthy diet can be conducted in primary schools, higher institutions, church groups and the community. The school canteens and takeaway shops should be encouraged to sell healthy and affordable foods to school children. It is easier to educate the young school children about healthy balanced diets than adults, who have already established their eating habits.

Exercise and physical activity

Exercise and physical activity control body weight and prevent people with impaired glucose tolerance from developing diabetes mellitus (21-24) and reduce the risk of cardiovascular disease (2). The urban and periurban dwellers need to be encouraged to do some exercise such as walking and backyard gardening in order to maintain a healthy weight. In urban areas, the urban councils should build secure and safe exercise and walking tracks for its residents, especially women.

Conclusions

The BMI of urban and periurban dwellers is higher than in the remote rural areas of Papua New Guinea. There are more overweight and obese people in the urban communities and periurban villages. The lower level of normal BMI for Papua New Guineans should be 18.5 kg/m^2 rather than the WHO standard level of 20 kg/m^2.

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REFERENCES

1 **Craig PL, Swinburn BA, Matenga-Smith T, Matangi H, Vaughn G.** Do Polynesians still believe that big is beautiful? Comparison of body size perceptions and preferences of Cook Islanders, Maoris and Australians. *NZ Med J* 1996;109:200-203.

2 **Haslam DW, James WPT.** Obesity. *Lancet* 2005;366:1197-1209.

3 **World Health Organization.** Obesity: preventing and managing the global epidemic. Technical Report Series No 894. Geneva: World Health Organization, 2000.

4 **National Health and Medical Research Council.** Report of the 100th session of the National Health and Medical Research Council. Canberra: Australian Government Publishing Services, 1985.

5 **Garrow JS, Webster J.** Quetelet's index (W/H^2) as a measure of fatness. *Int J Obes* 1985;9:147-153.

6 **Rush EC, Plank LD, Laulu MS, Robinson SM.** Prediction of percentage body fat from anthropometric measurements: comparison of New Zealand European and Polynesian young women. *Am J Clin Nutr* 1997;66:2-7.

7 **Royal College of Physicians.** Report of a Working Party on Obesity. Royal College of Physicians, London, 1983.

8 **Lindeberg S, Berntorpe E, Nilsson-Ehle P, Terént A, Vessby B.** Age relations of cardiovascular risk factors in a traditional Melanesian society: the Kitava Study. *Am J Clin Nutr* 1997;66:845-852.

9 **Taufa T, Benjamin AL.** Diabetes: the by-product of westernization in Papua New Guinea. *PNG Med J* 2001;44:108-110.

10 **Benjamin AL.** Community screening for diabetes mellitus in selected urban and rural areas of Papua New Guinea and determination of associated risk factors. MD Thesis, University of Papua New Guinea, Port Moresby, 2004.

11 **Benjamin AL, Margis D.** Betelnut chewing: a contributing factor in the poor glycaemic control in diabetic patients attending Port Moresby General Hospital, Papua New Guinea. *PNG Med J* 2005;48:174-182.

12 **Dowse GK, Zimmet PZ, Gareeboo H, Alberti KGMM, Tuomilehto J, Finch CF, Chitson P, Tulsidas H.** Abdominal obesity and physical inactivity as risk factors for NIDDM and impaired glucose tolerance in Indian Creole and Chinese Mauritians. *Diabetes Care* 1991;14:271-282.

13 **Zimmet PZ.** Challenges in diabetes epidemiology – from West to the rest. *Diabetes Care* 1992;15:232-252.

14 **World Health Organization.** Prevention of diabetes mellitus. Report of a WHO Study Group. WHO Technical Report Series No 844. Geneva: World Health Organization, 1985.

15 **Freeman WL.** The dietary management of non-insulin-dependent diabetes mellitus in the obese patient. *Prim Care* 1988;15:327-352.

16 **Eason RJ, Pada J, Wallace R, Henry A, Thornton R.** Changing patterns of hypertension, diabetes, obesity and diet among Melanesians and Micronesians in the Solomon Islands. *Med J Aust* 1987;146:465-469,473.

17 **McDermott R, Rowley KG, Lee AJ, Knight S, O'Dea K.** Increase in prevalence of obesity and diabetes and decrease in plasma cholesterol in a central Australian Aboriginal community. *Med J Aust* 2000;172:480-484.

18 **Guest CS.** Diabetes in Aborigines and Torres Strait Islanders: effects, causes and strategies. *Aust NZ J Public Health* 1995;19:442-444.

19 **O'Dea K, Rowley KG.** Macrovascular disease risk factors and insulin resistance in Aboriginal and Torres Strait Islander people. *J. Diabetes Complications* 2002;16:9-16.

20 **Leonard D, McDermott R, O'Dea K, Rowley KG, Pensio P, Sambo E, Twist A, Toolis R, Lowson S, Best JD.** Obesity, diabetes and associated cardiovascular risk factors among Torres Strait Islander people. *Aust NZ J Public Health* 2002;26:144-149.

21 **Reitman JS, Vasquez B, Klimes I, Nagulesparan M.** Improvement of glucose homeostasis after exercise training in non-insulin-dependent diabetes. *Diabetes Care* 1984;7:434-441.

22 **Helmrich SP, Ragland DR, Leung RW, Paffenbarger RS Jr.** Physical activity and reduced occurrence of non-insulin-dependent diabetes mellitus. *N Engl J Med* 1991;325:147-152.

23 **Manson JE, Rimm EB, Stampfer MJ, Colditz GA, Willett WC, Krolewski AS, Rosner B, Hennekens CH, Speizer FE.** Physical activity and incidence of non-insulin-dependent diabetes mellitus in women. *Lancet* 1991;338:774-778.

24 **Pan XR, Li GW, Hu YH, Wang JX, Yang WY, An ZX, Hu ZX, Lin J, Xiao JZ, Cao HB, Liu PA, Jiang XG, Jiang YY, Wang JP, Zheng H, Zhang H, Bennett PH, Howard BV.** Effects of diet and exercise in preventing NIDDM in people with impaired glucose tolerance. The Da Qing IGT and Diabetes Study. *Diabetes Care* 1997;20:537-544.

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Scrub typhus (*Orientia tsutsugamushi*), spotted fever (*Rickettsia australis*) and dengue fever as possible causes of mysterious deaths in the Strickland Gorge area of Southern Highlands and West Sepik Provinces of Papua New Guinea

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SUMMARY

A medical investigation was carried out in April 2001 into an outbreak of a mysterious haemorrhagic disease and deaths in the remote picturesque Strickland River area of Papua New Guinea (PNG). The area is in part of the Southern Highlands and West Sepik Provinces and situated downstream from the Porgera Joint Venture gold mine. 9 villages were visited and 140 persons, consisting of immediate blood relatives of the deceased (cases) and others in the village picked at random (controls), were physically examined. Specimens of blood, urine and faeces were collected from each person for laboratory tests in PNG and Australia. Positive sera for dengue (15%) and Japanese encephalitis (JE) (6%) were identified. Surprisingly, a number of the sera were positive for scrub typhus (*Orientia tsutsugamushi*) (28%) and spotted fever (*Rickettsia australis*) (11%). The last reported cases of scrub typhus in PNG were during World War Two among the allied troops. This is the first time spotted fever (*R. australis*) has been reported in PNG. These conditions may have been the cause of the deaths described by the villagers. However, there were significantly more dengue-positive results among relatives of the deceased than non-relatives though no such difference was found with rickettsial infections: haemorrhagic dengue fever is thus the most likely cause of this recurring outbreak. Mining did not appear to be a direct causal factor for the deaths in the area.

Introduction

The aim of the survey was to determine the epidemiology and possible causes of unaccountable deaths that occurred over a ten-year period in parts of the Strickland Gorge area of Southern Highlands and West Sepik Provinces of Papua New Guinea (PNG).

In 1998 Robinson and Haley (1), who were doing social and genealogical studies in the area, first reported 42 deaths. This new condition was thought to have started ten years earlier. The signs and symptoms of the disease, as described by the villagers, varied and included, among the various cases, a distended abdomen, a distended abdomen with swelling of the arms, legs and face, intermittent fever, severe jaundice and red eyes. Some described a body rash that involved the soles of the feet. Most descriptions included bleeding from nose, mouth and ears. Deaths occurred within 24-48 hours of the start of symptoms. This presentation could easily fit several infectious diseases or syndromes that have been reported in PNG and other parts of the world.

All age groups were affected. The villages with most cases were Bulago, Egali and Magali and it was these villages that

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recommended that Porgera Joint Venture (PJV) send a medical patrol to investigate this new haemorrhagic type of disease. It was Saleu (2), in his report presented at the 2000 Annual PNG Medical Symposium, who raised the public, media and National Parliament debates on this issue (Figure 1).

In October 2000 an independent medical team was set up to investigate the cause of these deaths. This survey was sponsored by the PNG Government Department of Environment with PJV providing logistical assistance.

Methods

Selection of villages and subjects

Because of many unknown variables, cost and time factors, it was decided it would be more appropriate to visit villages where deaths had been reported and to also include one control village, Biamin, where no cases had been reported. In each affected village identified, one first-degree relative of every deceased person was included, eg, a son, father, sister etc. This method is fraught with statistical problems as it presumes that the condition is either genetically influenced or transmitted by close contact, eg, typhoid infection. 10 other persons were selected at random as controls and similar examinations and specimens collected from each.

Conditions considered and screened

The following conditions were specifically targeted clinically and in specimens collected to confirm current or past infections: dengue and dengue haemorrhagic fever, rickettsial diseases (scrub typhus and spotted fever), typhoid, malaria, tuberculosis (TB), *Taenia solium* cysticercosis and HIV/AIDS (human immunodeficiency virus/acquired immune deficiency syndrome) . After the results were available, Ebola virus antibodies were requested for 18 specimens chosen at random from relatives of the deceased and others from the nine villages.

Drinking water samples were collected and tested for coliform counts and heavy metal levels. Urine samples were collected from study persons and checked for protein and sugar and cultured for pathogenic organisms. Faecal samples were also collected from the study persons for examination and culture.

Serological tests were performed for flaviviruses and the rickettsia groups. Samples were screened by flavivirus

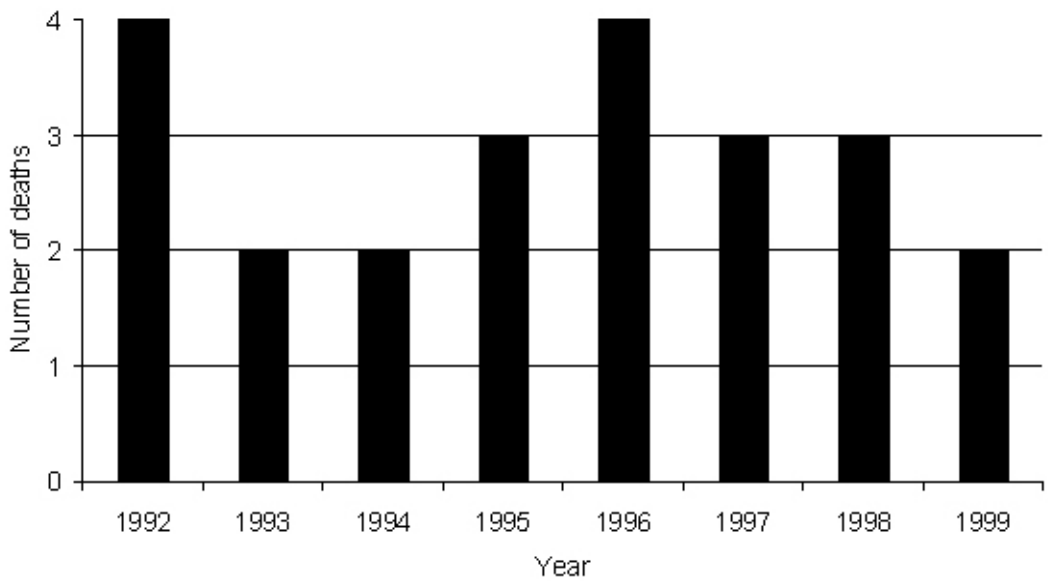

Figure 1. Haemorrhagic deaths in the Strickland Gorge area, Papua New Guinea, by year (23 deaths). This is based on the field patrol report compiled by Gerard Saleu in January 2000 and presented to Porgera Joint Venture (2) and at the Annual Symposium of the Medical Society of Papua New Guinea in September 2000 in Port Moresby.

competitive ELISAs using flavivirus-reactive and Japanese encephalitis (JE)-specific assays. The positive sera in the JE assay were further analyzed by Murray Valley encephalitis virus (MVEV)-specific and Kunjin virus (KUNV)-specific competitive assays as per Hall et al. (3) and by micro-neutralization against JE. This is the same method as used in the study of JE antibodies in a group of people in Indonesian Papua (4).

Samples for the rickettsia groups were screened by two assays: Panbio Rickettsia spotted fever group IgM ELISA test for IgM antibodies to *Rickettsia australis, R. honei, R. rickettsii, R. conorii, R. siberica* and *R. akari*, and Panbio Rickettsia scrub typhus group IgM ELISA test for IgM antibodies to *Orientia tsutsugamushi* (group r56 antigen). These are both screening assays. All reactive samples were sent to the Reference Laboratory at Queensland Health Scientific Services for confirmation by immunofluorescence assay (IFA).

The number and types of village houses, ventilation, cooking places, toilets and water supply were checked. Approximately 50 ml of drinking water was collected in sterile bottles from each participating village for laboratory bacterial and heavy metal testing. Samples of water were also taken from the Ok Om (Om river) and the Lagaip and Strickland rivers.

Ethical clearance

Ethical clearance was obtained from the PNG Government Medical Research Advisory Committee, Port Moresby and the James Cook University Ethical Committee, Townsville.

Results

From villagers' reports and focus group discussions the number of deaths appeared much larger than originally reported by Saleu (2) and Robinson and Haley (1). Unfortunately there is no independent verification of these reports. One has to consider other normal causes of death in a community with high mortality, low literacy rate and no compulsory registration of births and deaths. Haemorrhagic deaths, as reported by the villagers to the team, peaked in 1993 then gradually declined (Figure 2). Some of the deaths in question appeared to occur before mining commenced in the area and generally affected males significantly more than females ($\chi^2 = 13.6$, $p < 0.001$) (Figures 2 and 3), although there were more female than male deaths in 2000 and 2001. It appears that the epidemic is still active (Figure 2), even though a case was not seen in the field.

Figure 2. Haemorrhagic deaths as reported by the villagers to the team, by year and gender (130 deaths).

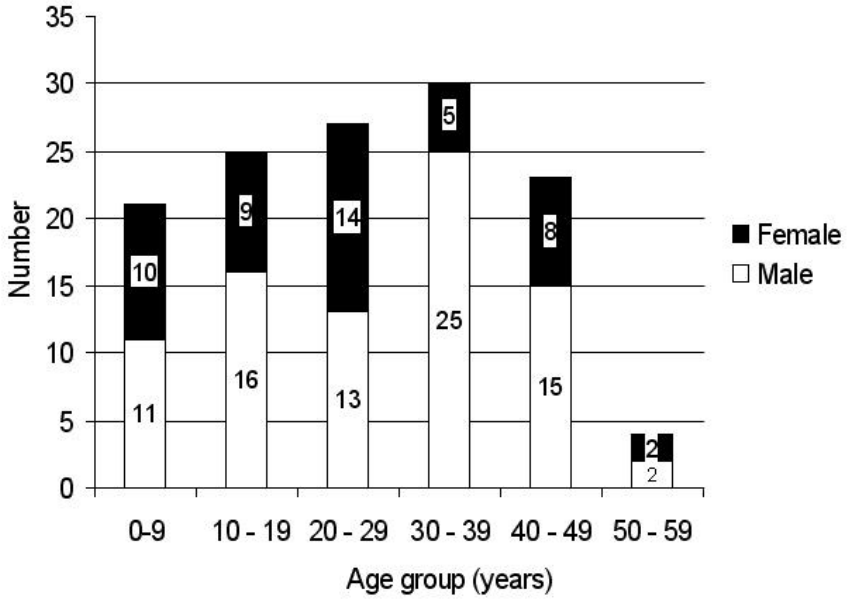

Figure 3. Haemorrhagic deaths as reported by the villagers to the team, by age and gender (130 deaths).

Water and heavy metal testing showed that none of the drinking sources or rivers in the area had levels of heavy metals above normal. Bacterial tests from sources of drinking water and the Strickland River, all except one, were heavily polluted with *E. coli* using the World Health Organization (WHO) accepted levels for *E. coli* as <10/100 ml of water. This confirmed the findings of Shearman (5).

Respiratory illnesses, including upper respiratory tract infections (URTI), pneumonia and chronic obstructive airway disease (COAD), were common, affecting 15% of those seen. Skin infections such as sores, scabies, tropical ulcers and tinea were the commonest of the medical conditions seen, affecting 33% of those in the study.

99% of 110 urine samples tested positive for protein with 36% having proteinuria of 100 mg/dl or above.

Faecal testing showed 25% of samples positive for parasites, *Entamoeba histolytica* being present in 65% of the positive samples. Helminth loads were low.

Blood samples were serologically tested for *Taenia solium* infection, a new condition introduced from neighbouring West Papua in the 1960s (6) and serologically confirmed for the first time in PNG in 1997 (7-9). There were two seropositive samples – a 70-year-old male and an 18-year-old male from different villages. Neither was related to a deceased person. The serological test at times has cross-reactivity to *Echinococcus* infections.

Malaria, a formidable problem in PNG, was assessed clinically, by blood slide examination and indirectly through the presence of splenic enlargement (10). There was a rough correlation between splenic enlargement and altitude (Table 1).

HIV/AIDS was introduced into PNG in 1987 and is increasing alarmingly (11); however, all 136 tests performed in this survey were serologically negative.

Serological testing for the Ebola/Marburg virus and leptospirosis was performed on 18 samples, 2 specimens from each village, one case and one control. All were negative.

Flaviviruses

The results of the screening tests for flavivirus and rickettsial infections are shown in Table 2 and Figures 4 and 5. There were 21 sera (15%) positive for dengue out of 136 samples and 8 (6%) positive for JE. The 8 samples positive in the JE assay were further analyzed by Murray Valley encephalitis virus (MVEV)-specific and Kunjin virus (KUNV)-

TABLE 1

MALARIA BLOOD SLIDE RESULTS, PALPABLE SPLEEN AND VILLAGE ALTITUDE

Village	Total	Blood slide		Palpable spleen (%)	Altitude* (feet asl)
		Positive (%)	Negative		
Egali**	12	-	-	7 (58.3)	2800
Bulago	14	2 (14.3)	12	5 (35.7)	4000
Bimin	18	0 (0)	18	0 (0)	5900
Kunanap	18	0 (0)	18	3 (16.7)	4000
Aluni	25	0 (0)	25	1 (4.0)	5200
Dupan	11	0 (0)	11	1 (9.1)	3500
Gawa	13	0 (0)	13	0 (0)	5200
Yokona	15	0 (0)	15	6 (40)	4500
Sisimin	9	0 (0)	9	0 (0)	1750
Total	135	2 (1.6)***	121	23 (17.0)	

*Generally there is a correlation between altitude and palpable spleen: places above 5000 feet above sea level had no palpable spleen – the exception is Aluni; at Sisimin, which is at 1750 feet, surprisingly no spleens were palpated
**Egali blood slides were wet and damaged in the ice-packs en route from the field to Porgera
***The percentage does not include the 12 from Egali

specific competitive assays, as previously done by Hall et al. (3) and Spicer et al. (4). One reacted in the KUNV assay but none in the MVEV assay. None of the 8 JE ELISA-reactive sera were confirmed by micro-neutralization against JEV. This suggests that none of the samples were positive for JEV-specific antibody. Based on similar results from a serological study in South Sulawesi during 1999 (Sjahril and Hall, unpublished data), the non-specific cross-reactions in the JEV competitive assay are likely to be due to skewed antibody responses after multiple infections with different dengue serotypes (R. Hall, personal communication).

Rickettsia groups

There were 140 samples of sera tested for scrub typhus, of which 39 (28%) were positive (Table 2). There were 16 (11%) positive sera for spotted fever out of the 140 samples. All villages demonstrated *O. tsutsugamushi* (scrub typhus) serum-positive findings including the control village of Bimin. Scrub typhus had a higher positive rate than spotted fever (Figure 5).

There was no significant difference in the rickettsial infections between males and females (Table 3 and Figure 6).

Confirmatory assays (IFA) for *Orientia tsutsugamushi* (scrub typhus) were the same as the IgM positive results with 39 (28%) positives out of 140. Similarly for *Rickettsia australis* (spotted fever) the results were essentially the same, with 15 (11%) positive out of 140.

Results in relatives of the deceased and controls

There were significantly more dengue

TABLE 2

POSITIVE SCREENING SEROLOGY TESTS FOR DENGUE, JAPANESE ENCEPHALITIS, SCRUB TYPHUS AND SPOTTED FEVER

Village	Dengue		Japanese encephalitis		Scrub typhus		Spotted fever	
	No	Positive (%)	No	Positive (%)	No	Positive (%)	No	Positive (%)
Egali	8	1 (12.5)	8	1 (12.5)	10	7 (70.0)	10	1 (10.0)
Bulago	11	1 (9.1)	11	0 (0)	11	3 (27.3)	11	0 (0)
Bimin	16	0 (0)	16	0 (0)	16	2 (12.5)	16	1 (6.3)
Kunanup	21	4 (19.0)	21	1 (4.8)	21	8 (38.1)	21	3 (14.3)
Aluni	25	5 (20.0)	25	2 (8.0)	25	3 (12.0)	25	4 (16.0)
Dupan	12	0 (0)	12	0 (0)	12	3 (25.0)	12	0 (0)
Gawa	13	0 (0)	13	0 (0)	13	2 (15.4)	13	1 (7.7)
Yokona	15	3 (20.0)	15	0 (0)	15	9 (60.0)	15	3 (20.0)
Sisimin	15	7 (46.7)	15	4 (26.7)	17	2 (11.8)	17	3 (17.6)
Total	136	21 (15.4)	136	8 (5.9)	140	39 (27.9)	140	16 (11.4)

Only one case of Kunjin/West Nile (KUN/WN) positive at Kunanap village
Confirmatory assays (IFA) for *Orientia tsutsugamushi* (scrub typhus) were the same as the IgM positive results with 39 positive out of 140 or 28%; similarly for *Rickettsia australis* (spotted fever) the results were essentially the same, with 15 positive out of 140 or 11%.

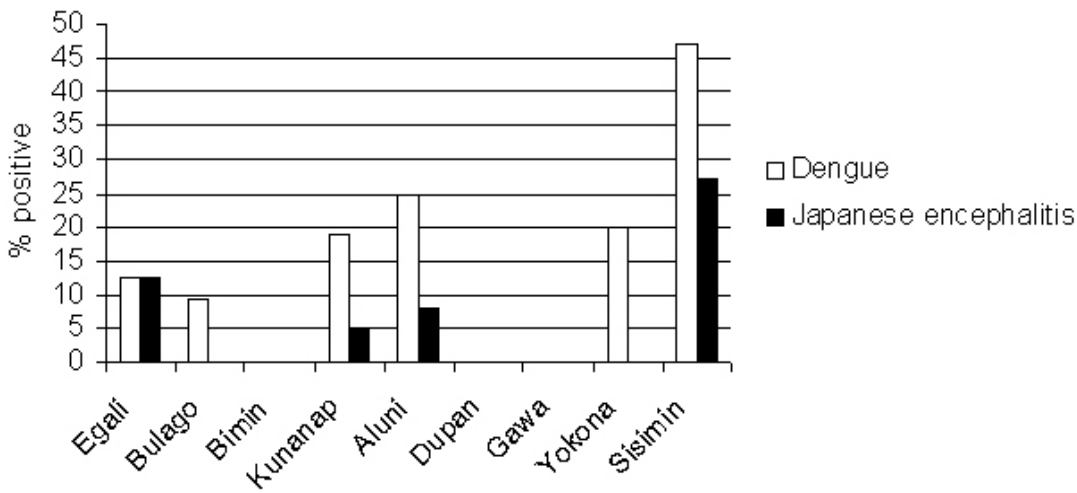

Figure 4. Dengue and Japanese encephalitis serological ELISA results by village. The percentage positive in the people tested in each village is plotted. The lowest altitude village, Sisimin, had lots of *Aedes aegypti* mosquitoes and this is reflected in it having the highest serum-positive rates.

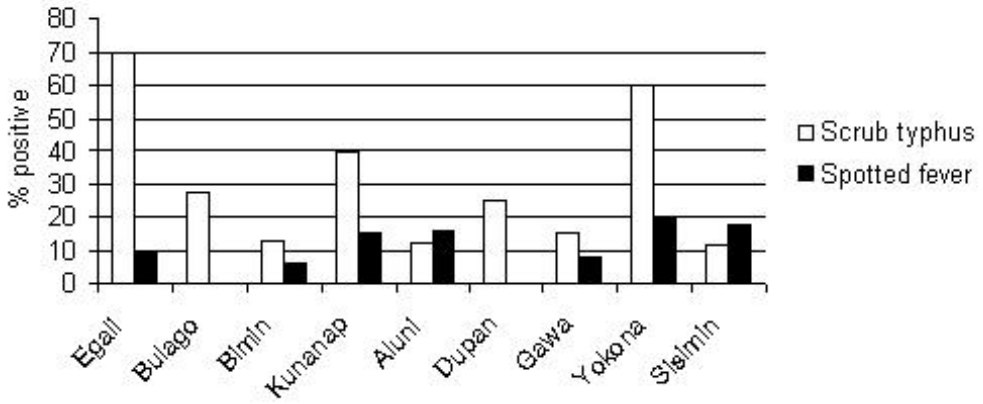

Figure 5. Scrub typhus and spotted fever serum-positive (IgM) results by village. All villages had *O. tsutsugamushi* (scrub typhus) serum-positive findings including the control village of Bimin. Scrub typhus had a higher positive rate than spotted fever.

TABLE 3

SCRUB TYPHUS SERUM-POSITIVE (IGM) RESULTS BY GENDER

	Scrub typhus positive	Scrub typhus negative
Male	24	56
Female	15	40
Total	39	96

χ^2 (Yates corrected) = 0.16, p = 0.95: there is no significant difference in scrub typhus positive between males and females

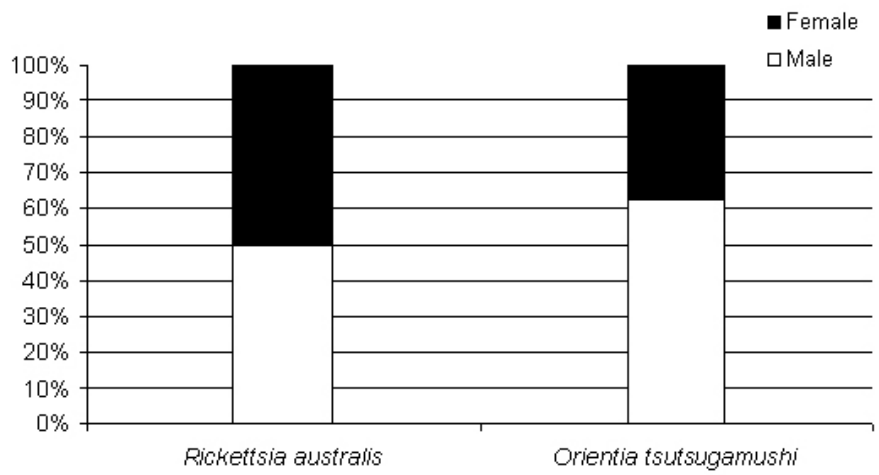

Figure 6. Rickettsial serum positives by gender. There is no significant difference in rickettsial infections between males and females (χ^2 = 1.6, df = 3, p = 0.762).

TABLE 4

DENGUE SERUM POSITIVES (ELISA) AND SCRUB TYPHUS SERUM POSITIVES (IGM) BY RELATIVES OF THE DECEASED AND OTHERS

	Dengue		Scrub typhus	
	Positive	**Negative**	**Positive**	**Negative**
Relatives of the deceased	11	25	8	23
Non-relatives	10	94	31	73
Total	21	119	39	96

Relatives of the deceased have significantly more dengue-positive serum results than non-relatives: dengue χ^2 (Yates correction) = 8.7, $p < 0.01$; for scrub typhus there is no significant difference in serum positives between relatives and non-relatives of the deceased: χ^2 (Yates correction) = 0.30, $p = 0.658$

serum-positive results among relatives of the deceased than non-relatives. With scrub typhus there was no significant difference in positive serum results between relatives and non-relatives of the deceased (Table 4).

Discussion

This retrospective investigation was made much more difficult because of the lack of definitive medical evidence and because of an illiterate community with no sense of time to help chronological recollections of events. The stories obtained from villagers during focus group discussions after the field clinics were at times conflicting.

The logistics of keeping the collected specimens cool and in a suitable condition for analysis by the various laboratories in PNG and Australia proved difficult, but not insurmountable, and was achieved by making use of iceboxes and frequent helicopter trips provided by PJV.

There have been previous epidemics of dengue in the country (12-14) and the latest recorded was in 1991. These have been identified as dengue serotypes 1, 2 and 3 but not 4 (15). No cases of dengue haemorrhagic fever (DHF) have been described in PNG but it is common in other parts of the Pacific and South-East Asia. Dengue serotypes 2 and 3 are associated with haemorrhagic fever and these are the types recorded in PNG. The signs and symptoms, as described by local villagers, fit DHF and the more acute dengue shock syndrome (DSS). It is likely the villagers may have experienced an outbreak of DHF and DSS and that there was a dengue epidemic in the area without the knowledge of the health authorities (Tables 2 and 4, Figure 4).

There have been outbreaks of JE in the Western and Milne Bay Provinces in the 1990s (J. Wangi, personal communication, 2000), in neighbouring West Papua (4,16) and Torres Strait Islands (17). The JE picture does not appear appropriate here but it is interesting to note that it has now spread to the highlands of PNG. The vector *Aedes aegypti* thrives in the PNG lowlands and predictably Sisimin, the village at the lowest altitude, has the highest serum positives for the flaviviruses. There were no positives among the people living in the high altitude villages of Bimin, Dupan and Gawa (Figure 4).

There are two groups within the rickettsiae. The scrub typhus group (STG), formerly *Rickettsia tsutsugamushi*, is now classified as *Orientia tsutsugamushi*. It is found in the Orient including northern Australia and PNG. The second group is divided into two subgroups, the spotted fever group (SFG) and the typhus group (TG).

The scrub typhus illness is caused by *Orientia tsutsugamushi*. Clinical features of the haemorrhagic disease described are similar to those found in studies in Japan (18), in Taiwan (19) and in Cambodia (20). Ticks and mites are the vectors for rickettsial infections and normally live on animals,

particularly rodents that inhabit the grasslands. Humans are accidental hosts. Scrub typhus is spread by mites whilst ticks generally transmit spotted fever infections. Ticks and mites are widely distributed in the Americas, Africa, Europe, Asia and North Australia. The lush 'kunai' grasslands of the magnificent Strickland Gorge and particularly Kunanup village are the ideal habitat for the rodents and the vector for rickettsial infections. The scrub typhus mite cycle in coastal PNG was studied by the Americans during World War Two (21,22). The spotted fever tick or mite cycle in PNG is unknown and urgently needs investigation in the light of this outbreak. The presence of IgM antibody is an indication of recent or active scrub typhus disease. This is supported by the epidemiological distribution of the cases (Figure 3) and the reports to the visiting follow-up team of 6 more new deaths since this survey was completed.

The finding of positive serological tests to the rickettsiae (Table 2) was a surprise since there have been no cases of scrub typhus (*Orientia tsutsugamushi*) reported in PNG since the end of the Second World War (21). There was no significant difference in scrub typhus positives between males and females or between relatives of the deceased and others (Tables 3 and 4). This is also the first time that spotted fever (*Rickettsia australis*) has been reported in PNG. The nearest recorded cases since World War Two have been in the Eastern Solomon Islands and Vanuatu (23) and in Biak in West Papua (24). During World War Two scrub typhus caused havoc among allied troops and carried a case fatality rate of 20% (25). Having a scrub typhus infection resulted in evacuation of a soldier from the front line. The Army also abandoned the island of Bat in Manus Province, PNG because of the high rate of scrub typhus infections among soldiers (26). According to Chin (27) the fatality rate in untreated cases can be as high as 60%. Professor Ian Maddocks (personal communication), a longtime resident physician in PNG, suggested that the local coastal people have developed an immunity to the disease and only people from non-endemic areas, such as the highlanders, would be affected. He had seen one highlander with scrub typhus. The allied troops would have fallen into this non-immune category. This theory is supported by a recent report on a serological survey by Kende and Graves (28) in 2003 of the PNG highlands and coastal communities. It shows that rickettsial serum tests were positive among the coastal people of Central Province and not in the highlanders. The following questions then arise:

- Why might there have been an epidemic of scrub typhus in the Strickland high country? Could it have been recently introduced from the coastal areas and caused infection in a non-immune group? The fact that health workers have not reported it before supports this view. However, the area is very remote and inaccessible and this could account for poor reporting.

- Did it exist before, as reported by some at Bulago and Aluni villages, and no notice was taken until the mining started? In support of this view Saleu (2) reported a Japanese anthropology student who stated that 10 people had died of scrub typhus in the Firamin and Telefomin areas within the highlands but away from the Strickland Gorge region. The clinical picture of these rickettsial diseases (27) if not treated is similar to the description of the mysterious deaths. Videotape pictures of a maculopapular rash on the extremities, including the soles of the feet, of a person from the Strickland River area was reported as "dying of chemical poisoning" by Sopas Hospital staff in 1995 (29). This fits very well the clinical picture of rickettsial spotted fever infection. Unfortunately no serological tests for rickettsial infections were performed.

It is interesting to note that there is serological evidence for the continued presence of rickettsiosis in Southern India. It was generally believed to have disappeared from there. However, serological tests confirmed that spotted fever, epidemic typhus, endemic typhus and scrub typhus continue to occur in many parts of India (30) .

During the last six years it appears that rickettsial diseases have become more prevalent, particularly in the South-East Asia region. Studies have noted the emergence of rickettsiosis in the Myanmar-Thailand region (31,32), in Laos (33), in South Korea (34,35), in Sri Lanka (36), and in Japan and

the Far East (37). These results may be, however, a reflection of more awareness of rickettsial diseases. Improved serological techniques for detection may also be responsible for the apparent increase in recorded cases. Jensenius and colleagues warn that rickettsiosis must now be a consideration for the international traveller who presents with a benign febrile illness accompanied by headache, myalgia and cutaneous eruptions. Severe complications and death are occasionally seen (38).

Conclusion

The survey and tests have excluded malaria, typhoid, tuberculosis, *Taenia solium*, HIV/AIDS and Ebola/Marburg virus as likely causes of the haemorrhagic fever deaths. Water tests for bacteriology and heavy metals also exclude them as likely causes. The only person to have survived from the disease had negative leptospirosis serology and his blood heavy metal results were all well below accepted cut-off points.

The serological findings of positive antibodies to flaviviruses and rickettsiae indicate that there was an epidemic of these infections in the area, of which health authorities were unaware. It appears that the haemorrhagic disease problem is an ongoing issue (Figure 2). This is reinforced by the fact that a further 6 deaths have been reported after this survey was concluded. The follow-up team explained the findings to the villagers, discussed preventive measures and left supplies of doxycycline and chloramphenicol in each village. It is essential that these drugs are readily available within the heath services and are constantly made accessible to the villagers. The signs, symptoms and mortality of the condition as described by the villagers strongly suggest dengue haemorrhagic fever or infection with *Orientia tsutsugamushi* (scrub typhus) or *Rickettsia australis* (spotted fever).

The finding of evidence for these infections in these communities is of considerable importance. However, the only difference found between the results in relatives of the diseased and non-relatives was in the dengue serology (Table 4). This makes dengue haemorrhagic fever the most likely cause of the haemorrhagic disease and deaths that occurred in this mysterious outbreak. The present expansion of dengue infections in the Pacific region and the apparent continuation of the epidemic in the Strickland Gorge are consistent with this explanation.

The villages, except for Sisimin, are high in the mountains. The villagers descend to the lower Strickland River areas to hunt. Here it is open grassland with wild game available. One theory is that the El Niño dry spells of the 1990s forced more wild game towards the Strickland River. It is likely that during hunting trips the hunters are, in their turn, hunted by the vector mosquitoes that transmit the flaviviruses and the mites and ticks that transmit rickettsial infections.

Recommendations

- Findings to be made known to the villagers in order that they may take appropriate preventive measures. The villagers should be made aware that it is almost certainly not the water from the Strickland River that caused the deaths.
- A medical and entomology team to be given the task of establishing the exact life cycle of the agents of scrub typhus and spotted fever in the Strickland areas.
- The distribution and use of chemically impregnated nets plus education regarding the life cycle of day-biting *Aedes aegypti* mosquitoes to prevent them from biting and thus transmitting dengue and other flaviviruses.
- Avoidance of scrub typhus areas.

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- Dr Mike Catton and Dr Julian Druce at the Commonwealth Haemorrhagic Fever Reference Laboratory, Melbourne, Victoria for performing the Ebola/Marburg viral tests.

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REFERENCES

1 **Robinson R, Haley N.** A social and genealogical study of the Bogaia, Yeru, Duna and Hewa people of the Lagaip and Upper Strickland rivers. Report to Porgera Joint Venture, Mt Hagen, 1998.

2 **Saleu G.** Community relations patrol, Lake Kopiago District, Southern Highlands and Oksapmin District, West Sepik Province. Report to Porgera Joint Venture, Mt Hagen, 2000.

3 **Hall RA, Broom AK, Hartnett AC, Howard MJ, Mackenzie JS.** Immunodominant epitopes on the NS1 protein of MVE and KUN viruses serve as targets for a blocking ELISA to detect virus-specific antibodies in sentinel animal serum. *J Virol Methods* 1995;51:201-210.

4 **Spicer PE, Phillips D, Pike A, Johansen C, Melrose W, Hall RA.** Antibodies to Japanese encephalitis virus in human sera collected from Irian Jaya. Follow-up of a previously reported case of Japanese encephalitis in that region. *Trans R Soc Trop Med Hyg* 1999;93:511-514.

5 **Shearman P.** The environmental and social impact of the Porgera Gold Mine on the Strickland River system. BSc Hons Thesis, University of Tasmania, 1995.

6 **Gajdusek DC.** Introduction of *Taenia solium* into West New Guinea with a note on an epidemic of burns from cysticercus epilepsy in the Ekari people of the Wissel Lakes area. *PNG Med J* 1978:21:329-342.

7 **Taufa T.** Human health. CSIRO review of the riverine impacts, Porgera Joint Venture. Commonwealth Scientific and Industrial Research Organization, Canberra, Australia, 1996.

8 **Taufa T.** Baseline health survey in parts of Lagaip and Strickland Rivers and the Lake Murray areas, January, June and November 1996. Unisearch Report. University of Papua New Guinea, Port Moresby, 1997.

9 **Taufa T.** Baseline health survey of the lower Ok Tedi area for the OTML waste management. Unpublished report of the Community Medicine Department, Faculty of Medicine, University of Papua New Guinea, Port Moresby, 1997.

10 **Hackett LW.** Spleen enlargement in malaria. *J Natl Malar Soc* 1943;3:121-133.

11 **Malau C.** HIV/AIDS toll to soar. *Post-Courier*, 18 Feb 2002:5.

12 **Papua New Guinea Department of Health.** Papua New Guinea National Health Plan 1991-1995. Port Moresby: Department of Health, Jun 1990.

13 **Papua New Guinea Department of Health.** Papua New Guinea National Health Plan 2001- 2010. Port Moresby: Department of Health, Aug 2000.

14 **Farrell KT.** An epidemic of dengue fever in Wewak. *PNG Med J* 1978:21:191-196.

15 **Trevett AJ, Sanders RC.** Arbovirus disease in Papua New Guinea. *PNG Med J* 1994:37:116-124.

16 **Spicer PE.** Japanese encephalitis in Western Irian Jaya. *J Travel Med* 1997;4:146-147.

17 **Hanna JN, Ritchie SA, Phillips DA, Shield J, Bailey MC, Mackenzie JS, Poidinger M, McCall BJ, Mills PJ.** An outbreak of Japanese encephalitis in the Torres Strait, Australia 1995. *Med J Aust* 1996;165:256-260.

18 **Ogawa M, Hagiwara T, Kishimoto T, Shiga S, Yoshida Y, Furuya Y, Kaiho I, Ito T, Nemoto H, Yamamoto N, Masukawa K.** Tsutsugamushi disease (scrub typhus) in Japan: clinical features. [Jp] *Kansenshogaku Zasshi* 2001;75:359-364.

19 **Lee HC, Ko WC, Lee HL, Chen HY.** Clinical manifestations and complications of rickettsiosis in southern Taiwan. *J Formos Med Assoc* 2002;101:385-392.

20 **Thap LC, Supanaranond W, Treeprasertsuk S, Kitvatanachai S, Chinprasatsak S, Phonrat B.** Septic shock secondary to scrub typhus: characteristics and complications. *Southeast Asian J Trop Med Public Health* 2002;33:780-786.

21 **Blake FG, Maxcy KF, Sadusk JF, Kohls GM, Bell EJ.** Studies on tsutsugamushi disease (scrub typhus, mite-borne typhus) in New Guinea and adjacent islands: epidemiology, clinical observations, and aetiology in Dobanura area. *Am J Hyg* 1945;41:243-373.

22 **Kohls GM, Armbrust CA, Irons EN, Philip CB.** Studies on tsutsugamushi disease (scrub typhus, mite-borne typhus) in New Guinea and adjacent islands. *Am J Hyg* 1945;41:374-399.

23 **Miles JA, Austin FJ, Jennings IG.** Scrub typhus

in the Eastern Solomon Islands and Northern Vanuatu (New Hebrides). *Am J Trop Med Hyg* 1981;30:849-854.

24 **Hadi TR, Nalim S, Sukaeri S, Dennis DT.** Scrub typhus survey of Biak and Owi islands: ectoparasites of small mammals and rickettsial isolations. *Southeast Asian J Trop Med Public Health* 1980;1:220-226.

25 **Williams SW, Sinclair AJM, Jackson AV.** Mite-borne typhus in Papua and the Mandated Territory of New Guinea. *Med J Aust* 1944;2:525-539.

26 **Philip CB, Kohls GM.** Studies on tsutsugamushi disease (scrub typhus, mite-borne typhus) in New Guinea and adjacent islands. *Am J Hyg* 1945;42:195-203.

27 **Chin J.** Control of Communicable Diseases Manual, 17th edition. Washington DC: American Public Health Association, 2000.

28 **Kende M, Graves S.** Survey of rickettsial antibodies at two local sites and review of rickettsiosis in Papua New Guinea. *PNG Med J* 2003;46:53-62.

29 **Special Broadcasting Service.** SBS Television Special Report Videotape, 1995.

30 **Mathai E, Lloyd G, Cherian T, Abraham OC, Cherian AM.** Serological evidence for the continued presence of human rickettsiosis in southern India. *Ann Trop Med Parasitol* 2001;95:395-398.

31 **Parola P, Miller RS, McDaniel P, Telford SR 3rd, Rolain JM, Wongsrichanalai C, Raoult D.** Emerging rickettsiosis of the Thai-Myanmar border. *Emerg Infect Dis* 2003;9:592-595.

32 **Pickard AL, McDaniel P, Miller RS, Uthaimongkol N, Buathong N, Murray CK, Telford SR 3rd, Parola P, Wongsrichanalai C.** A study of febrile illnesses on the Thai-Myanmar border: predictive factors of rickettsiosis. *Southeast Asian J Trop Med Public Health* 2004;35:657-663.

33 **Phongmany S, Rolain JM, Phetsouvanh R, Blacksell SD, Soukkhaseum V, Rasachack B, Phiasakha K, Soukkhaseum S, Frichithavong K, Chu V, Keolouangkhot V, Martinez-Aussel B, Chang K, Darasavath C, Rattanavong O, Sisouphone S, Mayxay M, Vidamaly S, Parola P, Thammavong C, Heuangvongsy M, Syhavong B, Raoult D, White NJ, Newton PN.** Rickettsial infections and fever, Vientiane, Laos. *Emerg Infect Dis* 2006;12:256-262.

34 **Jang WJ, Kim JH, Choi YJ, Jung KD, Kim YG, Lee SH, Choi MS, Kim IS, Walker DH, Park KH.** First serological evidence of human spotted fever group rickettsiosis in Korea. *J Clin Microbiol* 2004;42:2310-2313.

35 **Choi YJ, Jang WJ, Ryu JS, Lee SH, Park KH, Paik HS, Koh YS, Choi MS, Kim IS.** Spotted fever group and typhus group rickettsiosis in humans, South Korea. *Emerg Infect Dis* 2005;11:237-244.

36 **Kularatne SA, Edirisingha JS, Gawarammana IB, Urakami H, Chenchittikul M, Kaiho I.** Emerging rickettsial infections in Sri Lanka: the pattern in the hilly Central Province. *Trop Med Int Health* 2003;8:803-811.

37 **Mahara F.** Rickettsiosis in Japan and the Far East. *Ann NY Acad Sci* 2006;1078:60-73.

38 **Jensenius M, Fournier PE, Raoult D.** Rickettsiosis and the international traveler. *Clin Infect Dis* 2004;39:1493-1499.

List of Medical Research Projects in Papua New Guinea

Approved or Noted

By the Medical Research Advisory Committee in 2006

Malarial anaemia following drug treatment of people in endemic areas

Dr Louis Schofield, Dr Harin Karunajeewa and Dr Ivo Mueller (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province 511, Papua New Guinea)

Updating filarial antigen tests

Dr Wayne Melrose, Dr Billy Selve, Dr Jethro Usurup and Dr Jeffrey Warner (WHO Collaborating Centre for Control of Lymphatic Filariasis, James Cook University, Townsville, Queensland 4811, Australia)

Aetiology of opportunistic intestinal parasitic infections among HIV infected patients at Goroka Base Hospital

Dr Peter Siba, Dr Paul Harino and Dr Suparat Phuanukoonnon (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Evaluation of a program to improve the interaction between community and health systems in Papua New Guinea: its impact on improved maternal and child health

Ms Helen Ashwell and Mrs Doreen Dawadawareta (PO Box 740, Redcliffe, Queensland 4020, Australia)

Monitoring and evaluation of sentinel sites for National Lymphatic Filariasis Control Program

Dr Wayne Melrose, Mr Leo Makita and Dr Ishmael Robert (WHO Collaborating Centre for Control of Lymphatic Filariasis, James Cook University, Townsville, Queensland 4811, Australia)

An ethnicity-based analysis of genetic polymorphism and susceptibility to infectious diseases

Dr Peter Zimmerman and Dr John Reeder (Centre for Global Health and Diseases, Case Western University School of Medicine, W147D, Cleveland, Ohio 44106-4983, USA)

Investigating the benefits and costs of an integrated malaria control program in a large oil palm plantation in Papua New Guinea

Dr Ivo Mueller and Ms Bianca Plüss (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province 511, Papua New Guinea)

Immune maturation and function in newborns in Papua New Guinea and Australia

Dr Suparat Phuanukoonnon and Dr Anita van den Biggelaar (Telethon Institute for Child Health Research, PO Box 855, West Perth, WA 6872, Australia)

Mass drug treatment and vector control of filariasis, phase 1 (pre-intervention observational): demography, disease, and transmission survey of lymphatic filariasis in Papua New Guinea

Professor James Kazura, Associate Professor Peter Siba, Professor John Reeder and Dr Moses Bockarie (Case Western University, Centre for Global Health and Diseases, Wolstein Research Bldg, 10900 Euclid Avenue, Cleveland, OH 44106-7286, USA)

Documentary film about life and work of Carleton Gajdusek

Mr Bosse Lindquist (Swedish Television, Documentary Department, NYH 60E, SE 105 10 Stockholm, Sweden)

Impacts of cross-cultural interaction and international migration on a group of Papua New Guineans adopted by Dr Carleton Gajdusek

Dr Ceridwen Spark (School of Political and Social Enquiry, Faculty of Arts, Monash University, Victoria 3800, Australia)

Investigation of dengue viral infection in Papua New Guinea

Mr Dagwin Suarkia, Associate Professor Peter Siba and Mr Leo Makita (Papua New Guinea Institute of Medical Research, PO

Box 60, Goroka, EHP 441, Papua New Guinea)

Enhancing pregnancy outcomes in Papua New Guinea

Associate Professor Peter Siba, Dr A. Abramov, Dr Grace Kariwiga, Dr A. B. Amoa, Dr Ivo Mueller and Professor John C. Reeder (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Better use of public health campaigns for child survival

Dr Chris Morgan, Associate Professor Peter Siba and Ms Rachael Hinton (Burnet Institute, Centre for International Health, GPO Box 2284, Melbourne, Victoria 3001, Australia)

Study of health seeking behaviour, non-compliance and role of social capital in TB program: implication for TB sufferers and home care providers in selected areas of PNG

Dr Peter Siba, Ms Geraldine Maibani, Dr Katie Thomas, Dr Paison Dakulala and Dr Maxine Whittaker (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Evaluation of community based health promotion program and assessment of program sustainability in the Eastern Highlands of Papua New Guinea

Ms Geraldine Maibani (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Health seeking behaviour for chronic cough and tuberculosis in selected sites: a gendered analysis

Ms Geraldine Maibani (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Oral health survey of Papua New Guinea 2006

Dr Emily Wesley (Oral Health Services, Department of Health, PO Box 807, Waigani, NCD 131, Papua New Guinea)

Assessment of the efficacy and safety of amodiaquine plus sulphadoxine-pyrimethamine (S-P) combination and sulphadoxine-pyrimethamine alone as intermittent preventive treatment for malaria in pregnancy

Dr Francis Hombhanje, Dr John Sairere, Dr William Selve, Dr Ilomo Hwaihwanje and Dr Steven Toraso (Divine Word University, School of Health Sciences, PO Box 483, Madang, Madang Province 511, Papua New Guinea)

An economic evaluation of health initiatives funded by private enterprise within a developing economy

Dr Christian Wium (Lihir Gold Mine, Port Moresby Office, PO Box 789, Port Moresby, NCD 121, Papua New Guinea)

Genetic epidemiology of severe malaria in PNG children

Dr Ivo Mueller, Dr John Reeder, Dr Pascal Michon, Dr Dominic Kwiatkowski, Dr Ilomo Hwaihwanje, Dr Stephen Allen, Dr Tim Davis, Dr David Weatherall, Dr James Beeson, Dr Stephen Rogerson and Dr Louis Schofield (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Rapid assessment and response: drug and alcohol in Papua New Guinea

Dr Betty Koka and Mr Simon Baldwin (Social Changes and Mental Health Services, Department of Health, PO Box 807, Waigani, NCD 131, Papua New Guinea)

What proportion of infants in Papua New Guinea seroconvert after receiving 1st dose measles vaccine at 6 months?

Dr Jonah Kurubi, Dr John Vince, Dr David Mokela, Dr Paulus Ripa, Mr Enoch Posanai, Dr William Lagani, Professor Steve Wesselingh and Dr Trevor Duke (School of Medicine and Health Sciences, University of Papua New Guinea, PO Box 5623, Boroko, NCD 111, Papua New Guinea)

Does successful universal salt iodization guarantee optimal iodine nutrition in mother and infant?

Dr Victor J. Temple, Professor F. Delange and Prof. C. J. Eastman (School of Medical Sciences, Division of Basic Medical Sciences, University of Papua New Guinea, PO Box 5623, Boroko, NCD 111, Papua New Guinea)

Baseline burden of *Haemophilus influenzae* disease among children in Papua New Guinea

Dr William Lagani, Professor John Vince and Dr Trevor Duke (Health Improvement Branch, Department of Health, PO Box 807,

Waigani, NCD 131, Papua New Guinea)

Social research methods training program strengthening HIV social research in Papua New Guinea

Dr Heather Worth (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, EHP 441, Papua New Guinea)

Health facility capacity assessment in Papua New Guinea

Dr Chris Morgan and Dr Hilda Polume (Centre for International Health, Burnet Institute, GPO Box 2284, Melbourne, Victoria 3001, Australia)

Clinical epidemiology of sago haemolytic disease in Papua New Guinea: a randomised and longitudinal study addressing the risk factors

Dr Miila Gena, Professor Jeffrey Warner and Professor Peter Siba (PO Box 3858, Boroko, NCD 111, Papua New Guinea)

A randomised non-inferiority clinical trial to assess the efficacy and safety of Duo-Cotexcin® and Coartem® for treatment of uncomplicated malaria

Dr Goa Tau, Dr David Linge, Dr Lloyd Ipai, Dr James Amini, Dr Joseph Kaven, Dr Jackson Taviri and Dr Felix Feiling (Port Moresby General Hospital, Private Mail Bag, Boroko, NCD 111, Papua New Guinea)

Use of an immunohistochemical stain for identification of *Treponema* as an aid in the diagnosis of yaws

Dr Jacob Morewaya, Dr Whitney High and Dr Graham Ogle (Division of Pathology, University of Papua New Guinea, Taurama Campus, PO Box 5623, Boroko, NCD 111, Papua New Guinea)

Participatory HIV prevention programs 'targeting' young people: rhetoric, reality, and pictures of future

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Note:

These projects have been examined and cleared by the MRAC but they have not all started, nor is there any guarantee that they all will, since in many cases this still depends on funding. It should be noted that the project funds for the MRAC were deleted from the Health Budget from 1997 to 2006.

Information about these projects may be obtained from the investigators or from the Chairperson of the Medical Research Advisory Committee (Director of Research and Monitoring, Department of Health, PO Box 807, Waigani, NCD 131)

MEDLARS BIBLIOGRAPHY

PUBLICATIONS OF RELEVANCE TO PAPUA NEW GUINEA AND MELANESIA

Bibliographic Citation List generated from MEDLARS

1 **Aaskov J, Buzacott K, Field E, Lowry K, Berlioz-Arthaud A, Holmes EC.**
Multiple recombinant dengue type 1 viruses in an isolate from a dengue patient.
J Gen Virol 2007 Dec;88(Pt 12):3334-3340.

Between 2000 and 2004, dengue virus type 1 (DENV-1) genotypes I and II from Asia were introduced into the Pacific region and co-circulated in some localities. Envelope protein gene sequences of DENV-1 from 12 patients infected on the island of New Caledonia were obtained, five of which carried genotype I viruses and six genotype II viruses. One patient harboured a mixed infection, containing viruses assigned to both genotypes I and II, as well as a number of inter-genotypic recombinants. This is the first report of a population of dengue viruses isolated from a patient containing both parental and recombinant viruses.

2 **The AIDS Reader**
Papua New Guinea AIDS crisis may mirror Africa's, UN says.
AIDS Read 2007 Nov;17(11):534.

3 **Anga G, Vince JD, Kaupa M.**
Early introduction of solids and pneumonia in young infants in Papua New Guinea: a case control study.
J Trop Pediatr 2007 Dec 21. [Epub ahead of print]

A prospective case control study to determine the association of early introduction of solids with admission to hospital with pneumonia was undertaken at Mount Hagen General Hospital (MHGH) in the highlands of Papua New Guinea (PNG) over a 3-month period in 2005. Twenty-three infants up to 6 months of age admitted with radiologically confirmed pneumonia were compared with 24 infants of similar age attending the well baby clinic for immunization and with 35 infants admitted to the hospital with conditions other than pneumonia or meningitis. There was a highly significant difference in feeding patterns between the groups. Children with pneumonia were much more likely than the control children to have started solids before the age of 2 months [OR = 18.06 (4.8-72.86)]. They were also significantly more likely to have been admitted previously with a diagnosis of pneumonia ($p < 0.001$). The children in each group were of comparable age and weight and there were no obvious confounding factors. This study provides clear evidence for the association between early introduction of solids and pneumonia in PNG highlands children. The findings are consistent with other international data. While the reasons for the association remain speculative, the association strongly reinforces the need to educate the community on best infant feeding practices and to discourage the early introduction of solids.

4 **Antony L, Laim E, Chung SS.**
Hand-assisted laparoscopy on a shoestring.
ANZ J Surg 2007 Sep;77(9):765-767.

BACKGROUND: Hand-assisted laparoscopic surgery is useful in difficult laparoscopic operations. Expensive and cumbersome devices are necessary to ensure airtightness between the surgeon's hand and the abdominal wall so that pneumoperitoneum can be maintained. METHOD: A simple method of maintaining pneumoperitoneum in hand-assisted laparoscopic surgery was carried out by tying a strong nylon suture in a criss-cross fashion on one end of the incision. Airtightness was maintained by tightening the suture around the wrist and wedging the dorsum of the hand against the abdominal wall. RESULT: The method was used successfully to remove a pyonephrotic left kidney in a 28-year-old man from Papua New Guinea. CONCLUSION: Hand-assisted laparoscopic operations can be carried out expediently and inexpensively without specialized equipment by simply tying a shoestring suture at one end of the wound.

5 **Ardian M, Meokbun E, Siburian L, Malonda E, Waramori G, Penttinen P, Lempoy J, Kenangalem E, Tjitra E, Kelly PM.**
A public-private partnership for TB control in Timika, Papua Province, Indonesia.
Int J Tuberc Lung Dis 2007 Oct;11(10):1101-1107.

SETTING: A district-level tuberculosis (TB) control programme in Papua Province, Indonesia. OBJECTIVE: To describe a successful partnership between the District Health Department, a private company and non-governmental health care providers. METHODS: Routinely collected surveillance data were analysed. A conceptual model was constructed to describe TB control in the district. Data were compared with the National TB Control Programme (NTP) performance indicators. RESULTS: Funding for the programme's TB clinic is provided by a private company (PT Freeport Indonesia). The NTP provides the policy framework, treatment guidelines and some supplies. TB clinic staff are included in training programmes and the TB laboratory in the provincial quality assurance system. TB clinic staff are responsible for diagnosis, treatment, default tracing, recording and reporting, health education and community mobilisation. The largest proportion of TB patient referrals came from the community hospital (41%). The TB notification rate (311/100000), TB-HIV (human immunodeficiency virus) co-infection (12%) and multidrug-resistant (MDR) TB (2%) are significantly higher in Mimika, but the treatment success rate for smear-positive patients (91%) is similar to Indonesian national figures. CONCLUSIONS: For true progress in attaining the United Nations Millennium Development Goals for TB in Indonesia, innovative local solutions utilising public-private partnerships are essential. The Mimika model is one such solution that should be tested elsewhere.

6 **Atkinson QD, Gray RD, Drummond AJ.**
mtDNA variation predicts population size in humans and reveals a major Southern Asian chapter in

human prehistory.
Mol Biol Evol 2008 Feb;25(2):468-474. Epub 2007 Dec 18.

The relative timing and size of regional human population growth following our expansion from Africa remain unknown. Human mitochondrial DNA (mtDNA) diversity carries a legacy of our population history. Given a set of sequences, we can use coalescent theory to estimate past population size through time and draw inferences about human population history. However, recent work has challenged the validity of using mtDNA diversity to infer species population sizes. Here we use Bayesian coalescent inference methods, together with a global data set of 357 human mtDNA coding-region sequences, to infer human population sizes through time across 8 major geographic regions. Our estimates of relative population sizes show remarkable concordance with the contemporary regional distribution of humans across Africa, Eurasia, and the Americas, indicating that mtDNA diversity is a good predictor of population size in humans. Plots of population size through time show slow growth in sub-Saharan Africa beginning 143-193 kya, followed by a rapid expansion into Eurasia after the emergence of the first non-African mtDNA lineages 50-70 kya. Outside Africa, the earliest and fastest growth is inferred in Southern Asia approximately 52 kya, followed by a succession of growth phases in Northern and Central Asia (approximately 49 kya), Australia (approximately 48 kya), Europe (approximately 42 kya), the Middle East and North Africa (approximately 40 kya), New Guinea (approximately 39 kya), the Americas (approximately 18 kya), and a second expansion in Europe (approximately 10-15 kya). Comparisons of relative regional population sizes through time suggest that between approximately 45 and 20 kya most of humanity lived in Southern Asia. These findings not only support the use of mtDNA data for estimating human population size but also provide a unique picture of human prehistory and demonstrate the importance of Southern Asia to our recent evolutionary past.

7 **Barcus MJ, Basri H, Picarima H, Manyakori C, Sekartuti , Elyazar I, Bangs MJ, Maguire JD, Baird JK.**
Demographic risk factors for severe and fatal vivax and falciparum malaria among hospital admissions in northeastern Indonesian Papua.
Am J Trop Med Hyg 2007 Nov;77(5):984-991.

Between January 1998 and December 2000, the Jayapura Provincial Public Hospital in northeastern Indonesian New Guinea (Papua) admitted 5,936 patients with a diagnosis of malaria. The microscopic diagnosis at admission was *Plasmodium falciparum* (3,976, 67%), *Plasmodium vivax* (1,135, 19%), *Plasmodium malariae* (8, <1%), and mixed species infections (817, 14%). Approximately 9% (367) of patients were classified as having severe malaria (277 *P. falciparum*, 36 *P. vivax*, 53 mixed infections, and 1 *P. malariae*) and 88 died (79 *P. falciparum*/ mixed infections and 9 *P. vivax*). Risk of fatal outcomes among severe malaria patients was indistinguishable between those with falciparum versus vivax malaria (OR =0.89; P = 0.771). Compared with non-pregnant women, pregnant women showed no higher risk of severe malaria (p = 0.643) or death caused by severe malaria (p = 0.748). This study compares admissions per population (based on census data), parasitemia, morbidity, and mortality among children versus adults, pregnant versus non-pregnant women, and urban/suburban versus rural residents.

8 **Beeson JG, Ndungu F, Persson KE, Chesson JM, Kelly GL, Uyoga S, Hallamore SL, Williams TN, Reeder JC, Brown GV, Marsh K.**
Antibodies among men and children to placental-binding *Plasmodium falciparum*-infected erythrocytes that express *var2csa*.
Am J Trop Med Hyg 2007 Jul;77(1):22-28.

During pregnancy, specific variants of *Plasmodium falciparum*-infected erythrocytes (IEs) can accumulate in the placenta through adhesion to chondroitin sulfate A (CSA) mediated by expression of PfEMP1 encoded by *var2csa*-type genes. Antibodies against these variants are associated with protection from maternal malaria. We evaluated antibodies among Kenyan, Papua New Guinean and Malawian men and Kenyan children against two different CSA-binding *P. falciparum* isolates expressing *var2csa* variants. Specific IgG was present at significant levels among some men and children from each population, suggesting exposure to these variants is not exclusive to pregnancy. However, the level and prevalence of antibodies was substantially lower overall than exposed multigravidas. IgG-binding was specific and did not represent antibodies to subpopulations of non-CSA-binding IEs, and some sera inhibited IE adhesion to CSA. These findings have significant implications for understanding malaria pathogenesis and immunity and may be significant for understanding the acquisition of immunity to maternal malaria.

9 **Butt L, Munro J.**
Rebel girls? Unplanned pregnancy and colonialism in highlands Papua, Indonesia.
Cult Health Sex 2007 Nov-Dec;9(6):585-598.

In highlands Papua, Indonesia, rapid social change under a colonial system of governance has created novel sexual opportunities for young indigenous women. Recent scholarship has viewed similar youthful sexual practices that challenge the status quo as expressions of personal agency. By looking at how young women and their families cope with unplanned pregnancies, we suggest that a more viable analytic approach would be to view sexuality, pregnancy and childbirth as a single unit of analysis. From this perspective, young women's experiences are primarily ones of constraint. Case studies offer insights into the ways a political context of colonial domination limits options and choices for young women who have children born out of wedlock. In particular, this paper describes how the 'settler gaze' - omnipresent colonial norms and judgments – creates regulatory effects in the realm of reproduction.

10 **Carr G, Raszek M, Van Soest R, Matainaho T, Shopik M, Holmes CF, Andersen RJ.**
Protein phosphatase inhibitors isolated from *Spongia irregularis* collected in Papua New Guinea.
J Nat Prod 2007 Nov;70(11):1812-1815. Epub 2007 Nov 3.

Irregularasulfate (1), a new nitrogen-containing sesterterpenoid, and the known sesterterpenoids hipposulfate C (2), halisulfate-7 (3), and igernellin (4), have been isolated from the marine sponge *Spongia irregularis* collected in Papua New Guinea. The structure of 1 was elucidated via analysis of its spectroscopic data. Sesterterpenoids 1, 2, and 3

are moderate inhibitors of the catalytic subunits of the mammalian Ser/Thr protein phosphatases calcineurin, PP-1, and PP-2A. The phosphate analogue of 3 and the thiophosphate analogue of 2 have been prepared from the corresponding natural products and evaluated for their ability to inhibit the phosphatase activity of calcineurin.

11 **Cassar O, Afonso PV, Bassot S, Plancoulaine S, Duprez R, Capuano C, Abel M, Martin PM, Gessain A.**
Novel human herpesvirus 8 subtype D strains in Vanuatu, Melanesia.
Emerg Infect Dis 2007 Nov;13(11):1745-1748.

We show human herpesvirus 8 with diverse molecular subtype D variants to be highly endemic among the Ni-Vanuatu population. Most K1 genes were nearly identical to Polynesian strains, although a few clustered with Australian or Taiwanese strains. These results suggest diverse origins of the Ni-Vanuatu population and raise questions about the ancient human population movements in Melanesia.

12 **Cassar O, Capuano C, Bassot S, Charavay F, Duprez R, Afonso PV, Abel M, Walter H, Mera W, Martin PM, Chungue E, Gessain A.**
Human T lymphotropic virus type 1 subtype C Melanesian genetic variants of the Vanuatu Archipelago and Solomon Islands share a common ancestor.
J Infect Dis 2007 Aug 15;196(4):510-521. Epub 2007 Jun 28.

BACKGROUND: Melanesia is endemic for human T lymphotropic virus type 1 (HTLV-1) subtype C. In 2005, we identified 4 infected women from Ambae Island, Vanuatu. Subsequently, 4247 Ni-Vanuatu originating from 18 islands were enrolled to define HTLV-1 epidemiological determinants and to characterize the viral strains molecularly. METHODS: Plasma from 1074 males and 3173 females were screened for HTLV-1/2 antibodies by particle agglutination (PA) and an immunofluorescence assay (IFA). Positive and/or borderline samples were then tested by a Western blot (WB) confirmatory assay. DNAs were amplified to obtain a 522-bp *env* gene fragment. Phylogenetic and molecular-clock analyses were performed. RESULTS: Of 4247 samples, 762 were positive and/or borderline by IFA/PA, and 26 of them were confirmed to be HTLV-1 positive by WB. The overall HTLV-1 seroprevalence was 0.62%. Viral transmission was found within families of infected index case patients. A geographic heterogeneity of HTLV-1 seroprevalence was observed among the islands. All 41 of the new *env* sequences belonged to HTLV-1 subtype C. Phylogenetic and molecular-clock analyses suggested that Ni-Vanuatu and Solomon Islander strains emerged from a common ancestor ~10,000 years ago. CONCLUSION: The Vanuatu archipelago is endemic for HTLV-1 with a diversity of subtype C variants. These strains were probably introduced into Vanuatu during ancient migration of the original settlers a few thousand years ago.

13 **Cole-Tobian JL, Michon P, Dabod E, Mueller I, King CL.**
Dynamics of asymptomatic *Plasmodium vivax* infections and Duffy binding protein polymorphisms in relation to parasitemia levels in Papua New Guinean children.
Am J Trop Med Hyg 2007 Nov;77(5):955-962.

The interaction between *Plasmodium vivax* Duffy binding protein II (PvDBPII) and human erythrocyte Duffy antigen is necessary for blood stage infections. However, PvDBPII is highly polymorphic. We recently observed that certain recombinant DBPII variants bind better to erythrocytes in vitro. To examine the hypothesis that haplotypes with enhanced binding have increased parasitemia levels, we followed 206 Papua New Guinean children biweekly for six months with a total of 713 *P. vivax* samples genotyped. Twenty-seven PvDBPII haplotypes were identified, and 3 haplotypes accounted for 57% of the infections. The relative frequencies of dominant haplotypes remained stable throughout the study. There was no significant association of PvDBPII alleles or haplotypes with *P. vivax* parasitemia. The dominant haplotype (26% of samples), however, corresponded to a high-binding haplotype. Thus, common haplotypes are not likely to have arisen from increased fitness as measured by greater parasitemia levels. The restricted number of common haplotypes increases the feasibility of a PvDBPII-based vaccine.

14 **Cournil A, Defay R, Lacroux A, Barny S, Fontbonne A, CALDIA Study Group.**
Paradoxical relationships between anthropometric variables and phenotypic expression of the metabolic syndrome in nondiabetic Polynesians of New Caledonia.
Diabetes Care 2007 Jul;30(7):1909-1911. Epub 2007 Apr 19.

15 **Davis RA, Simpson MM, Nugent RB, Carroll AR, Avery VM, Rali T, Chen H, Qurallo B, Quinn RJ.**
Pim2 inhibitors from the Papua New Guinean plant *Cupaniopsis macropetala*.
J Nat Prod 2007 Dec 29 [Epub ahead of print]

Bioassay-guided fractionation of an organic extract from the leaves of *Cupaniopsis macropetala* resulted in the isolation of a new alkaloid, galloyl tyramine (1), together with the known flavonoid glycoside quercitrin (2). The structure of 1 was determined following 1D and 2D NMR, IR, UV, and MS data analysis. Compounds 1 and 2 displayed IC 50 values of 161 and 25 microM, respectively, in a Pim2 enzyme assay.

16 **Dent AE, Yohn CT, Zimmerman PA, Vulule J, Kazura JW, Moormann AM.**
A polymerase chain reaction/ligase detection reaction fluorescent microsphere assay to determine *Plasmodium falciparum* MSP-1(19) haplotypes.
Am J Trop Med Hyg 2007 Aug;77(2):250-255.

The merozoite surface protein-1 (MSP-1) is a blood stage antigen currently being tested as a vaccine against *Plasmodium falciparum* malaria. Determining the MSP-1(19) haplotype(s) present during infection is essential for assessments of MSP-1 vaccine efficacy and studies of protective immunity in human populations. The C-terminal fragment (MSP-1(19)) has four predominant haplotypes based on point mutations resulting in non-synonymous amino acid changes: E-TSR (PNG-MAD20 type), E-KNG (Uganda-PA type), Q-KNG (Wellcome type), and Q-TSR (Indo type). Current techniques using direct DNA sequencing are laborious and expensive. We present an MSP-1(19) allele-specific polymerase chain reaction (PCR)/ligase detection reaction-fluorescent microsphere assay (LDR-FMA) that allows simultaneous detection of the four predominant MSP-1(19) haplotypes with a sensitivity

and specificity comparable with other molecular methods and a semi-quantitative determination of haplotype contribution in mixed infections. Application of this method is an inexpensive, accurate, and high-throughput alternative to distinguish the predominant MSP-1(19) haplotypes in epidemiologic studies.

17 **Dubey SP, Molumi CP.**
Critical look at the surgical approaches of nasopharyngeal angiofibroma excision and 'total maxillary swing' as a possible alternative.
Ann Otol Rhinol Laryngol 2007 Oct;116(10):723-730.

OBJECTIVES: We critically analyzed different surgical approaches used for the excision of nasopharyngeal angiofibroma (NPA) at our hospital in a 10-year period and proposed 'total maxillary swing' as a possible alternative approach. METHODS: Retrospective review of the clinical and operative records was done regarding 39 cases of NPA that were surgically managed from 1995 to 2005. The duration of the surgical procedures, amount of blood loss and transfusion, time to recurrence, frequency of recurrence, functional and cosmetic deformity, and complications of all surgical procedures were thoroughly analyzed. These parameters were compared with total maxillary swing, a new approach recently being used by us, for the one-time excision of NPA. RESULTS: A total of 61 operations including revision surgeries for recurrences were done in the 39 cases of NPA. Conventional surgical approaches were performed in 37 cases, and the total maxillary swing approach was used in 2 cases. This new approach provided a wider surgical exposure for complete tumor resection and better hemostasis without any recurrence or major functional deformity, which were seen with some of the conventional approaches. CONCLUSIONS: We found total maxillary swing to be a relatively safe alternative approach for the resection of NPA. We removed the entire tumors in a wider surgical field under the microscope with reduced blood loss and minimal complications or chance of recurrences.

18 **Fotinatos N, Warmington A, Walker T, Pilbeam M.**
Estimates for cervical abnormalities in Vanuatu.
Aust NZ J Public Health 2007 Dec;31(6):571-575.

OBJECTIVE: To use the Pap smear to establish a recent prevalence of cervical abnormalities within a select population in Vanuatu, a developing country. METHODS: Cervical smears (n=907) were collected from Ni-Vanuatu women from both urban and rural islands within Vanuatu between August 2001 and September 2005. RESULTS: The prevalence of low-grade epithelial abnormalities for the total population was 2.9% and the prevalence of the high-grade epithelial abnormalities/cancer was 2.0%. There was a significant difference ($p < 0.05$) in prevalence of high-grade epithelial abnormalities/cancer between the urban and rural populations sampled, with a higher prevalence in the urban population. CONCLUSIONS: The prevalence of pre-cancer and cancer in Vanuatu is high compared with Victorian (Australian) statistics yet comparable with other developing countries with no cervical screening programs available. IMPLICATIONS: This study will hopefully assist in future planning of women's health programs and relevant preventive strategies to combat cervical cancer in Vanuatu.

19 **Graves P, Gelband H.**
Vaccines for preventing malaria.
Cochrane Database Syst Rev 2007 Jul 18;(4):CD000129.

BACKGROUND: Four types of malaria vaccine, SPf66 and MSP/RESA vaccines (against the asexual stages of the *Plasmodium* parasite) and CS-NANP and RTS,S vaccines (against the sporozoite stages), have been tested in randomized controlled trials in endemic areas. OBJECTIVES: To assess malaria vaccines against *Plasmodium falciparum*, *P. vivax*, *P. malariae* and *P. ovale* in preventing infection, disease and death. SEARCH STRATEGY: We searched the Cochrane Infectious Diseases Group Specialized Register (April 2004), CENTRAL (The Cochrane Library Issue 2, 2004), MEDLINE (1966 to April 2004), EMBASE (1980 to April 2004), Science Citation Index (1981 to April 2004), and reference lists of articles. We also contacted organizations and researchers in the field. SELECTION CRITERIA: Randomized controlled trials comparing vaccines against *Plasmodium falciparum*, *P. vivax*, *P. malariae* or *P. ovale* with placebo or routine antimalarial control measures in people of any age receiving a challenge malaria infection. DATA COLLECTION AND ANALYSIS: Two reviewers independently assessed trial quality and extracted data. MAIN RESULTS: Eighteen efficacy trials involving 10,971 participants were included. There were ten trials of SPf66 vaccine, four trials of CS-NANP vaccines, two trials of RTS,S vaccine, and two of MSP/RESA vaccine. Results with SPf66 in reducing new malaria infections (*P. falciparum*) were heterogeneous: it was not effective in four African trials (Peto odds ratio (OR) 0.96, 95% confidence interval (CI) 0.81 to 1.14), but in five trials outside Africa the number of first attacks was reduced (Peto OR 0.77, 95% CI 0.67 to 0.88). Trials to date have not indicated any serious adverse events with SPf66 vaccine. In three trials of CS-NANP vaccines, there was no evidence for protection by these vaccines against *P. falciparum* malaria (Peto OR 1.12, 95% CI 0.64 to 1.93). In a small trial in non-immune adults in the USA, RTS,S gave strong protection against experimental infection with *P. falciparum*. In a trial in an endemic area of The Gambia in semi-immune people, there was a reduction in clinical malaria episodes in the second year of follow up, corresponding to a vaccine efficacy of 66% (CI 14% to 85%). In a trial in Papua New Guinea, MSP/RESA had no protective effect against episodes of clinical malaria. There was evidence of an effect on parasite density, but this differed according to whether the participants had been pretreated with sulfadoxine/pyrimethamine or not. The prevalence of infections with the parasite subtype of MSP2 in the vaccine was reduced compared with the other subtype (Peto OR 0.35, CI 0.23 to 0.53). AUTHORS' CONCLUSIONS: There is no evidence for protection by SPf66 vaccines against *P. falciparum* in Africa. There is a modest reduction in attacks of *P. falciparum* malaria following vaccination with SPf66 in other regions. Further research with SPf66 vaccines in South America or with new formulations of SPf66 may be justified. There was not enough evidence to evaluate the use of CS-NANP vaccines. The RTS,S vaccine showed promising result, as did the MSP/RESA vaccine, but it should include the other main allelic form of MSP2. The MSP/RESA trial demonstrated that chemotherapy during a vaccine trial may reduce vaccine efficacy, and trials should consider very

carefully whether this practice is justified.

20 **Greenhill AR, Shipton WA, Blaney BJ, Warner JM.**
Fungal colonization of sago starch in Papua New Guinea.
Int J Food Microbiol 2007 Nov 1;119(3):284-290. Epub 2007 Aug 19.

Sago starch is an important source of dietary carbohydrates in lowland Papua New Guinea. Over the past 30 years there have been sporadic reports of severe illness following consumption of sago starch. A common assumption is that fungal metabolites might be associated with the illness, leading to the need for a more thorough investigation of the mycoflora of sago starch. Sago starch was collected from areas of high sago consumption in Papua New Guinea for fungal analysis (69 samples). Storage methods and duration were recorded at the time of collection and pH on arrival at the laboratory. Yeasts were isolated from all samples except two, ranging from $1.2x10^3$ to $8.3x10^7$ cfu/g. Moulds were isolated from 65 of the 69 samples, ranging from $1.0x10^2$ to $3.0x10^6$ cfu/g. Of 44 samples tested for ergosterol content, 42 samples showed the presence of fungal biomass. Statistical analyses indicated that sago starch stored for greater than five weeks yielded significantly higher ergosterol content and higher numbers of moulds than sago stored for less than five weeks. The method of storage was also shown to influence mould numbers with storage in natural woven fibre containers returning significantly greater numbers than present in other storage methods tested. Potentially mycotoxigenic genera of moulds including *Aspergillus* and *Penicillium* were commonly isolated from sago starch, and such storage factors that influence the growth of these and other filamentous fungi might contribute to the safety of traditional sago starch in PNG.

21 **Greenhill AR, Shipton WA, Omoloso AD, Amoa B, Warner JM.**
Bacterial contamination of sago starch in Papua New Guinea.
J Food Prot 2007 Dec;70(12):2868-2872.

Sago starch is an important food in lowland Papua New Guinea. Extraction of the starch from the palm and storage were performed by way of traditional methods that have been used for thousands of years. Currently, very little is known about the microbiology of sago starch. Sago samples were collected from areas of high starch utilization and analyzed for the presence of bacterial pathogens and indicator organisms. Storage methods and duration were recorded at the time of collection, and pH and water activity on arrival at the laboratory. Sago starch was found to harbor high levels of fecal contamination, as well as various food pathogens including *Salmonella*, *Bacillus cereus*, and coagulase-positive staphylococci. *Clostridium perfringens* was only present infrequently in samples and in very low numbers, while *Listeria monocytogenes* was not isolated from sago starch. The presence of high levels of fecal contamination in sago starch is of particular concern, and may contribute to diarrheal disease in rural Papua New Guinea.

22 **Grimberg BT, Udomsangpetch R, Xainli J, McHenry A, Panichakul T, Sattabongkot J, Cui L, Bockarie M, Chitnis C, Adams J, Zimmerman PA, King CL.**
Plasmodium vivax invasion of human erythrocytes inhibited by antibodies directed against the Duffy binding protein.
PLoS Med 2007 Dec;4(12):e337.

BACKGROUND: *Plasmodium vivax* invasion requires interaction between the human Duffy antigen on the surface of erythrocytes and the *P. vivax* Duffy binding protein (PvDBP) expressed by the parasite. Given that Duffy-negative individuals are resistant and that Duffy-negative heterozygotes show reduced susceptibility to blood-stage infection, we hypothesized that antibodies directed against region two of *P. vivax* Duffy binding protein (PvDBPII) would inhibit *P. vivax* invasion of human erythrocytes. METHODS AND FINDINGS: Using a recombinant region two of the *P. vivax* Duffy binding protein (rPvDBPII), polyclonal antibodies were generated from immunized rabbits and affinity purified from the pooled sera of 14 *P. vivax*-exposed Papua New Guineans. It was determined by ELISA and by flow cytometry, respectively, that both rabbit and human antibodies inhibited binding of rPvDBPII to the Duffy antigen N-terminal region and to Duffy-positive human erythrocytes. Additionally, using immunofluorescent microscopy, the antibodies were shown to attach to native PvDBP on the apical end of the *P. vivax* merozoite. In vitro invasion assays, using blood isolates from individuals in the Mae Sot district of Thailand, showed that addition of rabbit anti-PvDBPII Ab or serum (antibodies against, or serum containing antibodies against, region two of the *Plasmodium vivax* Duffy binding protein) (1:100) reduced the number of parasite invasions by up to 64%, while pooled PvDBPII antisera from *P. vivax*-exposed people reduced *P. vivax* invasion by up to 54%. CONCLUSIONS: These results show, for what we believe to be the first time, that both rabbit and human antibodies directed against PvDBPII reduce invasion efficiency of wild *P. vivax* isolated from infected patients, and suggest that a PvDBP-based vaccine may reduce human blood-stage *P. vivax* infection.

23 **Guignard R, Truong T, Rougier Y, Baron-Dubourdieu D, Guénel P.**
Alcohol drinking, tobacco smoking, and anthropometric characteristics as risk factors for thyroid cancer: a countrywide case-control study in New Caledonia.
Am J Epidemiol 2007 Nov 15;166(10):1140-1149. Epub 2007 Sep 12.

Exceptionally high incidence rates of thyroid cancer are observed in New Caledonia, particularly in Melanesian women. To investigate further the etiology of thyroid cancer and to clarify the reasons of this elevated incidence, the authors conducted a countrywide population-based case-control study in this multiethnic population. The study included 332 cases with histologically verified papillary or follicular carcinoma (293 women and 39 men) diagnosed in 1993-1999 and 412 population controls (354 women and 58 men) frequency matched by gender and 5-year age group. Thyroid cancer was negatively associated with tobacco smoking and alcohol drinking, but no inverse dose-response relation was observed. Height was positively associated with thyroid cancer, particularly in men. Strong positive associations with weight and body mass index were observed in Melanesian women aged 50 years or more, with an odds ratio of 5.5 (95% confidence interval: 1.5, 20.3) for a body mass index of 35 kg/m^2 or greater compared with normal-weight women, and there was a clear dose-response trend. This

study clarifies the role of overweight for thyroid cancer in postmenopausal women. Because of the high prevalence of obesity among Melanesian women of New Caledonia, this finding may explain in part the exceptionally elevated incidence of thyroid cancer in this group.

24 **Hall AJ, Quinnell RJ, Raiko A, Lagog M, Siba P, Morroll S, Falcone FH.**
Chitotriosidase deficiency is not associated with human hookworm infection in a Papua New Guinean population.
Infect Genet Evol 2007 Dec;7(6):743-747. Epub 2007 Jul 28.

Human chitotriosidase (CHIT1) is a chitinolytic enzyme with suggested anti-fungal properties. Previous studies have suggested that chitotriosidase may also protect individuals against filarial nematode infections and malaria. A mutant allele, which renders chitotriosidase unstable and enzymatically inactive, is found at a frequency of >20% in Caucasians and other populations. This allele is found at much lower frequency in parts of West Africa where malarial and intestinal helminth infections are endemic. Here, we investigate whether there is a significant association between chitotriosidase genotype and the intensity of hookworm infection in 693 individuals from five villages in Papua New Guinea. Individuals were genotyped for chitotriosidase using a PCR-based assay. There was no association between CHIT1 genotype and the intensity of hookworm infection as determined by faecal egg counts. The frequency of the mutant allele was 0.251, very similar to that found in non-endemic countries. The extent of geographical variation in allele frequencies across worldwide populations was not high (F(st)=0.11), and does not provide evidence for directional selection at this locus between different geographical areas. We conclude that the CHIT1 genotype does not play a crucial role in protection against hookworm infection. This does not correlate with a previous study that linked the mutant CHIT1 genotype to filariasis susceptibility. The possible reasons for this discrepancy are discussed.

25 **Hietala SF, Bhattarai A, Msellem M, Röshammar D, Ali AS, Strömberg J, Hombhanje FW, Kaneko A, Björkman A, Ashton M.**
Population pharmacokinetics of amodiaquine and desethylamodiaquine in pediatric patients with uncomplicated falciparum malaria.
J Pharmacokinet Pharmacodyn 2007 Oct;34(5):669-686. Epub 2007 Jul 10.

The study aimed to characterize the population pharmacokinetics of amodiaquine (AQ) and its major metabolite N-desethylamodiaquine (N-DEAQ), and to assess the correlation between exposure to N-DEAQ and treatment outcome. Blood samples from children in two studies in Zanzibar and one in Papua New Guinea were included in the pharmacokinetic analysis (n = 86). The children had been treated with AQ in combination with artesunate or sulphadoxine-pyrimethamine. The population pharmacokinetics of AQ and N-DEAQ were modeled using the non-linear mixed effects approach as implemented in NONMEM. Bayesian post-hoc estimates of individual pharmacokinetic parameters were used to generate individual profiles of N-DEAQ exposure. The correlation between N-DEAQ exposure and effect was studied in 212 patients and modeled with logistic regression in NONMEM. The pharmacokinetics of AQ and N-DEAQ were best described by two parallel two-compartment models with a central and a peripheral compartment for each compound. The systemic exposure to AQ was low in comparison to N-DEAQ. The t(1/2 lambda) of N-DEAQ ranged from 3 days to 12 days. There was a statistically significant, yet weak, association between N-DEAQ concentration on day 7 and treatment outcome. The age-based dosing schedule currently recommended in Zanzibar appeared to result in inadequate exposure to N-DEAQ in many patients.

26 **Hume JC, Tunnicliff M, Ranford-Cartwright LC, Day KP.**
Susceptibility of *Anopheles gambiae* and *Anopheles stephensi* to tropical isolates of *Plasmodium falciparum*.
Malar J 2007 Oct 24;6:139.

BACKGROUND: The susceptibility of anopheline mosquito species to *Plasmodium* infection is known to be variable with some mosquitoes more permissive to infection than others. Little work, however, has been carried out investigating the susceptibility of major malaria vectors to geographically diverse tropical isolates of *Plasmodium falciparum* aside from examining the possibility of infection extending its range from tropical regions into more temperate zones. METHODS: This study investigates the susceptibility of two major tropical mosquito hosts (*Anopheles gambiae* and *Anopheles stephensi*) to *P. falciparum* isolates of different tropical geographical origins. Cultured parasite isolates were fed via membrane feeders simultaneously to both mosquito species and the resulting mosquito infections were compared. RESULTS: Infection prevalence was variable with African parasites equally successful in both mosquito species, Thai parasites significantly more successful in *An. stephensi*, and PNG parasites largely unsuccessful in both species. CONCLUSION: Infection success of *P. falciparum* was variable according to geographical origin of both the parasite and the mosquito. Data presented raise the possibility that local adaptation of tropical parasites and mosquitoes has a role to play in limiting gene flow between allopatric parasite populations.

27 **Johansen CA, Susai V, Hall RA, Mackenzie JS, Clark DC, May FJ, Hemmerter S, Smith DW, Broom AK.**
Genetic and phenotypic differences between isolates of Murray Valley encephalitis virus in Western Australia, 1972-2003.
Virus Genes 2007 Oct;35(2):147-154. Epub 2007 Mar 29.

Murray Valley encephalitis virus (MVEV) is a medically important mosquito-borne flavivirus found in Australia and Papua New Guinea (PNG). Partial envelope gene nucleotide sequences of 28 isolates of MVEV from Western Australia (WA) between 1972 and 2003 were aligned and compared phylogenetically with the prototype MVE-1-51 from Victoria in 1951 and isolates from northern Queensland and PNG. Monoclonal antibody-binding patterns were also investigated. Results showed that the majority of isolates of MVEV from widely disparate locations in WA were genetically and phenotypically homogeneous. Furthermore, isolates of MVEV from WA and northern Queensland were almost identical, confirming results from earlier studies. Recent isolates of MVEV from Western

Province in PNG were more similar to Australian isolates of MVEV than to isolates from PNG in 1956 and 1966, providing further evidence for the movement of flaviviruses between PNG and Australia. Additional representatives of a unique variant of MVEV (OR156) from Kununurra in the northeast Kimberley region of WA were also detected. This suggests that the OR156 lineage is still intermittently active but may be restricted to a small geographic area in northern WA, possibly due to altered biological characteristics.

28 **Karunajeewa HA, Ilett KF, Mueller I, Siba P, Law I, Page-Sharp M, Lin E, Lammey J, Batty KT, Davis TM.**
Pharmacokinetics and efficacy of piperaquine and chloroquine in Melanesian children with uncomplicated malaria.
Antimicrob Agents Chemother 2008 Jan;52(1):237-243. Epub 2007 Oct 29.

The disposition of chloroquine (CQ) and the related 4-aminoquinoline, piperaquine (PQ), were compared in Papua New Guinean children with uncomplicated malaria. Twenty-two children were randomized to 3 days of PQ phosphate at 20 mg/kg/day (12 mg of PQ base/kg/day) coformulated with dihydroartemisinin (DHA-PQ), and twenty children were randomized to 3 days of CQ at 10 mg base/kg/day with a single dose of sulfadoxine-pyrimethamine (CQ-SP). After a 42-day intensive sampling protocol, PQ, CQ, and its active metabolite monodesethylchloroquine (DECQ) were assayed in plasma by using high-performance liquid chromatography. A two-compartment model with first-order absorption was fitted to the PQ and CQ data. There were no significant differences in age, gender, body weight, or admission parasitemia between the two groups. The PCR-corrected 42-day adequate clinical and parasitological responses were 100% for DHA-PQ and 94% for CQ-SP, but *P. falciparum* reinfections during follow-up were common (33 and 18%, respectively). For PQ, the median volume of distribution at steady state, allowing for bioavailability (V(ss)/F), was 431 liters/kg (interquartile range [IQR], 283 to 588 liters/kg), the median clearance (CL/F) was 0.85 liters/h/kg (IQR, 0.67 to 1.06 liters/h/kg), the median distribution half-life (t(1/2)(alpha)) was 0.12 h (IQR, 0.05 to 0.66 h), and the median elimination half-life (t(1/2)(beta)) was 413 h (IQR, 318 to 516 h). For CQ, the median V(ss)/F was 154 liters/kg (IQR, 101 to 210 liters/kg), the median CL/F was 0.80 liters/h/kg (IQR, 0.52 to 0.96 liters/h/kg), the median t(1/2)(alpha) was 0.43 h (IQR, 0.05 to 1.82 h), and the median t(1/2)(beta) was 233 h (IQR, 206 to 298 h). The noncompartmentally derived median DECQ t(1/2)(beta) was 290 h (IQR, 236 to 368 h). Combined molar concentrations of DECQ and CQ were higher than those of PQ during the elimination phase. Although PQ has a longer t(1/2)(beta) than CQ, its prompt distribution and lack of active metabolite may limit its posttreatment malaria-suppressive properties.

29 **King E, Webster T, Siba P, Pantumari J.**
Medical interventions, social science, and resource-poor countries.
Lancet 2007 Sep 1;370(9589):739-740.

30 **Linington RG, Edwards DJ, Shuman CF, McPhail KL, Matainaho T, Gerwick WH.**
Symplocamide A, a potent cytotoxin and chymotrypsin inhibitor from the marine cyanobacterium *Symploca* sp.
J Nat Prod 2008 Jan;71(1):22-27. Epub 2007 Dec 29.

Investigation of a *Symploca* sp. from Papua New Guinea has led to the isolation of symplocamide A (1), a potent cancer cell cytotoxin, which also inhibits serine proteases with a 200-fold greater inhibition of chymotrypsin over trypsin. The complete stereostructure of symplocamide A was determined by detailed NMR and MS analysis as well as chiral HPLC analysis of the component amino acid residues. The presence of several unusual structural features in symplocamide A provides new insights into the pharmacophore model for protease selectivity in this drug class and may underlie the potent cytotoxicity of this compound to H-460 lung cancer cells (IC50=40 nM) as well as neuro-2a neuroblastoma cells (IC50=29 nM).

31 **Lum JK, Kaneko A, Taleo G, Amos M, Reiff DM.**
Genetic diversity and gene flow of humans, *Plasmodium falciparum*, and *Anopheles farauti s.s.* of Vanuatu: inferred malaria dispersal and implications for malaria control.
Acta Trop 2007 Aug;103(2):102-107. Epub 2007 May 25.

A comparison of the patterns of gene flow within and between islands and the genetic diversities of the three species required for malaria transmission (humans, *Plasmodium falciparum*, and *Anopheles farauti s.s.*) within the model island system of Vanuatu shows that the active dispersal of *An. farauti s.s.* is responsible for within island movement of parasites. In contrast, since both *P. falciparum* and *An. farauti s.s.* populations are largely restricted to islands, movement of parasites between islands is likely due to human transport. Thus, control of vectors is crucial for controlling malaria within islands, while control of human movement is essential to control malaria transmission across the archipelago.

32 **Marfurt J, Mueller I, Sie A, Maku P, Goroti M, Reeder JC, Beck HP, Genton B.**
Low efficacy of amodiaquine or chloroquine plus sulfadoxine-pyrimethamine against *Plasmodium falciparum* and *P. vivax* malaria in Papua New Guinea.
Am J Trop Med Hyg 2007 Nov;77(5):947-954.

Because of increasing resistance to 4-aminoquinolines in Papua New Guinea, combination therapy of amodiaquine (AQ) or chloroquine (CQ) plus sulfadoxine-pyrimethamine (SP) was introduced as first-line treatment against uncomplicated malaria in 2000. The purpose of this study was to monitor in vivo efficacy of the current standard combination therapy against *Plasmodium falciparum* and *P. vivax* malaria. Studies were conducted between 2003 and 2005 in the Simbu, East Sepik, and Madang Provinces in Papua New Guinea according to the revised protocol of the World Health Organization (WHO) for assessment of antimalarial drug efficacy. Children between six months and seven years of age with clinically overt and parasitologically confirmed *P. falciparum* or *P. vivax* malaria were treated according to the new policy guidelines (ie, AQ plus SP given to patients weighing <14 kg and CQ plus SP given to patients weighing ≥14 kg). Children were monitored up to day 28 and classified according to clinical and parasitological outcome as adequate clinical and parasitological response (ACPR), early treatment failure (ETF), late clinical

failure (LCF), or late parasitological failure (LPF). For *P. falciparum* malaria, polymerase chain reaction (PCR)-corrected treatment failure rates up to day 28 ranged between 10.3% and 28.8% for AQ plus SP and between 5.6% and 28.6% for CQ plus SP, depending on the region and the year of assessment. Overall treatment failure rate with AQ or CQ plus SP for *P. vivax* malaria was 12%. Our results suggest that the current first-line treatment in Papua New Guinea is not sufficiently effective. According to the new WHO guidelines for the treatment of malaria, a rate of parasitological resistance greater than 10% in the two dominant malaria species in the country justifies a change in treatment policy.

33 **Massey P, Durrheim DN, Speare R.**
Inadequate chemoprophylaxis and the risk of malaria.
Aust Fam Physician 2007 Dec;36(12):1058-1060.

BACKGROUND: Malaria is an important disease for Australian travellers, particularly to Papua New Guinea. Travellers often seek health advice from their general practitioner before travel or if they develop illness after travel. METHOD: A retrospective cohort investigation into malaria risk in a group of adult Australians that trekked the Kokoda trail in Papua New Guinea. RESULTS: Six of 38 group members were diagnosed with malaria on return from Papua New Guinea. None of the 12 individuals who took chemoprophylaxis for the recommended period post-travel developed malaria compared to 4/24 travellers who terminated prophylaxis prematurely or 2/2 who took no chemoprophylaxis. DISCUSSION: Chemoprophylaxis is effective if taken for the full recommended period following travel to a malaria endemic area; 4 weeks for doxycycline and mefloquine, and 7 days for atovaquone+proguanil. Malaria is a likely cause of illness in recently returned travellers from Papua New Guinea who develop a febrile illness.

34 **Mehlotra RK, Bockarie MJ, Zimmerman PA.**
CYP2B6 983T>C polymorphism is prevalent in West Africa but absent in Papua New Guinea: implications for HIV/AIDS treatment.
Br J Clin Pharmacol 2007 Sep;64(3):391-395. Epub 2007 Mar 28.

AIMS: To determine the prevalence of the novel *CYP2B6* functional polymorphism 983T>C in Papua New Guinea where HIV/AIDS poses a significant health problem. METHOD: We genotyped Papua New Guineans (PNG, n = 174), West Africans (WA, n = 170), and North Americans (NA, n = 361). RESULTS: The polymorphism was absent in PNG, while its overall frequency was 4.7% in WA. Among NA, the polymorphism was present in African-Americans (7.5%) and Hispanic-Americans (1.1%) but not in Caucasian-Americans and Asian-Americans. Haplotype analysis indicated that 983T>C was present alone as the *CYP2B6*18* allele in WA and African-Americans. CONCLUSIONS: Significant interethnic differences occur at the *CYP2B6* locus, which may influence treatment outcomes with efavirenz.

35 **Mona S, Tommaseo-Ponzetta M, Brauer S, Sudoyo H, Marzuki S, Kayser M.**
Patterns of Y-chromosome diversity intersect with the Trans-New Guinea hypothesis.
Mol Biol Evol 2007 Nov;24(11):2546-2555. Epub 2007 Sep 10.

The island of New Guinea received part of the first human expansion out of Africa (>40,000 years ago), but its human genetic history remains poorly understood. In this study, we examined Y-chromosome diversity in 162 samples from the Bird's Head region of northwest New Guinea (NWNG) and compared the results with previously obtained data from other parts of the island. NWNG harbors a high level of cultural and linguistic diversity and is inhabited by non-Austronesian (ie, Papuan)-speaking groups as well as harboring most of West New Guinea's (WNG) Austronesian-speaking groups. However, 97.5% of its Y-chromosomes belong to 5 haplogroups that originated in Melanesia; hence, the Y-chromosome diversity of NWNG (and, according to available data, of New Guinea as a whole) essentially reflects a local history. The remaining 2.5% belong to 2 haplogroups (O-M119 and O-M122) of East Asian origin, which were brought to New Guinea by Austronesian-speaking migrants around 3,500 years ago. Thus, the Austronesian expansion had only a small impact on shaping Y-chromosome diversity in NWNG, although the linguistic impact of this expansion to this region was much higher. In contrast, the expansion of Trans-New Guinea (TNG) speakers (non-Austronesian) starting about 6,000-10,000 years ago from the central highlands of what is now Papua New Guinea, presumably in combination with the expansion of agriculture, played a more important role in determining the Y-chromosome diversity of New Guinea. In particular, we identified 2 haplogroups (M-P34 and K-M254) as suggestive markers for the TNG expansion, whereas 2 other haplogroups (C-M38 and K-M9) most likely reflect the earlier local Y-chromosome diversity. We propose that sex-biased differences in the social structure and cultural heritage of the people involved in the Austronesian and the TNG expansions played an important role (among other factors) in shaping the New Guinean Y-chromosome landscape.

36 **Naab PJ, Russell JA.**
Judgments of emotion from spontaneous facial expressions of New Guineans.
Emotion 2007 Nov;7(4):736-744.

The claim that specific discrete emotions can be universally recognized from human facial expressions is based mainly on the study of expressions that were posed. The current study (N=50) examined recognition of emotion from 20 spontaneous expressions from Papua New Guinea photographed, coded, and labeled by P. Ekman (1980). For the 16 faces with a single predicted label, endorsement of that label ranged from 4.2% to 45.8% (mean 24.2%). For 4 faces with 2 predicted labels (blends), endorsement of one or the other ranged from 6.3% to 66.6% (mean 38.8%). Of the 24 labels Ekman predicted, 11 were endorsed at an above-chance level, and 13 were not. Spontaneous expressions do not achieve the level of recognition achieved by posed expressions.

37 **Naepi D.**
Doing things her way – the Pacific way. Interview by Teresa O'Connor.
Nurs NZ 2007 Aug;13(7):18-19.

38 **Ng CY, Gu F, Phong WY, Chen YL, Lim SP, Davidson A, Vasudevan SG.**
Construction and characterization of a stable subgenomic dengue virus type 2 replicon system

for antiviral compound and siRNA testing.
Antiviral Res 2007 Dec;76(3):222-231. Epub 2007 Jul 26.

Self-replicating, non-infectious flavivirus subgenomic replicons have been broadly used in the studies of trans-complementation, adaptive mutation, viral assembly and packaging in Kunjin, yellow fever and West Nile viruses. We describe here the construction of subgenomic EGFP- or *Renilla* luciferase-reporter based dengue replicons of the type 2 New Guinea C (NGC) strain and the establishment of stable BHK21 cell lines harboring the replicons. In replicon cells, viral proteins and RNAs are stably expressed at levels similar to cells transfected with the full length NGC infectious RNA. Furthermore, the replicon can be packaged by separately transfected C (core)-prM (pre-membrane)-E (envelope) polyprotein constructs. The replicon cells were subjected to treatment with several antiviral compounds and inhibition of the replicon was observed in treatment with known nucleoside analog inhibitors of NS5 such as 2'-C-methyladenosine (EC(50)=2.42 +/- 0.59 microM), or ribavirin (EC(50)=6.77 +/-1.33 microM), mycophenolic acid (EC(50)=1.31 +/- 0.27 microM) and siRNA against NS3. The BHK-replicon cells have been stably maintained for about 10 passages without significant loss in reporter intensity and are sufficiently robust for both research and drug discovery.

39 **Okumiya K, Fujisawa M, Ishine M, Wada T, Sakamoto R, Hirata Y, Del Saz EG, Griapon Y, Togodly A, Sanggenafa N, Rantetampang AL, Kokubo Y, Kuzuhara S, Matsubayashi K.**
Fieldwork survey of neurodegenerative diseases in West New-Guinea in 2001-02 and 2006-07 [Jp]
Rinsho Shinkeigaku 2007 Nov;47(11):977-978.

40 **Owen IL, Reid SA.**
Survival of *Trichinella papuae* muscle larvae in a pig carcass maintained under simulated natural conditions in Papua New Guinea.
J Helminthol 2007 Dec;81(4):429-432. Epub 2007 Nov 16.

In Papua New Guinea, *Trichinella papuae*, a non-encapsulated species, is circulating among wild and domestic pigs and saltwater crocodiles. Since an important phase of the life cycle of nematodes of the genus *Trichinella* is the time of survival of infective larvae in decaying muscle tissues of the hosts, the carcass of a pig, experimentally infected with larvae of *T. papuae*, was exposed to the environmental conditions of Papua New Guinea to establish how long these larvae would survive and remain infective to a new host. Larvae retained their infectivity in the pig carcass up to 9 days after slaughtering, during which time the temperature within the carcass reached 35.0 degrees C on 2 days; the average relative humidity was 79.0%. A low number of larvae survived up to day 14 after the pig was killed, when the carcass temperature reached 38.0 degrees C, but they lost their infectivity to laboratory mice. This result suggests that the larvae of *T. papuae* can survive in a tropical environment for a time, favouring their transmission to a new host in spite of the lack of a collagen capsule.

41 **Price RN, Hasugian AR, Ratcliff A, Siswantoro H, Purba HL, Kenangalem E, Lindegardh N, Penttinen P, Laihad F, Ebsworth EP, Anstey NM, Tjitra E.**
Clinical and pharmacological determinants of the therapeutic response to dihydroartemisinin-piperaquine for drug-resistant malaria.
Antimicrob Agents Chemother 2007 Nov;51(11):4090-4097. Epub 2007 Sep 10.

Dihydroartemisinin-piperaquine (DHP) is an important new treatment for drug-resistant malaria, although pharmacokinetic studies on the combination are limited. In Papua, Indonesia, we assessed determinants of the therapeutic efficacy of DHP for uncomplicated malaria. Plasma piperaquine concentrations were measured on day 7 and day 28, and the cumulative risk of parasitological failure at day 42 was calculated using survival analysis. Of the 598 patients in the evaluable population 342 had infections with *Plasmodium falciparum*, 83 with *Plasmodium vivax*, and 173 with a mixture of both species. The unadjusted cumulative risks of recurrence were 7.0% (95% confidence interval [CI]: 4.6 to 9.4%) for *P. falciparum* and 8.9% (95% CI: 6.0 to 12%) for *P. vivax*. After correcting for reinfections the risk of recrudescence with *P. falciparum* was 1.1% (95% CI: 0.1 to 2.1%). The major determinant of parasitological failure was the plasma piperaquine concentration. A concentration below 30 ng/ml on day 7 was observed in 38% (21/56) of children less than 15 years old and 22% (31/140) of adults ($p = 0.04$), even though the overall dose (mg per kg of body weight) in children was 9% higher than that in adults ($p < 0.001$). Patients with piperaquine levels below 30 ng/ml were more likely to have a recurrence with *P. falciparum* (hazard ratio [HR] = 6.6 [95% CI: 1.9 to 23]; $p = 0.003$) or *P. vivax* (HR = 9.0 [95% CI: 2.3 to 35]; $p = 0.001$). The plasma concentration of piperaquine on day 7 was the major determinant of the therapeutic response to DHP. Lower plasma piperaquine concentrations and higher failure rates in children suggest that dose revision may be warranted in this age group.

42 **Pritchard DI, Hooi DS, Brown A, Bockarie MJ, Caddick R, Quinnell RJ.**
Basophil competence during hookworm (*Necator americanus*) infection.
Am J Trop Med Hyg 2007 Nov;77(5):860-865.

A popular hypothesis to explain parasite survival in the presence of a pronounced T helper 2 phenotype in helminth-parasitized populations has been FcepsilonRI blockade by parasite-induced polyclonal IgE. To begin to test the hypothesis that FcepsilonRI-bearing cells would be refractory to activation in parasitized populations, we investigated basophil function in 43 individuals from a hookworm endemic area. Study individuals had high levels of total IgE and eosinophilia and a mean hookworm burden of 2,257 epg. Basophils from all members of this parasitized population were shown to release histamine to a number of agonists, including anti-IgE and a hookworm allergen, calreticulin. These data would indicate that FcepsilonRI blockade at the level of the basophil did not occur in this parasitized population despite the presence of possible immunologic blocking agents. This would suggest that this effector arm of the T helper 2 phenotype remains operative in infected populations.

43 **Proellocks NI, Kovacevic S, Ferguson DJ, Kats LM, Morahan BJ, Black CG, Waller KL, Coppel RL.**
Plasmodium falciparum Pf34, a novel GPI-anchored

rhoptry protein found in detergent-resistant microdomains.
Int J Parasitol 2007 Sep;37(11):1233-1241. Epub 2007 Apr 19.

Apicomplexan parasites are characterised by the presence of specialized organelles, such as rhoptries, located at the apical end of invasive forms that play an important role in invasion of the host cell and formation of the parasitophorous vacuole. In this study, we have characterised a novel *Plasmodium falciparum* rhoptry protein, Pf34, encoded by a single exon gene located on chromosome 4 and expressed as a 34kDa protein in mature asexual stage parasites. Pf34 is expressed later in the life cycle than the previously described rhoptry protein, Rhoptry Associated Membrane Antigen (RAMA). Orthologues of Pf34 are present in other *Plasmodium* species and a potential orthologue has also been identified in *Toxoplasma gondii*. Indirect immunofluorescence assays show that Pf34 is located at the merozoite apex and localises to the rhoptry neck. Pf34, previously demonstrated to be glycosyl-phosphatidyl-inositol (GPI)-anchored [Gilson, P.R., Nebl, T., Vukcevic, D., Moritz, R.L., Sargeant, T., Speed, T.P., Schofield, L., Crabb, B.S. (2006) Identification and stoichiometry of GPI-anchored membrane proteins of the human malaria parasite *Plasmodium falciparum*. Mol. Cell. Proteomics 5, 1286-1299.], is associated with parasite-derived detergent-resistant microdomains (DRMs). Pf34 is carried into the newly invaded ring, consistent with a role for Pf34 in the formation of the parasitophorous vacuole. Pf34 is exposed to the human immune system during infection and is recognised by human immune sera collected from residents of malaria endemic areas of Vietnam and Papua New Guinea.

44 **Reiff DM, Kaneko A, Taleo G, Amos M, Lum JK.**
Population structure and gene flow of *Anopheles farauti s.s.* (Diptera: Culicidae) among ten sites on five islands of Vanuatu: implications for malaria control.
J Med Entomol 2007 Jul;44(4):601-607.

The *Anopheles punctulatus* (Diptera: Culicidae) group is the main vector for malaria and bancroftian filariasis in Vanuatu. *Anopheles* larvae were collected from 10 localities on five islands of Vanuatu during the 2004 dry season for species identification as well as for estimating population structure and gene flow within and among islands. Species identification was determined using polymerase chain reaction-restriction fragment length polymorphism analysis of the internal transcribed spacer 2 region. Population structure and gene flow were examined by sequencing a portion of the ND4/ND5 region of the mitochondrial genome. Only one species of the *An. punctulatus* group, *An. farauti s.s.,* was identified, consistent with previous studies in Vanuatu. A nonrandom distribution of *An. farauti s.s.* lineages was observed with one cosmopolitan lineage shared by eight sites on all five islands and a preponderance of island-specific lineages (36/40), indicating the introduction of a single main lineage into Vanuatu followed by dispersal, diversification, and limited lineage exchange between islands. Network analysis suggests a possible second introduction of *An. farauti s.s.* into the northern islands of Gaua and Malekula. Gene flow was high on three of the five islands, whereas Tanna and Santo have significant population structure. Among islands, gene flow was limited, indicating active mosquito dispersal only over short distances and a paucity of passive human-mediated dispersal over long distances. Minimal risk of active dispersal among these islands indicates that vector control can be effectively initiated at the island level within the archipelago of Vanuatu.

45 **Ricciardelli LA, McCabe MP, Mavoa H, Fotu K, Goundar R, Schultz J, Waqa G, Swinburn BA.**
The pursuit of muscularity among adolescent boys in Fiji and Tonga.
Body Image 2007 Dec;4(4):361-371. Epub 2007 Sep 20.

The desire for muscularity is tied to Western views of the male gender role, which prescribe that men be strong, physically fit and athletically successful. Although these ideals have been primarily studied among Western adolescent boys, there is emerging evidence that the same ideals are valued and promoted among males from the Pacific Islands. The aim of the present study was to examine body image concerns associated with muscularity and the reasons for these concerns among Fijian and Tongan adolescent boys. Semi-structured interviews were conducted with 24 Indigenous Fijian, 24 Indo-Fijian, and 24 Tongan boys aged between 13 and 20 years. A thematic analysis of boys' narratives showed that the pursuit of muscularity was a dominant theme for many boys. Boys' reasons for pursing muscularity included the attainment of strength and fitness, sporting performance, physical work, dominance, and health. These findings are examined in relation to previous research with Western adolescent boys.

46 **Ryan CE, Gare J, Crowe SM, Wilson K, Reeder JC, Oelrichs RB.**
The heterosexual HIV type 1 epidemic in Papua New Guinea is dominated by subtype C.
AIDS Res Hum Retroviruses 2007 Jul;23(7):941-944.

Papua New Guinea is in the midst of a generalized HIV epidemic. As part of a larger behavioral survey aiming to further characterize the HIV epidemic occurring in PNG, samples were collected from 1175 participants from seven different provinces. Seventy-one (6%) of these samples were HIV-1 positive, and 35 (49%) successfully underwent a double nested RT-PCR that was designed to amplify the C2-V4 region of the HIV-1 envelope. Sequence analysis showed that 33 (94%) samples were subtype C and the remaining 2 (6%) were subtype B. Further phylogenetic analysis demonstrated that there was no province-specific clustering among the samples and that within the global pandemic, PNG subtype C isolates most closely resembled those from East Africa.

47 **Sharma R, Maimanuku LR, Morse Z, Pack AR.**
Preterm low birth weights associated with periodontal disease in the Fiji Islands.
Int Dent J 2007 Aug;57(4):257-260.

AIM: To determine any association between pre-term low birth weight (PTLBW) neonates and periodontal disease during the mother's pregnancy. DESIGN: A multi-centered prospective case cohort study. SETTING: Ante-natal clinics at the Colonial War Memorial and Lautoka Hospitals, Fiji from 1st January to 30th June 2004. PARTICIPANTS: 670 multiethnic pregnant women. METHODS: Participants were interviewed to identify confounding variables – medical conditions, smoking, alcohol

consumption, maternal age and history of preterm birth. Oral examination was conducted and included the Community Periodontal Index of Treatment Needs (CPITN). Delivery outcome was recorded for each woman. RESULTS: The mean age of participants was 25.80 ± 5.56 years. 1.9% (n=13) women delivered preterm babies. More than 50% of this group displayed moderate to severe periodontitis compared with 13% of women who had a normal delivery. Preterm birth was also associated with the mother having had a previous preterm birth and who was more likely to be Indo-Fijian ($p < 0.01$). There was no significant association with where the mother lived; however, rural women with PTLBW babies had more severe periodontal disease ($p = 0.0001$). CONCLUSION: There is a highly significant association between pre-term birth and moderate to severe periodontal disease ($p = 0.0001$).

48 **Stutchfield BM, Jagilly R, Tulloh BR.**
Second opinions in remote surgical practice using email and digital photography.
ANZ J Surg 2007 Nov;77(11):1009-1012.

BACKGROUND: Email offers the opportunity to improve communication between surgeons across the world. This experimental study aimed to assess the feasibility of obtaining clinical opinions by email and digital photography in remote surgical practice. METHODS: Over a 3-week period, all adult general surgical cases with a visual component to their condition admitted to a remote developing-world hospital were invited to participate. Clinical details and digital images were emailed to a UK general surgeon who consulted specialist colleagues if required and emailed back a suggested diagnosis and management plan, rating the confidence with which these were made on a five-point scale. The concordance between diagnoses and management plans from each centre were rated by three independent general surgeons. RESULTS: In this prospective study of 32 patients, 56% of diagnoses and 78% of management plans were made by the UK surgeons with 'high' or 'total' confidence. Causes of low diagnostic confidence included vague swellings and low-resolution X-ray images. Diagnostic and management concordance between centres was adjudged 'high' or 'total' in 88 and 43% of cases, respectively. CONCLUSION: Obtaining second opinions using email and digital photography is feasible in adult general surgery, but its efficacy is limited in cases where image resolution or non-visual clues are important.

49 **Suwanarusk R, Russell B, Chavchich M, Chalfein F, Kenangalem E, Kosaisavee V, Prasetyorini B, Piera KA, Barends M, Brockman A, Lek-Uthai U, Anstey NM, Tjitra E, Nosten F, Cheng Q, Price RN.**
Chloroquine resistant *Plasmodium vivax*: in vitro characterisation and association with molecular polymorphisms.
PLoS ONE 2007 Oct 31;2(10):e1089.

BACKGROUND: Treatment failure of chloroquine for *P. vivax* infections has reached high levels in the eastern provinces of Indonesia; however, in vitro characterization of chloroquine resistance and its associated molecular profile have yet to be determined. METHODS: Using a modified schizont maturation assay we investigated the in vitro chloroquine susceptibility profile and molecular polymorphisms of *P. vivax* isolates collected from Papua, Indonesia, where high levels of clinical chloroquine treatment failure have been reported, and from Thailand, where chloroquine treatment is generally effective. RESULTS: The geometric mean chloroquine IC(50) for *P. vivax* isolates from Papua (n = 145) was 312 nM [95%CI: 237-411 nM] compared to 46.8 nM [95%CI: 34.7-63.1 nM] from Thailand (n = 81); $p<0.001$. Correlating with the known clinical efficacy of the area, a cut-off for chloroquine resistance was defined as 220nM, a level exceeded in 13.6% (11/81) of Thai isolates and 65% (94/145) of Papuan isolates; $p<0.001$. Several sequence polymorphisms in *pvcrt-o* and *pvmdr1*, and difference in *pvmdr1* copy number were identified. A Y976F mutation in *pvmdr1* was present in 96% (123/128) of Papuan isolates and 25% (17/69) of Thai isolates; $p<0.001$. Overall, the geometric mean chloroquine IC(50) in isolates with the Y976F mutation was 283 nM [95%CI: 211-379], compared to 44.5 nM [95%CI: 31.3-63.4] in isolates with the wild type; $p< 0.001$. *Pvmdr1* amplification occurred in 23% (15/66) of Thai isolates compared to none (0/104) of Indonesian isolates ($p<0.001$), but was not associated with increased chloroquine resistance after controlling for geographical location. CONCLUSIONS: In vitro susceptibility testing of *P. vivax* discriminates between populations with differing levels of clinical efficacy of chloroquine. The *pvmdr1* polymorphism at Y976F may provide a useful tool to highlight areas of emerging chloroquine resistance, although further studies defining its clinical correlates are needed.

50 **Tefuarani N, Vince J, Hawker R, Nunn G, Lee R, Crawford M, Kevau IH.**
Operation Open Heart in PNG, 1993-2006.
Heart Lung Circ 2007 Oct;16(5):373-377. Epub 2007 Jul 12.

OBJECTIVE: To report on the 'Operation Open Heart' (OOH) cardiac surgical program in Papua New Guinea (PNG). To document the short-term surgical outcome, the experience gained and the skill transfer from the visiting team members to their PNG counterparts. METHODOLOGY: Analysis of the database compiled from the records of the patients who were operated on by the visiting cardiothoracic surgical team. RESULTS: Four hundred and seventy patients from all regions of the country received operations. Three hundred and thirty seven (72%) were children less than 12 years of age, 39 (8%) were between 12 and 18 years of age and 263 (56%) were females. One hundred and eighty five (40%) patients had open heart procedures. Complications were unremarkable and the short-term mortality was 1.9%. Clinical skills were transferred to, and experience was gained by national anaesthetists, surgeons, paediatricians, physicians and nurses from intensive and full nursing care units and the operating theatre. CONCLUSIONS: The program not only achieved a higher annual operation rate than previous programs but also had a lower mortality rate. It achieved its objective of service delivery and, to a considerable extent, its objective of skill transfer. There now is an established and active group of PNG doctors and nurses with the skills, experience and confidence to perform patent ductus repair safely and efficiently. The program is cheaper than its predecessors, and is less disruptive for parents, patients and families.

51 **Tovosia S, Chen PH, Ko AM, Tu HP, Tsai PC, Ko YC.**
Prevalence and associated factors of betel quid use

in the Solomon Islands: a hyperendemic area for oral and pharyngeal cancer.
Am J Trop Med Hyg 2007 Sep;77(3):586-590.

Chewing betel quid is a popular habit in tropical areas. It is also a known fact that oral cancer is one of the most common cancers in the Solomon Islands, where betel quid chewing is prevalent. This study explores the prevalence of betel quid chewing in the Solomon Islands and related sociodemographic factors. A community-based survey was designed for the entire Solomon Islands people. Information on sociodemographic characteristics, habits of betel quid chewing, smoking, and drinking was collected by trained interviewers with a standard questionnaire. The prevalence of betel quid chewers, smokers, and drinkers was 76.8%, 53.0%, and 36.8%, respectively. Betel quid use was closely associated with smoking (aOR = 3.95; 95% CI: 1.87-8.33). Seventh-Day Adventist subjects were less likely to chew betel quid (aOR = 0.08; 95% CI: 0.04-0.18). Efforts to reduce habitual betel quid consumption and smoking might be of benefit in reduction of oral cancer incidence.

52 **Trnka S.**
Languages of labor: negotiating the 'real' and the relational in Indo-Fijian women's expressions of physical pain.
Med Anthropol Q 2007 Dec;21(4):388-408.

Medical personnel in public clinics in Fiji routinely contend that state-funded medical resources are misallocated on patients who complain of, but do not actually experience, physical pain. Frequently, these patients are identified as being Indo-Fijian women (ie, women of South Asian origin in Fiji). In this article, I examine clinical interactions between medical staff and female Indo-Fijian patients to demonstrate how 'real' and 'unreal' pain are distinguished in the clinical setting and to indicate some of the roles clinical encounters play in community processes that ascribe alternative meanings to physical pain. Focusing on how both physicians and women patients foster certain interpretations of physical pain over others, I argue that the category of 'unreal' pain, as employed by Fiji's physicians, consists of pain that medical professionals consider to be induced by psychological or physical, work-related stresses. I then show how Indo-Fijian women engage in a complementary but distinct discourse that emphasizes links between physical labor and pain and suggests that, in some cases, expressions of physical pain are as much an idiom of pride as an idiom of distress.

53 **Warner JM, Pelowa DB, Gal D, Rai G, Mayo M, Currie BJ, Govan B, Skerratt LF, Hirst RG.**
The epidemiology of melioidosis in the Balimo region of Papua New Guinea.
Epidemiol Infect 2007 Aug; 22:1-7. [Epub ahead of print]

The distribution of *Burkholderia pseudomallei* was determined in soil collected from a rural district in Papua New Guinea (PNG) where melioidosis had recently been described, predominately affecting children. In 274 samples, 2.6% tested culture-positive for *B. pseudomallei*. Pulsed-field gel electrophoresis using *Spe*I digests and rapid polymorphic DNA PCR with five primers demonstrated a single clone amongst clinical isolates and isolates cultured from the environment that was commonly used by children from whom the clinical isolates were derived. We concluded that individuals in this region most probably acquired the organism through close contact with the environment at these sites. *Burkholderia thailandensis*, a closely related *Burkholderia* sp. was isolated from 5.5% of samples tested, an observation similar to that of melioidosis-endemic areas in Thailand. This is the first report of an environmental reservoir for melioidosis in PNG and confirms the Balimo District in PNG as melioidosis endemic.

54 **Watters DA.**
Loch Ness, special operations executive and the first surgeon in paradise: Robert Kenneth Wilson (26.1.1899-6.6.1969).
ANZ J Surg 2007 Dec;77(12):1053-1057.

Lieutenant Colonel Robert Kenneth Wilson (1899-1969) was a surgeon who fought in both world wars and joined the Special Operations Executive parachuting behind enemy lines into Holland, France and Borneo, the last mission being with Australian forces (Semut II). He was an expert on firearms and gave opinion on ballistics at the Old Bailey during the 1930s. He also wrote a definitive text on automatic pistols with editions published in 1943 and 1975. He was an Edinburgh Fellow (1926), who had a practice in general surgery and gynaecology in Queen Anne Street during the 1930s. He took the famous 1934 'surgeon's photo' of the Loch Ness monster that was not admitted to be a hoax until 1994. After World War II, he became the first surgical specialist to work in the public service of the then Territory of Papua and New Guinea (1950-1956), where he wrote several papers on surgical topics. He married Gwen (1924), the daughter of Henrietta Gulliver, an Australian painter. They had two sons, Richard and Phillip. After practice he retired to Melbourne, where he died of carcinoma of the oesophagus.

55 **Wildig J, Mueller I, Kiniboro B, Maraga S, Siba P, Cossart Y.**
Seroprevalence of antibodies to parvovirus B19 among children in Papua New Guinea.
Am J Trop Med Hyg 2007 Aug;77(2):354-357.

Parvovirus B19 (B19) is a common childhood infection that has recently been found to be associated with severe anemia in Papua New Guinean children. Population surveys were performed in 15 villages in Maprik District, East Sepik Province, Papua New Guinea in 2005. Plasma samples collected from children less than 10 years of age were tested for IgM and IgG antibodies to B19 by enzyme immunoassay. The prevalence of IgG antibody to B19 was 53.8% and ranged from 20% in those less than one year of age to 85.5% in those nine years of age. Considerable variation in IgG prevalence was observed between study areas, indicating complex patterns of transmission. Prevalence of IgM antibody to B19 was 1.5%. This study confirms that B19 infection is common among children in this tropical area. With 19.5% of children one year of age showing evidence of previous infection, any preventive measures should be targeted at the very young.

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3 **Garner PA, Hill G.** Brainwashing in tuberculosis management. *PNG Med J* 1985;28:291-293.

4 **Cochrane RG.** A critical appraisal of the present position of leprosy. In: Lincicome DP, ed. International Review of Tropical Medicine. New York: Academic Press, 1961:1-42.

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