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EDITORIAL

Malaria in Papua New Guinea 2000-2013: back from the brink, but where to now?

After a long drought in funding for malaria control in Papua New Guinea (PNG), the year 2004 marked the beginning of renewed efforts to curb malaria in this highly endemic setting. 6 years after the launch of the global Roll Back Malaria Partnership (1), the Government of PNG secured a first grant of 16 million USD from the Global Fund to Fight AIDS, Tuberculosis and Malaria (Round 3 grant) to support its malaria control efforts (2). Together with partners in the provinces and districts, and in collaboration with PNG-based Rotarians Against Malaria (RAM), the National Department of Health used these funds to re-establish a vector control effort based on the free nation-wide distribution of long-lasting insecticidal nets (LLINs). Between 2005 and 2009, a total of 2.4 million LLINs were distributed, resulting in a national coverage of 65% of households owning at least one LLIN (3).

Unfortunately, for most parts of PNG the impact of this initial LLIN distribution could not easily be assessed. Systematic pre-intervention baseline data on mosquito net coverage and population parasite rates are lacking and data on clinical cases derived from the National Health Information System are based mainly on unconfirmed clinical diagnoses and are hence difficult to interpret (4). In 2008, the Papua New Guinea Institute of Medical Research (PNGIMR) was contracted to carry out a scientific evaluation of the Global Fund Round 3 program to provide some of the missing evidence. The Institute conducted a national malaria survey and established five sentinel sites in areas that were yet to receive the first LLINs. Despite rather modest rates of actual LLIN use (33% overall and approximately 40% by children aged <5 years and by pregnant women) (3), the distribution resulted in very significant reductions in mosquito biting rates, eg, 68-95% around Dreikikir, East Sepik Province (5), and a notable drop in the number of malaria episodes in many health facilities. In the sentinel sites, for example, the number of patients presenting at the local health centres was reduced by up to 80% within one year of the LLIN distribution (PNGIMR, unpublished data). Similarly, in two cohorts of children aged <5 years followed in 2006-2007 pre-LLIN (6) and 2009 post-LLIN distribution (7), the incidence of *Plasmodium falciparum* and *P. vivax* malaria episodes was reduced by 73% and 54%, respectively (PNGIMR, unpublished data). In two cross-sectional surveys conducted by PNGIMR in Madang Province before (2006) and after (2010) the LLIN distribution, the prevalence of *P. falciparum and P. vivax* infection assessed by polymerase chain reaction (PCR) decreased from 39% to 18% and from 32% to 13%, respectively (8).

In 2009, PNG obtained a further grant of 120 million USD from the Global Fund (Round 8 grant) both to continue the LLIN distribution program and to improve case management by introducing arthemeter-lumefantrine (AL) as first-line antimalarial treatment plus parasitological diagnosis by rapid diagnostic test (RDT) or light microscopy. In addition, a behaviour change campaign is being conducted to support the introduction of the new treatment and increase LLIN use. National household surveys conducted by PNGIMR in 2008-2009 and 2010-2011 found a reduction in prevalence of infection nation-wide from 18.2% to 6.7% (9) alongside an increase in ownership and use of LLINs from 65% to 82% and from 33% to 48%, respectively (10). There were, however, substantial differences in *Plasmodium* spp. infections between geographical regions. While virtually absent in the highlands and at around 5% in the Southern Region, the prevalence of infection was still higher in the Momase and Islands Regions (8-14%), with little change in prevalence in Momase between 2008-2009 and 2010-2011 (9).

Learning from the past

The reductions in the burden of malaria observed since 2005 are impressive – and a reason for celebration! They are even more impressive because they were achieved under often difficult conditions and were tackled by a combined effort of all stakeholders, including the national and provincial governments,

church health services and non-governmental organizations as well as the private sector.

At the same time it is important to view the current success in the context of earlier malaria control efforts undertaken in PNG. Particularly interesting is a comparison with the strategies and achievements of the malaria elimination campaigns in the 1960s and 1970s (reviewed in the PNG Medical Journal special issue 'Focus on Malaria' in 1974 (11)). In selected areas of PNG, focal attempts to control malaria started in the early 20th century using methods such as environmental modification (eg, Samarai Island and Wahgi Valley), larviciding (Rabaul), larvivorous fish (Kavieng) or house spraying with dichloro-diphenyl-trichloroethane (DDT) (12). Yet it was a pilot project with DDT at Maprik (East Sepik) in 1957 that laid the foundation for country-wide indoor residual spraying (IRS) campaigns complemented with mass drug administration (chloroquine + pyrimethamine) (12,13). By the early 1970s, about 70% of the PNG population were covered by IRS with DDT (13,14). In the densely populated highlands, malaria transmission had been interrupted and the program had moved from an 'attack' to a 'consolidation' phase (15). In Milne Bay and the Islands Region, prevalence rates had dropped rapidly following the initiation of IRS and in many areas (including Gazelle, Kavieng and Namatanai, Buka/Kieta, Talasea, Manus and Misima Island) prevalence rates were at or below 1-3% after 5-10 years of IRS control (12). It was, however, much more difficult to achieve significant impact in highly endemic areas in the Momase Region. IRS with DDT had a significant initial impact in the Maprik area, reducing prevalence rates in children 0-4 years of age from >90% in 1957 to <25% in 1962 (ie, after five years of IRS) and the overall parasite rate to 10% in 1964 (ie, after seven years of IRS) (12). At the same time, however, significant changes in vector behaviour towards earlier and more outdoor biting had been observed (16). Worryingly, similar changes in mosquito biting patterns are again being detected following LLIN distribution (17).

In summary, the trends in malaria observed over the last ten years closely resemble the situation in the late 1960s/early 1970s following the rollout of the IRS campaign. It is therefore important also to remember what happened to malaria in PNG from the early 1970s to 2000. Despite the initial successes of the program, populations grew tired of the continued spraying. There appeared to be a lack of appreciation of the benefits of the program in part of the population as well as insensitivity of campaign implementers towards local perceptions of IRS (18). This was aggravated by some evidence of toxic effects of the insecticide on domestic animals and increases in the population of caterpillars of the moth *Herculia negrivitta*, which would damage the sago-palm-thatched roofs, thus decreasing the life span of traditionally built houses (12,19). Only a few years into the program, refusals increased; in East New Britain, for example, 25% of houses could not be sprayed in 1970 while eight years earlier coverage had been 100% (12). Around the same time, chloroquine resistance emerged and within a few years resistance of malaria parasites against chloroquine, amodiaquine and sulphadoxine-pyrimethamine (SP) started to spread (20,21). A review of the malaria program conducted in 1983 concluded that the program was not good value for money and the newly independent PNG should better use its sparse resources to tackle other health problems (22). As a consequence, the program was scaled back and spraying was stopped in the early 1980s, except for control of epidemic outbreaks in the highlands.

The gradual decline of the malaria control program had immediate consequences: malaria rapidly rebounded (23,24). At Maprik, prevalence started to increase again, reaching 32-68% in 1967-1968 (12,25). Similar rebounds were documented for Pagwi and Aitape (East Sepik) and Karimui (Simbu) in the highlands (12,24). By the early 1990s malaria had once again reached pre-IRS levels (26-28) and despite a modest drop in burden following the addition of SP to chloroquine (29), prevalence rates in the Momase Region reached over 50% by light microscopy and up to 85% by PCR in some villages (30-32) and children aged <5 years experienced up to 5 malaria episodes per year (6). In the Highlands Region, malaria was endemic in all areas below 1500 m and epidemics were once again common (33).

The way forward

At the end of the Global Fund Round 8 support (October 2014), PNG authorities and their partners will have a fundamental choice to make: either to ensure a continuation of the successful investment in malaria control,

reduce the malaria burden further, and possibly even consider the long-term goal of malaria elimination, or to be satisfied with what has been achieved, let malaria slip down the priorities, and risk a rebound comparable to that observed in the 1980s.

With the control tools currently available (34), the complexity of the PNG setting, and the current state of the PNG health system, nation-wide elimination of malaria (ie, the complete interruption of local transmission) may be unrealistic in the near future. The slide positivity of fever cases remains in most places well above the indicative 5% threshold suggested by the World Health Organization (WHO) as the criterion for entering the pre-elimination phase (35). A more achievable strategy would be to consolidate and further extend the gains made so far based on an intensified control approach. The ongoing fluctuation in clinical cases and slight rebounds that are being observed in different parts of PNG (MWH, IM and others, work in progress) indicate that even to maintain the status quo further intensification of control efforts will be inevitable. Stabilizing malaria transmission at a very low level may set the stage for focal sub-national elimination initiatives, for example a malaria-free Highlands Region or malaria elimination from certain islands in the medium term. Yet, unless intense control is sustained in the heartland of transmission where the burden remains greatest, such peripheral gains will be hard to maintain and investments may be difficult to justify.

Important decisions will therefore need to be made in the coming years. As PNG's gross domestic product (GDP) increases as a result of the current resource boom and global donor support for health may dwindle (36), will PNG be willing to make use of its own resources, financial as well as human, to continue and strengthen the fight against malaria? At the moment, funding for the National Malaria Control Program (NMCP) originates almost exclusively from the Global Fund, complemented with money from other donors. The current domestic financial contribution to malaria control is minimal (37).

Decisions on program directions need to be made based on a solid understanding of the current malaria situation and the impact achieved by the ongoing control activities. In addition, new and improved strategies will need to be evaluated so that they may be considered as additional control tools in PNG. To monitor the impact of the Global Fund Round 8 malaria program and provide such essential data for national policy and local decision-making, the PNGIMR has established a nation-wide evaluation program that includes both regular national household and health facility surveys as well as continuous monitoring of trends in a number of sentinel sites.

This focus issue of the PNG Medical Journal focuses on key outcomes of the first phase of this evaluation, which is described in detail in the article by Hetzel et al. (38). 5 studies provide insights into different challenges in malaria case management in PNG. Through interviews with malaria-affected individuals or their caregivers Angwin et al. (39) show an apparent preference for convenient, home-based responses to perceived malaria infection in the first instance, including the use of antimalarials obtained from an existing or easily accessible source. A variety of reasons for these treatment choices were given by participants. While these findings confirm that many malaria episodes may never be detected by the formal health system, they also highlight the need and potential for exploring different approaches for providing appropriate treatment for malaria episodes close to people's homes. When patients with a febrile illness do visit a health centre, the proportion that is actually sick with malaria varies widely between different PNG settings. In a surveillance of all patients presenting to four health centres in the Momase Region and one in Western Province, Hetzel et al. (40) found that, while overall just about half of all fevers were due to malaria, this proportion varied from 75% in Dreikikir, East Sepik Province, to only 2% in Wipim, Western Province. Bande et al. (41) further investigated the findings from Wipim and found that 11% of non-malaria fevers were due to a dengue 1 infection, but the vast majority were of unknown origin. The variation in malaria-attributable fevers underlines the importance of implementing parasitological diagnosis for all suspected malaria cases and of only treating test-positive cases with antimalarials while investigating other causes in the remaining patients. Unfortunately, as shown by Kurumop et al. (42), diagnostic capacity was low in Papua New Guinean health facilities before the full rollout of the new National Malaria Treatment Protocol and access to recommended first-line antimalarial

medication was variable. Previously published results showed that, as a consequence, adherence to the new treatment guidelines was poor (43). However, with the nation-wide rollout of the new treatment strategy starting in 2010, diagnostic capacities substantially improved and RDTs and AL are now available in most health centres, yet still only in a minority of aid posts (44). A challenge remains in the clinical management of cases with *P. vivax* malaria because of the hypnozoites that cause relapses and contribute significantly to the disease burden (7). Betuela et al. discuss the importance of primaquine for treating *P. vivax* and the challenges encountered when implementing primaquine guidelines in clinical practice in PNG (45). One of these challenges is glucose-6-phosphate-dehydrogenase (G6PD) deficiency, which can lead to serious side-effects in combination with primaquine treatment.

The investigation of the composition of parasite species is one aspect discussed by Koepfli et al. (46) in their review of molecular epidemiology approaches. As the malaria burden is reduced, more sophisticated methods are required to detect parasites and investigate the complex and changing epidemiology of malaria in PNG. A potential threat to the current program may lie in the emergence of *Anopheles* mosquitoes resistant to insecticides used on mosquito nets. Yet, after showing earlier that there is no resistance to pyrethroids, the insecticide class used on the distributed LLINs, in PNG so far (47), Katusele et al. (48) now provide evidence that the LLINs used in PNG remain effective for up to 5 years and that proper net care can extend the duration of efficacy.

Together with earlier studies exploring reasons for non-use of LLINs (49) and investigating the use of text-message reminders to improve health worker adherence to malaria treatment guidelines (50), this array of studies provide essential baseline evidence against which the ongoing progress of the PNG NMCP can be evaluated and by which future policy decisions should be guided.

Dedication

This focus issue of the PNG Medical Journal is dedicated to five young staff members of the PNGIMR who disappeared from West New Britain Province in August 2011, never to be seen again. While travelling to a study village sampled for the 2010-2011 national malaria survey, the boat carrying Gibson Gideon, Tania Oakiva, Lydia Petrus, Leonard Vavana, George Dogoya and two local men as skipper and crew was hijacked at sea. The dead body of the skipper was later discovered in Manus Province but, to date, no trace has been found of the others. At the end of this issue, the PNGIMR team pay tribute to their missing friends and colleagues (51).

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REFERENCES

1 **Nabarro DN, Tayler EM.** The “roll back malaria”

campaign. *Science* 1998;280:2067-2068.
2 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Community-based Malaria Prevention and Control in Papua New Guinea – Nationwide Insecticide-treated Nets (ITN) Distribution and Expansion of Confirmed Diagnosis and Appropriate Treatment of Malaria. Grant Number: PNG-304-G01-M. Geneva: Global Fund, 2004. http://portfolio.theglobalfund.org/en/Grant/Index/PNG-304-G01-M
3 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.
4 **World Health Organization, Roll Back Malaria, United Nations Children's Fund.** World Malaria Report 2005. Geneva: World Health Organization, Roll Back Malaria, UNICEF, 2005.
5 **Reimer LJ, Thomsen EK, Tisch DJ, Henry-Halldin CN, Zimmerman PA, Baea ME, Dagoro H, Susapu M, Hetzel MW, Bockarie MJ, Michael E, Siba PM, Kazura JW.** Insecticidal bed nets and filariasis transmission in Papua New Guinea. *N Engl J Med* 2013;369:745-753.
6 **Lin E, Kiniboro B, Gray L, Dobbie S, Robinson L, Laumaea A, Schöpflin S, Stanisic D, Betuela I, Blood-Zikursh M, Siba P, Felger I, Schofield L, Zimmerman P, Mueller I.** Differential patterns of infection and disease with *P. falciparum* and *P. vivax* in young Papua New Guinean children. *PLoS One* 2010;5:e9047.
7 **Betuela I, Rosanas-Urgell A, Kiniboro B, Stanisic DI, Samol L, de Lazzari E, Del Portillo HA, Siba P, Alonso PL, Bassat Q, Mueller I.** Relapses contribute significantly to the risk of *Plasmodium vivax* infection and disease in Papua New Guinean children 1-5 years of age. *J Infect Dis* 2012;206:1771-1780.
8 **Koepfli C, Robinson LJ, Rarau P, Salib M, Sambale N, Wampfler R, Betuela I, Nuitragool W, Barry AE, Siba P, Felger I, Mueller I.** Blood-stage parasitaemia and age determine *Plasmodium falciparum* and *P. vivax* gametocytaemia in Papua New Guinea. *PLoS One* 2014;10:e0126747.
9 **World Health Organization.** World Malaria Report 12. Geneva: World Health Organization, 2012.
10 **Hetzel MW, Choudhury AAK, Pulford J, Ura Y, Whittaker M, Siba PM, Mueller I.** Progress in mosquito net coverage in Papua New Guinea. *Malar J* 2014;13:242.
11 **Papua New Guinea Medical Journal.** Focus on Malaria. *PNG Med J* 1974;17:1-125.
12 **Ewers WH, Jeffrey WT.** Parasites of Man in Niugini. Milton, Queensland: Jacaranda Press, 1971.
13 **Parkinson AD.** Malaria in Papua New Guinea 1973. *PNG Med J* 1974;17:8-16.
14 **Van Dijk WJOM, Parkinson AD.** Epidemiology of malaria in New Guinea. *PNG Med J* 1974;17:17-21.
15 **Black RH.** Malaria control. Letter. *PNG Med J* 1969;12:142-143.
16 **Spencer TET, Spencer M, Venters D.** Malaria vectors in Papua New Guinea. *PNG Med J* 1974;17:22-30.
17 **Koimbu G, Katusele M, Hetzel MW, Burkot T, Kazura J, Siba PM, Reimer LJ.** Assessing the biological coverage of long lasting insecticidal nets in Papua New Guinea. Abstract 14 in Abstracts and Program of the Forty-ninth Annual Medical Symposium of the Medical Society of Papua New Guinea, Lae, 1-6 Sep 2013:53-54.
18 **Zigas V, van Delden J.** Communication and community response in health programmes with special reference to Papua New Guinea. *PNG Med J* 1974;17:80-83.
19 **Moorhouse DE.** Enhanced damage to attap roofs by the caterpillars of *Herculia nigrivitta*, following house-spraying with DDT. *Med J Malaysia* 1964;19:56-57.
20 **Grimmond TR, Donovan KO, Riley I.** Chloroquine resistant malaria in Papua New Guinea. *PNG Med J* 1976;19:184-185.
21 **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;77:947-954.
22 **Papua New Guinea Department of Health.** Papua New Guinea National Health Plan 1986-1990. Port Moresby: Department of Health, 1986.
23 **Müller I, Bockarie M, Alpers M, Smith T.** The epidemiology of malaria in Papua New Guinea. *Trends Parasitol* 2003;19:253-259.
24 **Mueller I, Tulloch J, Marfurt J, Hide R, Reeder JC.** Malaria control in Papua New Guinea results in complex epidemiological changes. *PNG Med J* 2005;48:151-157.
25 **Desowitz RS, Spark RA.** Malaria in the Maprik area of the Sepik region, Papua New Guinea: 1957-1984. *Trans R Soc Trop Med Hyg* 1987;81:175-176.
26 **Genton B, Al-Yaman F, Beck HP, Hii J, Mellor S, Narara A, Gibson N, Smith T, Alpers MP.** The epidemiology of malaria in the Wosera area, East Sepik Province, Papua New Guinea, in preparation for vaccine trials. I. Malariometric indices and immunity. *Ann Trop Med Parasitol* 1995;89:359-376.
27 **Genton B, Al-Yaman F, Beck HP, Hii J, Mellor S, Rare L, Ginny M, Smith T, Alpers MP.** The epidemiology of malaria in the Wosera area, East Sepik Province, Papua New Guinea, in preparation for vaccine trials. II. Mortality and morbidity. *Ann Trop Med Parasitol* 1995;89:377-390.
28 **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.
29 **Kasehagen LJ, Mueller I, McNamara DT, Bockarie MJ, Kiniboro B, Rare L, Lorry K, Kastens W, Reeder JC, Kazura JW, Zimmerman PA.** Changing patterns of *Plasmodium* blood-stage infections in the Wosera region of Papua New Guinea monitored by light microscopy and high throughput PCR diagnosis. *Am J Trop Med Hyg* 2006;75:588-596.
30 **Mueller I, Widmer S, Michel D, Maraga S, McNamara D, Kiniboro B, Sie A, Smith T, Zimmerman P.** High sensitivity detection of *Plasmodium* species reveals positive correlations between infections of different species, shifts in age distribution and reduced local variation in Papua New Guinea. *Malar J* 2009;8:41.
31 **Schultz L, Wapling J, Mueller I, Ntsuke PO, Senn N, Nale J, Kiniboro B, Buckee CO, Tavul L, Siba PM, Reeder JC, Barry AE.** Multilocus haplotypes reveal variable levels of diversity and population structure of *Plasmodium falciparum* in Papua New Guinea, a region of intense perennial transmission. *Malar J* 2010;9:e336.
32 **Arnott A, Barnadas C, Senn N, Siba P, Mueller I, Reeder JC, Barry AE.** High genetic diversity of *Plasmodium vivax* on the north coast of Papua New Guinea. *Am J Trop Med Hyg* 2013;89:188-194.
33 **Betuela I, Maraga S, Hetzel MW, Tandrapah T, Sie A, Yala S, Kundi J, Siba P, Reeder JC, Mueller I.** Epidemiology of malaria in the Papua New Guinean highlands. *Trop Med Int Health* 2012;17:1181-1191.

34 **Alonso PL, Tanner M.** Public health challenges and prospects for malaria control and elimination. *Nat Med* 2013;19:150-155.

35 **World Health Organization.** Malaria elimination: a field manual for low and moderate endemic countries. Geneva: World Health Organization, 2007.

36 **Institute for Health Metrics and Evaluation.** Financing Global Health 2012: The End of the Golden Age? Seattle, Washington: Institute for Health Metrics and Evaluation, 2012.

37 **Papua New Guinea Department of Health.** Papua New Guinea Malaria Programme Review 2013. Port Moresby: Department of Health, 2013.

38 **Hetzel MW, Pulford J, Maraga S, Barnadas C, Reimer LJ, Tavul L, Jamea-Maiasa S, Tandrapah T, Maalsen A, Makita L, Siba PM, Mueller I.** Evaluation of the Global Fund-supported National Malaria Control Program in Papua New Guinea, 2009-2014. *PNG Med J* 2014;57:7-29.

39 **Angwin A, Hetzel MW, Mueller I, Siba PM, Pulford J.** A qualitative study of how affected individuals or their caregivers respond to suspected malaria infection in rural Papua New Guinea. *PNG Med J* 2014;57:30-38.

40 **Hetzel MW, Paul S, Benjamin L, Makita L, Mueller I, Siba PM.** The proportion of fevers attributable to malaria varies significantly between sites in Papua New Guinea. *PNG Med J* 2014;57:39-51.

41 **Bande G, Hetzel MW, Iga J, Barnadas C, Mueller I, Siba PM, Horwood PF.** An investigation into febrile illnesses of unknown aetiology in Wipim, Papua New Guinea. *PNG Med J* 2014;57:52-58.

42 **Kurumop SF, Pulford J, Mueller I, Siba PM, Hetzel MW.** Diagnostic capacity and antimalarial availability in Papua New Guinea before the introduction of a revised national malaria treatment protocol. *PNG Med J* 2014;57:59-67.

43 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

44 **Pulford J, Kurumop SF, Ura Y, Siba PM, Mueller I, Hetzel MW.** Malaria case management in Papua New Guinea following the introduction of a revised treatment protocol. *Malar J* 2013;12:433.

45 **Betuela I, Robinson LJ, Hetzel MW, Laman M, Siba PM, Bassat Q, Mueller I.** Primaquine treatment for *Plasmodium vivax* – an essential tool for malaria control and elimination in Papua New Guinea. *PNG Med J* 2014;57:68-74.

46 **Koepfli C, Barry AE, Javati S, Timinao L, Nate E, Mueller I, Barnadas C.** How molecular epidemiology studies can support the National Malaria Control Program in Papua New Guinea. *PNG Med J* 2014;57:75-85.

47 **Keven JB, Henry-Halldin CN, Thomsen EK, Mueller I, Siba PM, Zimmerman PA, Reimer LJ.** Pyrethroid susceptibility in natural populations of the *Anopheles punctulatus* group (Diptera: Culicidae) in Papua New Guinea. *Am J Trop Med Hyg* 2010;83:1259-1261.

48 **Katusele M, Gideon G, Thomsen EK, Siba PM, Hetzel MW, Reimer LJ.** Long-lasting insecticidal nets remain efficacious after five years of use in Papua New Guinea. *PNG Med J* 2014;57:86-93.

49 **Pulford J, Oakiva T, Angwin A, Bryant M, Mueller I, Hetzel MW.** Indifferent to disease: a qualitative investigation of the reasons why some Papua New Guineans who own mosquito nets choose not to use them. *Soc Sci Med* 2012;75:2283-2290.

50 **Kurumop SF, Bullen C, Whittaker R, Betuela I, Hetzel MW, Pulford J.** Improving health worker adherence to malaria treatment guidelines in Papua New Guinea: feasibility and acceptability of a text message reminder service. *PLoS One* 2013;8:e76578.

51 **Hetzel MW, Pulford J, Tandrapah T, Jamea-Maiasa S; PNGIMR MalCon Team.** Missing in the line of duty. *PNG Med J* 2014;57:94-102.

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Evaluation of the Global Fund-supported National Malaria Control Program in Papua New Guinea, 2009-2014

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SUMMARY

The Global Fund to Fight AIDS, Tuberculosis and Malaria is the major funder of the National Malaria Control Program in Papua New Guinea (PNG). One of the requirements of a Global Fund grant is the regular and accurate reporting of program outcomes and impact. Under-performance as well as failure to report can result in reduction or discontinuation of program funding. While national information systems should be in a position to provide accurate and comprehensive information for program evaluation, systems in developing countries are often insufficient. This paper describes the five-year plan for the evaluation of the Global Fund Round 8 malaria grant to PNG (2009-2014) developed by the Papua New Guinea Institute of Medical Research (PNGIMR). It builds on a complementary set of studies including national surveys and sentinel site surveillance for the assessment of program outcomes and impact. The PNGIMR evaluation plan is an integral part of the Global Fund grant. The evaluation program assesses intervention coverage (at individual, household and health facility levels), antimalarial drug efficacy, indicators of malaria transmission and morbidity (prevalence, incidence), and all-cause mortality. Operational research studies generate complementary information for improving the control program. Through the evaluation, PNGIMR provides scientific expertise to the PNG National Malaria Control Program and contributes to building local capacity in monitoring and evaluation. While a better integration of evaluation activities into routine systems would be desirable, it is unlikely that sufficient capacity for data analysis and reporting could be established at the National Department of Health (NDoH) within a short period of time. Long-term approaches should aim at strengthening the national health information system and building sufficient capacity at NDoH for routine analysis and reporting, while more complex scientific tasks can be supported by the PNGIMR as the de facto research arm of NDoH.

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Introduction

The Global Fund to Fight AIDS, Tuberculosis and Malaria (the Global Fund) invested US$4.4 billion in malaria control programs between 2002 and 2011, covering 97 countries across the world. In 2011, the Global Fund provided about half of all international malaria funding (1). While the Global Fund contributes significantly to the resources available for malaria control, its funding is strictly performance based (2). Grant agreements with recipients of funds define process, outcome and impact indicators and respective time-bound targets. As a result, grant recipients are bound not only to demonstrate their ability to implement an intervention but also to provide evidence of health impact, the latter of which is particularly important for decisions on continuation of funding after years 2 and 5 of a grant (2).

Adequate and accurate data are essential for providing evidence of performance. The Global Fund therefore recommends 5 to 10 percent of a grant's budget to be allocated to monitoring and evaluation (M&E) (2) in order to establish and maintain a functional M&E system. The Global Fund provides guidance and a toolkit to program implementers in order to ensure that appropriate mechanisms are in place that allow key indicators to be accurately and objectively assessed (3). Many of the proposed indicators depend on components of a national health information system, such as routine health facility reports and repeated national household surveys, including Demographic and Health Surveys (DHSs) or Multiple Indicator Cluster Surveys (MICSs). However, the functionality of health information systems is compromised in many resource-limited countries as a consequence of general health systems constraints. Long-term system-wide approaches would be required to fix health information systems in a sustainable manner. Yet, while there has been increasing appreciation of the importance of health systems, long-term investments in broader initiatives are still less readily available than disease-targeted funding support (4-6).

In Papua New Guinea (PNG), the Global Fund has been the main donor of the National Malaria Control Program (NMCP) since 2004, when the country secured a Round 3 Global Fund grant. Since 2009, the PNG NMCP has been funded largely by a subsequently acquired Round 8 grant that extends until 2014. The key components of the Round 8 program are the country-wide free distribution of long-lasting insecticidal nets (LLINs), the scaling-up of parasitological diagnosis by rapid diagnostic test (RDT) or microscopy and the introduction of artemisinin-based combination therapy (ACT) complemented by advocacy and behaviour change campaigns. The activities supported by the Round 8 grant are key components of the NMCP Strategic Plan 2009-2013, which was developed based on a comprehensive review of the NMCP and wide consultation of stakeholders (7). The main roles and responsibilities of stakeholders are listed in Table 1.

The Global Fund Round 3 malaria grant to PNG relied chiefly on routine data collections for M&E (8). This strategy was undermined by the weaknesses of the system resulting in often unsatisfactory quality of M&E data (9). In PNG, routine statistics from health facilities (hospitals and health centres, but not aid posts, the lowest level of health care provision), broken down by specific disease categories, are reported through the National Health Information System (NHIS), which was implemented as a harmonized system across all provinces after 1994 (10). In parallel, the Discharge Health Information System provides information specifically about diagnoses and outcomes of admissions, including International Classification of Diseases (ICD) codes. Yet in the case of malaria the current lack of parasitological diagnosis in many health facilities (11) compromises the accuracy of NHIS-derived estimates of the number of cases. At the same time, the scaling-up of diagnostics as part of the Global Fund support is expected to result in a decrease in reported malaria cases as a result of reclassifying non-malarial fevers, which often contribute significantly to the incidence of febrile illnesses (12). Adding to the complexity in interpreting NHIS malaria trends are changes that were made to the record forms before the start of the Round 8 grant in 2008. The last DHS that could provide complementary data was conducted in 2006; however, the report of this survey has not been made publicly available and may not be sufficiently accurate (13). Towards the end of the Round 3 grant in 2008, the Papua New Guinea Institute of Medical Research (PNGIMR) was therefore contracted to carry out an end-evaluation of the program (14). It concluded that the grant had contributed to a

significant increase in mosquito net coverage despite missing the program targets on a national scale (14).

Considering the shortcomings in existing systems, the Round 8 grant was designed differently and the evaluation of the grant was delegated to PNGIMR (15). This paper describes the institutional arrangements, organization and methodological approaches developed by the PNGIMR for the independent evaluation of the Global Fund Round 8 malaria grant between 2009 and 2014. At the time of writing, the program had been running for over two years.

Evaluation of the Global Fund Round 8 malaria grant

Institutional arrangements

The PNGIMR, founded in 1968 as the Institute of Human Biology, is a statutory body of the Government of PNG (16). Over the years, the Institute has built up an international reputation of excellence in biomedical research with significant involvement of local communities in its field and laboratory studies (17,18). PNGIMR's mission is to conduct research on the major health problems affecting Papua New Guinea and provide evidence to support health policy-making and the implementation of national health strategies. Growing institutional expertise in evaluating large-scale health interventions (14,19) enabled the Institute to become a Sub-Recipient of the Round 8 grant. One of the prerequisites was the Institute's capacity, which was assessed by the National Department of Health (NDoH) and approved by the Global Fund.

As Sub-Recipient, the Institute receives funds from a Principal Recipient (PR) of the grant (initially, NDoH, followed by Oil Search Health Foundation in 2012), to which it is directly accountable. The Principal Recipient keeps track of PNGIMR's performance of fulfilling its contractual obligations, such as conducting and reporting M&E activities, and controls its expenditures. M&E data generated by PNGIMR are reported to the Principal Recipient and to the Malaria Control Program Technical Working Group, a committee comprising representatives from all PNG-based stakeholders involved in the implementation and evaluation of the NMCP and chaired by the NDoH. A project management team consisting of project managers, accountants and a logistics officer was established at PNGIMR at the start of the project. The team is responsible for regular financial and progress reporting to the Principal Recipient following the strict requirements of the Global Fund. A team of approximately 40 scientific officers, research assistants, nurses, microscopists and auxiliary staff was recruited locally in PNG. Continuous training at all staff levels following the framework of university curricula or internal and on-the-job training contributes significantly to capacity building of the PNG workforce. Scientific and management leadership for the PNGIMR activities is provided by two senior research fellows recruited overseas due to a lack of local candidates.

Monitoring and evaluation framework

Progress towards the grant's goal of "reducing morbidity and mortality due to malaria in Papua New Guinea" is assessed against outcome and impact indicators while the progress in implementing the actual program is monitored against process indicators linked to each specific Service Delivery Area. While each PR is responsible for monitoring and reporting on the process indicators related to their individual Service Delivery Areas, the evaluation of the grant's outcomes and impact indicators was delegated to PNGIMR (Figure 1). Since the Institute is not involved in the implementation of the interventions the evaluation can be considered more independent than if it was performed by one of the implementing agencies.

Indicators and time-bound targets are defined in the Performance Framework, which is part of each Global Fund grant agreement (3). The Performance Framework formed the basis for the comprehensive five-year evaluation plan developed by PNGIMR. The outcome and impact indicators with their respective targets are presented in Table 2. The three impact indicators relate to child mortality (all-cause, due to the difficulty of performing verbal autopsies) and morbidity expressed as incidence of test-confirmed clinical cases and population prevalence of parasitaemia. Of the four outcome indicators, two relate to the rollout of LLINs and one to the introduction of ACT. No results-level indicator directly captures the introduction of diagnostic tests or outcomes of behaviour change campaigns yet, arguably, the latter activity

TABLE 1

Roles and responsibilities of Round 8 program partners

	Roles and responsibilities	
	Phase 1	**Phase 2 (Years 3-5)**
Principal Recipient (PR)		
National Department of Health (NDoH)	Delivery of first-line treatment (ACT) and improvement of diagnostics (RDT and microscopy). Strengthen M&E capacity.	(Resigned as PR.) Technical oversight, endorsement, coordination of program. Quantification of ACT/RDT requirements and monitoring of compliance with treatment guidelines.
Oil Search Health Foundation (OSHF)		(PR substituting NDoH.) Administration of grant finances. Monitor activities and implementation targets defined in Performance Framework, in partnership with NDoH. Ensure that documentation and deliverables meet Global Fund standards.
Rotarians Against Malaria (RAM)	Delivery of LLINs to all households, covering every province every three years. Delivery of LLINs to provinces for antenatal clinics, boarding schools, correctional centres, people living with HIV and for emergencies.	
Population Services International (PSI)	Behaviour change communications, advocacy, mass media campaigns.	Behaviour change communications (country-wide) and piloting of home-based management of malaria (HMM) in East New Britain, East Sepik and West Sepik Provinces.
Sub-Recipient (SR)		
Divine Word University/Diwai Pacific Ltd	Training health workers in malaria case management (particularly use of RDTs and ACT administration) and training of laboratory assistants. Reporting to RAM.	

PNG Institute of Medical Research	Evaluation of the NMCP and management of operational research program as described in this publication. Reporting to NDoH (phase 1) and OSHF (phase 2).
Save the Children	Pilot implementation of HMM program in East Sepik Province. Reporting to PSI.
Burnet Institute	Pilot implementation of HMM program in East New Britain Province. Reporting to PSI.
Other partners	
World Health Organization	Long-term technical assistance for planning, implementation, monitoring and evaluation of malaria control interventions, including guidelines revision, ACT/RDT rollout, quality assurance for diagnostics, strengthening surveillance and reporting, and coordination of the malaria program review.

ACT = artemisinin-based combination therapy
RDT = rapid diagnostic test
M&E = monitoring and evaluation
LLIN = long-lasting insecticidal net
HIV = human immunodeficiency virus
PNG = Papua New Guinea
NMCP = National Malaria Control Program

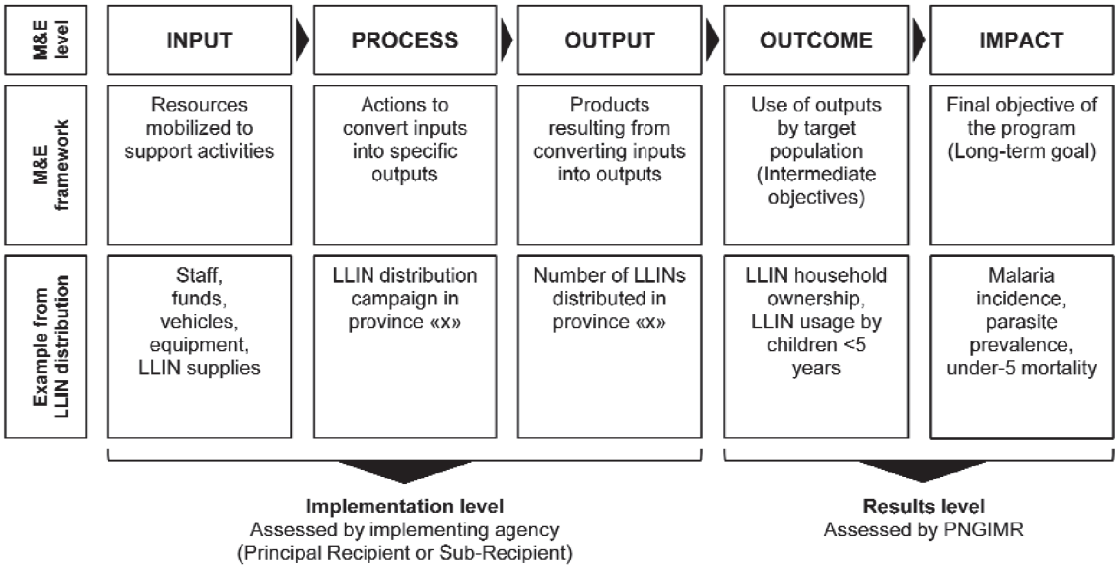

Figure 1. Overall monitoring and evaluation (M&E) framework and the Papua New Guinea Institute of Medical Research (PNGIMR) evaluation plan.
LLIN = long-lasting insecticidal net

should contribute to the level of mosquito net usage and treatment compliance. While each of the three Principal Recipients is using a different Performance Framework with process indicators pertaining to their specific Service Delivery Areas, the results-level indicators are universal across the grant. In the context of the phase 2 grant renewal process, some of the targets were reviewed and adjusted to account for updated baseline values.

The PNGIMR evaluation was designed primarily to assess outcome and impact indicators defined in the Performance Framework. At the same time, all data collection components also gather complementary information that can be used to triangulate monitoring data collected by program implementers and provide additional evidence of program outcomes and impact. Examples include data about uptake of RDTs and ACTs in health facilities, coverage with behaviour change campaigns, changes in mosquito-biting behaviour and malaria transmission (entomological inoculation rate), and trends in antimalarial drug resistance. All information feeds into key indicators defined in the NMCP Strategic Plan 2009-2013 (7).

The evaluation plan is based on a plausibility design with a before-after assessment (20). It combines several complementary data collection mechanisms aiming to assess changes in intervention coverage over time alongside trends in malaria morbidity, mortality and transmission. The evaluation activities address and combine interlinked aspects of malaria control and research at the levels of health care providers, the community, the malaria patients, the mosquito vector and the parasite. The comprehensiveness and scale of the evaluation plan is probably unique in the context of large donor-funded programs and will contribute to a better understanding of the contribution of the funded interventions to changes in malaria patterns. The evaluation plan, as well as the actual implementation of interventions, was confronted with operational realities that could not all be anticipated during the design phase. These include, for example, law and order problems, deterioration of transport infrastructure in certain locations, and difficulty in local recruitments in provinces with either low availability of the required human resources or other highly competitive

employers. The evaluation plan presented in detail below is a description of the evaluation activities as they were being implemented at the end of phase 1 rather than an exact reflection of the original plan. The key activities are based on two complementary approaches: repeated country-wide cross-sectional surveys in randomly sampled locations and longitudinal surveillance in selected sites (Figure 2). In the following five sections we outline the rationales and methodologies behind individual components of the Round 8 evaluation plan.

1. Country-wide household surveys

Study sites

Country-wide household surveys were scheduled for 2010-2011 and 2013-2014 with the baseline values derived from a comparable survey conducted during the end-evaluation of the Round 3 malaria grant in 2008-2009 (14). Each survey sample consists of five villages selected from each of 20 provinces using a simple random sampling approach. Villages that are inaccessible at the time of survey due to logistical or security reasons are replaced by another randomly sampled village from the same province. The sampling frame includes all geo-referenced villages ('census units') identified in the 2000 national population census (21). Census units include rural villages as well as suburbs, settlements or otherwise identifiable areas of urban centres. Within each selected village, up to 30 households are then randomly sampled from a list of households established ad hoc by the survey team leader and local village representatives.

Procedures and instruments

Surveys are carried out during the main transmission season roughly between November and June. Three field teams consisting of three to four PNGIMR staff including at least one nurse/community health worker simultaneously work at different sites. All field team members were trained extensively in the use of survey instruments and (in the case of clinical staff) blood collection and sample preparation techniques. Before a survey, the respective provincial and district health authorities are informed of the study objectives, sites and timetable. Each team is then accompanied by a representative of the respective health authority who assists in liaising with and informing the communities.

A structured questionnaire following the design of the Malaria Indicator Survey household questionnaire (22) is completed with adult heads of selected households. This form collects information about LLIN ownership and use, behaviour change campaigns and other interventions alongside background demographic information on each household member as well as indicators of the household's socioeconomic status. A structured treatment-seeking questionnaire is used to elicit information on treatment choices from household members who reported a fever in the two weeks before the survey. Information about village accessibility, nearest health services, village-level health initiatives (such as the healthy island/healthy village concept (23)) and the village leader's perception about local health problems and potential remedies is recorded in a village leader questionnaire. The questionnaires are administered by the PNGIMR field staff. All members of selected households aged six months and above are eligible for providing a finger-prick sample of capillary blood. One thick and one thin smear are prepared on the same glass slide for each participant. Microscopic diagnosis of malaria is performed at PNGIMR and each slide is read independently by at least two microscopists following standard World Health Organization (WHO) protocols. In 2013-2014, an additional sample of blood will be collected on filter paper for molecular detection of parasitaemia and markers of drug resistance (24-26). Symptomatic participants are diagnosed on the spot using a malaria RDT and positive cases are treated according to standard treatment guidelines. Haemoglobin (Hb) level is measured with a portable HemoCueHb 201+ analyser (HemoCue AB, Ängelholm, Sweden) and axillary temperature with an electronic thermometer. Information on previous malaria treatment and recent travel is recorded for each participant who provides a blood sample. Village locations and elevation above sea level are recorded with hand-held Global Positioning System (GPS) devices (Garmin etrex, Garmin Ltd, Olathe, Kansas, USA). Oral informed consent is sought from village leaders and all study participants.

Data analysis

Country and regional level proportions are calculated for indicators of intervention coverage, most importantly the evaluation

TABLE 2

Outcome and impact indicators of the Round 8 Performance Framework

Impact/outcome indicator		Indicator	Baseline Value	Baseline Source	Baseline (Year)	Target Year 1	Target Year 2	Target Year 3	Target Year 4	Target Year 5
Impact	1.	Parasite prevalence: percentage of children aged 6-59 months with malaria infection (detection of parasitaemia by microscopy)	24	Household survey	(2009)	24%	20%	n/a	n/a	17%
Impact	2.	Annual parasite incidence: number of malaria cases confirmed by microscopy detected per 1000 population during one year	205	Sentinel site surveillance	(2009)	158	205	185	17	150
Impact	3.	All-cause mortality rate among children younger than 5 years of age	n/a	Sentinel site surveillance	(2011)	n/a	n/a	Baseline to be set	n/a	To be determined
Outcome	1.	Proportion of households with at least 2 LLINs	38%	Household survey	(2009)	41%	61%	n/a	n/a	90%

Outcome	2.	Proportion of pregnant women who slept under LLIN the previous night	40%	Household survey	(2009)	40%	60%	n/a	n/a	70%
Outcome	3.	Proportion of children under 5 years old who slept under LLIN the previous night	40%	Household survey	(2009)	40%	60%	n/a	n/a	80%
Outcome	4.	Percentage of children younger than 5 years of age with fever in the last 2 weeks who received antimalarial treatment according to national policy	10%	Household survey	(2009)	10%	50%	n/a	n/a	60%

LLIN = long-lasting insecticidal net

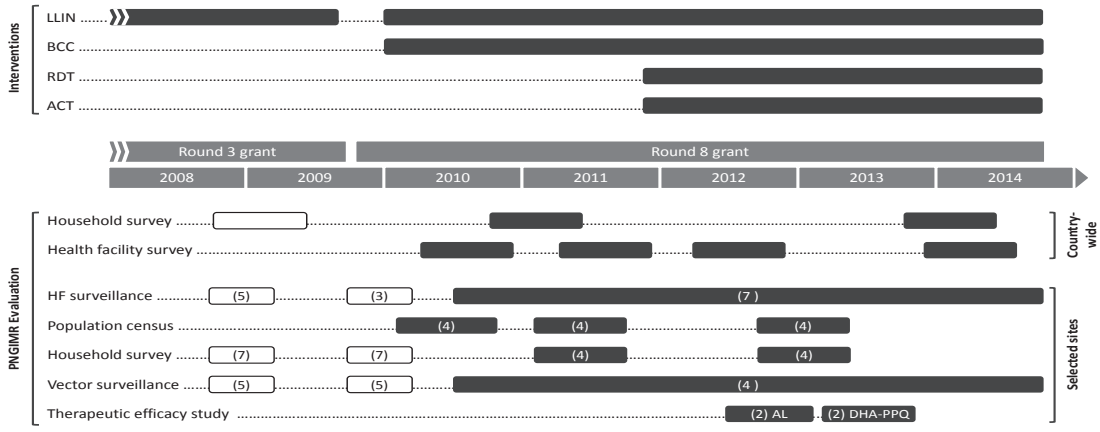

Figure 2. Interventions and evaluation activities by the Papua New Guinea Institute of Medical Research (PNGIMR) over the two Global Fund grants.
LLIN = long-lasting insecticidal net, BCC = behaviour change campaign, RDT = rapid diagnostic test, ACT = artemisinin-based combination therapy, HF = health facility, AL = artemether-lumefantrine, DHA-PPQ = dihydroartemisinin-piperaquine. PNGIMR evaluation: numbers in parentheses indicate number of sites.

indicators: proportion of households with at least two LLINs, proportion of pregnant women who slept under an LLIN the previous night, proportion of children under 5 years old who slept under an LLIN the previous night, and percentage of children younger than 5 years of age with fever in the last two weeks who received antimalarial treatment according to national policy. Analysis weights are used to adjust proportions and confidence intervals to the province-stratified sampling. Overall weights are calculated as the inverse of an observation's probability of selection taking into account the sampling stages. Prevalence data are presented for national and regional levels as weighted proportions of people infected with *Plasmodium* spp., where a positive result is based on at least two independent concordant light microscopy reads.

2. Country-wide health facility surveys

Study sites

The national health facility surveys were conducted in 2010, 2011 and 2012 and are planned for 2014 in areas with endemic or potentially epidemic malaria. Each survey sample consisted of two health centres or health subcentres and up to four aid posts selected from each province using a simple random sampling procedure. The sampling frame for each survey includes all health centres operational in March 2010 inclusive of government and mission-administered health facilities (N = 689) based on a list provided by NDoH. Aid posts are randomly selected on site at participating (ie, randomly selected and consenting) health centres. The sampling frame for aid posts includes all operational aid posts under the supervision of the health centre at the time of survey. Hospitals are excluded from the sampling frame as they are few in number and provide primary health care services only to a minority of the PNG population (27). Conversely, health centres and aid posts are widely available across the country as they are the main providers of primary care.

Procedures and instruments

Each survey is carried out from June to November by three trained field teams working simultaneously at different sites. The training program for field staff included intensive instruction and practice on the survey instruments. Members of each survey team spend between three to five days at

each participating health centre and up to one day at each participating aid post. Before any health facility visit, the respective health authorities are informed and requested to commission a health officer to accompany the field team.

Four survey instruments are completed at each health facility. A structured health facility checklist is completed with the officer in charge and designed to assess the availability of supplies relevant to malaria case management (quantity of RDT in stock, quantity of microscopes and availability of essential microscopy supplies, availability of antimalarials). All reported stock is observed by the respective field team leaders. An interviewer-administered questionnaire is completed with all available and consenting health workers employed at participating health facilities. The questionnaire contains open and closed questions pertaining to education, work experience and supervision, type and utility of any malaria-related training received (inclusive of training on new treatment policy), knowledge, attitudes and practices relevant to malaria case management and, if applicable, experiences implementing the new treatment policy. As a further component, features of the clinical case management of patients presenting with fever or a recent history of fever are recorded in a structured checklist. The instrument is divided into discrete sections including consultation and diagnosis, prescription and treatment counselling. The content of each section was informed by input from experienced medical and medical research professionals. The instrument is completed by a trained field team member who silently observes the management of fever patients from the point of initial contact with a health professional until service exit or admission on to a treatment ward. During the course of this observation, the field team member records whether specified actions do or do not occur and records the content of specific actions (eg, whether an RDT is conducted and, if yes, what is the outcome). Eligible patients are identified upon first contact with a health worker or, if circumstances allow, by screening in the waiting area before first contact with a health worker. Lastly, a questionnaire is administered to fever patients at the time of service discharge. It contains a range of open and closed questions pertaining to the patient's treatment experience, his or her retention of clinical information, treatment accessibility and cost, and pre-treatment behaviour. Oral informed consent is sought from the officer in charge at all participating health facilities and from all participating clinicians and patients before clinical observation.

Data analysis

Country-level proportions are calculated for indicators of intervention coverage, most importantly for: proportion of health facilities with working microscopy or with malaria RDT in stock; proportion of health facilities with the first-line drug artemether-lumefantrine (AL) in stock (for all four weight groups); proportion of health care providers trained in malaria case management (new treatment guidelines and use of RDTs); and proportion of fever cases presenting to health facilities diagnosed and treated according to national guidelines. The calculation of confidence intervals will adjust for possible clustering at the health facility level where required.

3. Sentinel surveillance sites

Sentinel surveillance sites have been established to follow morbidity and mortality trends alongside intervention coverage indicators in the same known population over the entire period of the Global Fund grant. The sentinel site activities include (i) morbidity surveillance in health facilities, (ii) demographic surveillance and repeated household surveys in the catchment area, and (iii) mosquito vector surveillance.

Study sites

Sentinel site locations were purposely selected considering accessibility of the site, local malaria epidemiology, presence of a functioning health centre and its case load and estimated catchment population. Two surveillance sites established in the frame of the end-evaluation of the Global Fund Round 3 malaria grant (2008-2009) were retained (12). A total of 7 sites were selected, 2 each located in Southern, Momase and Islands Regions and 1 located in the highlands (Figure 3). Basic characteristics of the sites are listed in Table 3. In each region, 1 site is dedicated to comprehensive surveillance including health facility- and community-based morbidity and mortality surveillance; the remaining 3 sites are considered complementary with only health facility-based activities.

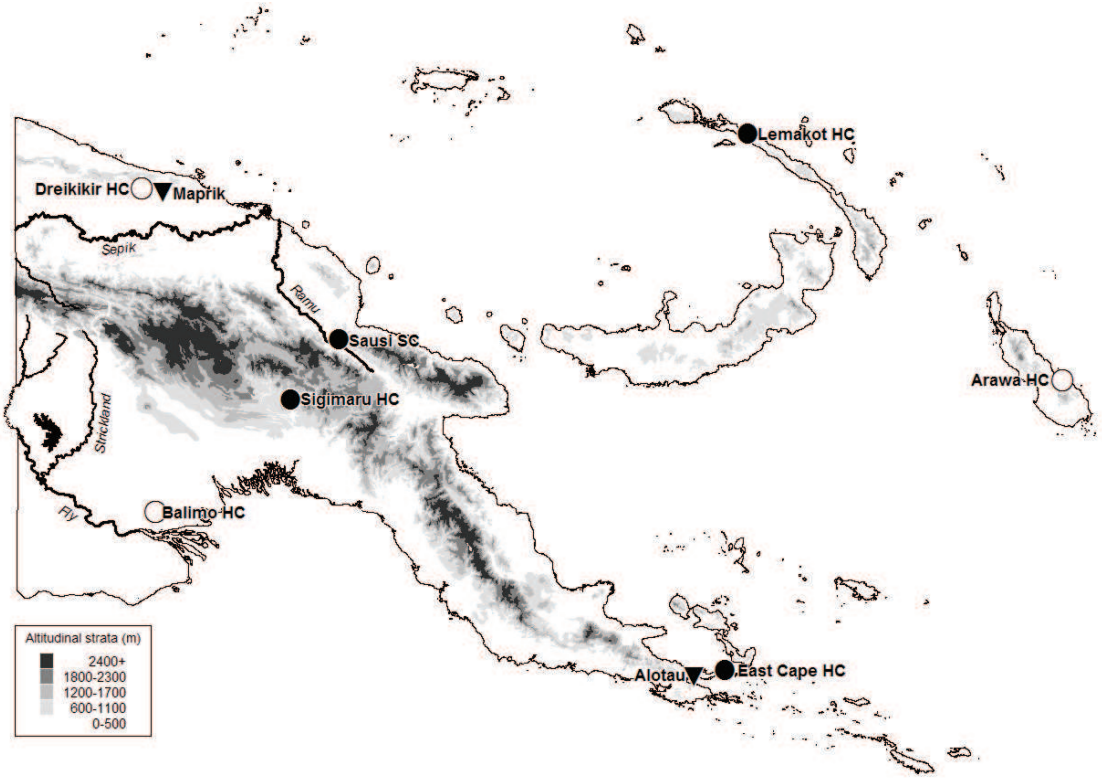

Figure 3. Map of sentinel surveillance and therapeutic efficacy study (TES) sites.
● indicates sentinel sites with demographic surveillance
○ sites without demographic surveillance
▼ therapeutic efficacy study sites
HC = health centre, SC = health subcentre

Establishment of sentinel surveillance sites

Health facility-based morbidity surveillance was established as the first activity in each site. In close dialogue with provincial health authorities the most suitable facility was selected and formal written approval obtained from the relevant authorities and organizations. During reconnaissance visits, the officer in charge of the facility was briefed, availability of working space, staff accommodation and basic infrastructure (eg, water) verified and information on catchment villages (eg, names of villages, population, resource persons) obtained. Health facilities lacking proper maintenance, staffing or leadership were not considered, whereas facilities with inpatients were preferred. In one case, a facility was excluded because of traditional beliefs of bad spirits inhabiting the facility, which was considered to negatively influence patient attendance. The establishment of the health facility surveillance was formalized for each site in a Memorandum of Understanding. A nurse or community health worker was then recruited (whenever possible locally) and based in the health facility. Training on institutional policies, rationale and details of the study and data collection procedures was provided on site.

Following the successful start of the health facility-based surveillance, the demographic component was established in the four 'fully fledged' sites. This successive implementation allowed the communities to get used to the presence of PNGIMR researchers. Meetings with village councillors of the proximate

catchment area were required to inform about the planned surveillance activities, assess accessibility and approximate population size of the villages and obtain the community leaders' approval. In subsequent village meetings ('tok save'), community members were provided with the same information in order to obtain collective community approval. Demographic surveillance was conducted in villages surrounding the sentinel site health facility with the aim to cover as much of the 'real' catchment area of the facility as operationally feasible. In certain locations, this does not exactly match the 'official' catchment area, which is often based on administrative considerations rather than accessibility.

Health facility surveillance procedures and instruments

Over a period of 6 to 12 months, including the period of the main transmission season, all outpatient cases and admissions to the sentinel site health facility are screened for fever in the past three days. The PNGIMR nursing officer then collects a capillary blood sample by finger-prick for on-the-spot diagnosis of malaria by RDT, preparation of thick and thin blood film for microscopic diagnosis and determining Hb level using a HemoCueHb 201+ Analyser. Demographic details of the patient are recorded in a one-page form alongside clinical signs and symptoms, previous health facility attendance and drug intake, axillary temperature (measured with an electronic thermometer), body weight and outcomes of the RDT and Hb measurement. Results of the clinical assessments are also recorded in the patient's clinic book. Following this procedure, the patient is transferred to a health facility staff member for further examination and treatment following routine procedures established by the facility. The final diagnosis and any treatment by the health facility clinician are recorded. Patients who are admitted follow the same procedure, but are later followed up until the time of discharge or referral.

Demographic surveillance procedures and instruments

An initial census was undertaken following the delineation of the demographic surveillance area. All households were registered, assigned to a specific village and numbered, with the assigned numbers painted on house walls for easier identification in subsequent rounds. Household locations were recorded by a hand-held Garmin GPS device. All household members were identified, demographics and relationships to other household members were recorded on census forms colour-coded by site and a personal identifier unique within each of the sites was assigned. No permanent village reporters could be engaged within the available funding frame, but village volunteers assisted in the identification and enumeration of households. Census update rounds are scheduled for every year. The frequency of updates was dictated by the financial and human resources available. During update rounds, the status of residence for each previously registered household member is updated in a household register book and all births, deaths and in- and out-migrations that occurred since the last census are recorded on separate dedicated forms. Particular attention is paid to the outcomes of pregnancies recorded during the previous round.

Household survey procedures and instruments

A household survey is conducted in a sub-sample of all households on a bi-annual basis in sites with demographic surveillance. The same methodology is applied as in the country-wide household survey described above. A simple random sample of households is drawn for each site from the demographic database. The aim of this component is to correlate incidence and mortality trends with changes in parasite prevalence and changes in intervention coverage observed in the same known catchment population.

Data analysis

Annual species-specific malaria incidence rates are calculated from light microscopy-confirmed cases presenting to four sentinel site health facilities and the respective catchment population. Supporting evidence on trends in malaria cases are calculated from health facility data derived from those sites without demographic surveillance and hence lacking a population denominator.

Mortality trends are followed in four sites by calculating all-cause under-five mortality as deaths per 1000 live births based on annual retrospective records of these events. Due to the difficulties inherent in establishing a cause of death, particularly in a community setting,

TABLE 3

Details of sentinel surveillance sites

Province	Sentinel site health facility	Altitude (metres)	Monthly OPD fever cases screened* (start of screening)	Proportion of fever cases with positive RDT*	Population under surveillance (2010)	Household surveys	Entomology surveys
Southern Region							
Milne Bay	East Cape HC	15	184 (07/2010)	52%	5684	Yes	Yes
Western	Balimo HC	15	66 (07/2011)	2%	-	-	-
Highlands Region							
Simbu	Sigimaru HC, Karimui	1140	71 (02/2011)	28%	8506	Yes	-
Momase Region							
Madang	Sausi SC	170	141 (09/2010)	26%	5158	Yes	Yes
East Sepik	Dreikikir HC	330	137 (07/2011)	21%	-	-	Yes

Islands Region

New Ireland	Lemakot HC	15	278 (01/2011)	32%	10384	Yes	Yes
Bougainville	Arawa HC	10	79 (05/2011)	6%	-	-	-

OPD = outpatient department
RDT = rapid diagnostic test
HC = health centre
SC = health subcentre
*Average of first 12 months under surveillance

all-cause mortality will be used as a proxy for changes in malaria-related deaths. No verbal autopsies as previously conducted in other surveillance sites in PNG (28) are conducted in the framework of this project.

4. Vector monitoring

Study sites

Entomological surveys are conducted at four-monthly intervals in two villages in each of the following four sentinel sites: Kokofine and Mauno from Sausi, Bou and Topa from East Cape, Lamusmus and Lemakot from Lemakot, and Nanaha and Yauatong from Dreikikir. Each village was divided into 6 geographical hamlets and 1 household was chosen per hamlet. Mosquitoes are collected by outdoor human landing catch for two consecutive nights per house (18:00-06:00), for a total collection effort of 12 person-nights per village and 24 person-nights per sentinel site. Eight mosquito collectors and two supervisors were employed from the village, with each collector working a six-hour shift. Collectors are systematically rotated between shifts and households to avoid individual differences in host attractiveness. Two PNGIMR technicians supervise the nightly collections and identify and process mosquitoes.

Data collection procedures and instruments

Mosquitoes are stored according to the location and collection hour. They are morphologically identified (29,30) the following morning and anophelines are stored dry on silica gel. Mosquitoes that are morphologically identified as members of the *Anopheles punctulatus* group (the major malaria vectors in PNG) are confirmed to species by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) of the ITS2 region (31) using either an individual leg or extracted DNA (deoxyribonucleic acid) from the DNeasy blood and tissue kit (QIAGEN, Maryland, USA). Lysate from whole mosquitoes is screened for *Plasmodium falciparum*, *P. vivax* 210 and *P. vivax* 247 circumsporozoite protein by enzyme-linked immunosorbent assay (32).

Data analysis

The geometric mean (33) man-biting rates are calculated based on nightly total catch per team for each village and month. Entomological inoculation rates for *P. falciparum* and *P. vivax* are calculated based on the estimated total number of bites per person per year and the sporozoite prevalence in mosquitoes.

5. Drug resistance monitoring

Study sites

Monitoring resistance of malaria parasites to the first-line (artemether-lumefantrine, AL) and second-line (dihydroartemisinin-piperaquine, DHA-PPQ) antimalarial drugs is an integral part of the evaluation plan. The Maprik area in East Sepik Province and Alotau in Milne Bay Province were selected as sites for therapeutic efficacy studies (TESs) (Figure 3) due to the high endemicity of both *P. falciparum* and *P. vivax.* The sites are located in the vicinity of a sentinel surveillance site (Dreikikir near Maprik, East Cape near Alotau), which can provide contextual data from a comparable environment. Study site selection was based chiefly on logistical considerations as a basic research infrastructure is required for conducting a therapeutic efficacy study. PNGIMR's Maprik branch provides basic laboratory, administrative, transport and staff housing infrastructure. The same sites had previously been used for clinical and epidemiological studies on malaria, including malaria drug efficacy studies (34-36). Alotau was established as a TES site before the start of the Round 8 evaluation and the Provincial Health Authority provides access to laboratory infrastructure within the provincial hospital.

Data collection procedures and instruments

The study protocol follows the WHO guidelines "Methods for surveillance of antimalarial drug efficacy" (37). Due to a marked decrease in malaria cases before the study onset, inclusion criteria for low-to-moderate transmission settings are being applied. Patients older than six months with mono-infection of *P. falciparum* (>1000 asexual parasites/µl of blood) or *P. vivax* (>250 asexual parasites/µl), acute fever or history of fever within the previous 48 hours, not presenting with any signs of severe illness (as per WHO guidelines) and resident in the study area are eligible for enrolment. Patients below 5 kg body weight, those with a clinically relevant concomitant illness, those having concurrent or recent (last two weeks) intake of an antimalarial drug, pregnant women and breastfeeding mothers are excluded.

Following written informed consent, patients are followed up for 42 days at the patient's home. Each morning dose of AL (Coartem®, Novartis) and every DHA-PQP (Eurartesim®, Sigma-Tau) dose is given under direct supervision. Blood samples are collected on each follow-up day on microscopy slides and filter paper for DNA extraction and PCR. Details on clinical signs and symptoms are recorded in standard case report forms. Microscopy slides will be read independently by two different microscopists.

Data analysis

Per-protocol and intention-to-treat analyses will be performed on adequate clinical and parasitological response (ACPR) on days 28 and 42. The primary endpoints will be PCR-corrected ACPR for *P. falciparum* and non-PCR-corrected ACPR for *P. vivax*. PCR-corrected *P. vivax* treatment outcomes following previously established and tested methodology (38) will be used as a secondary endpoint. Day 3 parasitaemia will be reported as an early indicator for therapeutic failure of artemisinin (39).

Operational research studies

To further support the successful implementation of the National Malaria Control Program, an operational research (OR) component was included in the monitoring and evaluation plan. The operational research program aims to close gaps in the knowledge about malaria control interventions in PNG, and provide program managers and key decision-makers with the evidence base for operational and policy decisions. The operational research program is determined on an annual basis by the Malaria Control Program Technical Working Group and is managed by the PNGIMR. Approved activities may be carried out by PNGIMR or contracted to other institutions depending on the research question and the available research capacities and expertise. Examples of operational research studies conducted to this date include a qualitative investigation into the reasons why some people who own mosquito nets do not use them (40), a study on the quality of antimalarial medicines available from public providers (41) and an ongoing study about the prevalence of glucose-6-phosphate dehydrogenase (G6PD) deficiency phenotypes and genotypic markers.

Discussion

Programmatic aspects

An accurate and timely evaluation of outcomes and impacts of the Global Fund-supported National Malaria Control Program is the aim of the described program. Ideally, data on program impact should originate from the existing health information system requiring minimal additional input (3). However, especially household-level indicators relating to program coverage cannot be captured easily with a facility-based data collection mechanism, such as the one established in PNG. Even though population-based surveys are considered an essential part of a country health information system (42), national household surveys are not regularly conducted as part of the PNG NHIS. The last DHS conducted in PNG dates back to 2006 and there are concerns about the reliability of its findings (13). Interestingly, even for malaria, which is recognized as one of the main health problems of PNG, no country-wide survey had been conducted before the evaluation of the Global Fund Round 3 grant. Lack of dedicated funding and operational capacity to conduct such surveys may be some of the reasons behind this shortfall. The country-wide malaria survey established in the frame of this evaluation therefore provides an opportunity to gain a more in-depth understanding of the malaria situation in PNG alongside generating data on program outcomes and impact. Nevertheless, this should not divert attention from the need to strengthen the health system and with it the NHIS so that data collected routinely is reliable enough to be used for program evaluation and planning (43). While disease-specific surveys can address pertinent program-specific questions, conducting regular national surveys with a broader focus (eg, MICS or DHS) in the frame of an NHIS could provide complementary data relevant to the PNG health system as a whole. Such surveys require a solid methodology and should involve a wide range of in-country stakeholders to be successful in a country that is as diverse and operationally difficult as PNG. Global Fund programs have previously been shown to be only partly integrated into the PNG health system, reinforcing the verticality of disease control programs by establishing parallel systems, particularly in the areas of M&E and supply chain management (44). It is hence unlikely that Global Fund programs contribute significantly to systems

strengthening and government-led initiatives in that area would be of paramount importance. In the case of the Round 8 grant, activities and targets are aligned with an NMCP Strategic Plan which was developed in the preparation phase of the grant application (7). It is likely that in the development of this plan a certain alignment to future donor requirements was sought. Since 2011, efforts are being made to improve the PNG NHIS in the areas of data transmission and management, reporting and analysis (43). Previous changes introduced in the monthly NHIS reporting form in 2008 eliminated some redundant malaria indicators (eg, 'Total nets in use', 'Number of nets retreated') and simultaneously introduced a new age and sex breakdown in the total number of outpatient and inpatient cases and a separate reporting of microscopy- and RDT-confirmed cases. Supposedly, the aim of this change was a more detailed reporting of malaria indicators (and possibly other disease-specific data) more closely aligned with donor reporting requirements. The direct consequence was an increase from 17 to 104 in the number of malaria fields in the monthly reporting form to be completed by each health centre and hospital (NHIS Monthly Report, Government Printer -266/80.-6.2006 vs NHIS Monthly Report 2009). It is very likely that this increase in reporting burden has some effect on data quality, particularly in light of anecdotal evidence that the species-specific results from RDTs are often misreported. It has been suggested, therefore, that reducing to a minimum the set of key indicators might be more valuable than collecting a maximum amount of data (45). This example also highlights that health systems interventions need to be designed and planned carefully, taking into consideration any unintended effects that the interventions may have on the system.

Currently, the insufficient data management and analysis capacity at the NDoH is unlikely to meet the additional reporting demands from the Global Fund (43,44). Outsourcing the evaluation to the PNGIMR relieves the NDoH from a burden the department was unlikely to be able to shoulder. Selecting a public institution of PNG as an evaluator agency has helped to engage and strengthen, to a maximum possible extent, in-country capacity. With a proven track record of large-scale program evaluations and in-depth clinical and epidemiological studies on malaria with a scientifically sound quality (19), the PNGIMR is well placed to support the NDoH in this area. The organizational arrangement of the current plan could therefore not only be adequate for the current grant evaluation but also serve as an example for other comparable disease control programs. Carefully considered partnerships can strengthen the system by using existing capacities in the health sector and creating synergies within the system (46). Past experience has shown that staff rotations between PNGIMR, NDoH and other partners can contribute to an exchange of expertise among institutions in the health sector. This could be strengthened with a more strategic formalized staff exchange or secondment program in the framework of strengthening the NHIS.

Methodological approach

The attribution of changes in morbidity or mortality to specific interventions is always difficult. In most cases, program evaluations are based on a before-after plausibility assessment due to the lack of an adequate control group (20). The advantage of the chosen evaluation design lies in the possibility to link impact data directly with program intervention coverage. During household surveys, mosquito net coverage is assessed alongside malaria prevalence data. In sentinel surveillance sites, coverage with interventions (such as LLINs and ACT) and community prevalence data are complemented by mortality figures, incidence data and vector-level transmission data collected by the project from the same known populations. These survey data can then again be triangulated and verified using process indicators which are primarily assessed by the implementing agencies. Ideally, all data should be linked to information collected through the NHIS, which could be used to model errors inherent in the routine system. While this latter approach was included in the original program proposal, difficulty in accessing the facility-level (raw) data from the Department of Health has so far prevented PNGIMR from engaging in such an analysis.

Evaluation data are shared regularly with the stakeholders in the NMCP and can hence provide the program with a more regular and up-to-date 'reality check' than large-scale surveys such as a DHS and, currently, more reliably than NHIS. However, financial and human resource capacities at PNGIMR limit the detail and timeliness of the generation

and dissemination of results. While this does not compromise the value of the data for the overall project evaluation it limits the degree to which the data can be used for actual program planning. In order for survey or surveillance results to be 'actionable' in this way, a higher resolution (eg, at district level or below) would be required (45). Arguably, sub-regional data are more valuable in general, considering the heterogeneities in health indicators (13,47) and the decentralized nature of the PNG health system (48). On the other hand, there is always a trade-off between optimal and essential data. This program is evaluating a country-wide program which is centrally managed, and the effects of a decentralized health system which may otherwise impact strongly on intervention rollout are unlikely to play a major role. In order to take into consideration at least larger variations in the malaria epidemiology, as well as major variations in intervention coverage uncovered in 2009 (49), geographical regions (Southern, Highlands, Momase and Islands) were considered as a secondary analysis level. The resolution of these two analysis levels was then considered sufficient for this program evaluation.

For providing country-level figures of community-level indicators, a simple random sample of villages can provide representative results at a fairly low sample size. In similar surveys, probability-proportional-to-size sampling is often applied (50,51). A prerequisite for such an approach is the availability of reliable population data from a census and the ability to clearly identify the sampled census unit demarcations in the field. With the latest available population census dating back to the year 2000 we found that both requirements were not met. A simple random sampling across PNG would be operationally challenging (and expensive) in a country in which many sites are difficult to access. A stratified multi-stage random sampling approach was therefore applied to allow for operational realities (by applying several sampling stages) and an interest in sub-national data (by stratification by province). The consequence of this approach is the need for applying probability weights to the analysis of population-level indicators. These weights are calculated as the inverse of an observation's probability of being included in the sample adjusting for differences in the number of households or individuals in the population that each observation represents (52).

The timing of household surveys roughly overlaps with the main malaria transmission season, which follows the pattern of the rainy season between January and April in large parts of the country (53). While there is marked seasonality in the incidence of clinical malaria episodes, the prevalence of malaria infections is more stable throughout the year (54). The precise timing may therefore be less relevant for the outcome of prevalence surveys. There are no data available so far about seasonality in net usage that has been found to occur elsewhere (55,56). Survey timing also has to take into consideration the availability of field staff and logistics capacity at PNGIMR by avoiding a large overlap between household survey and health facility surveys. In general, the timing of large malaria surveys is complicated as the periods with highest transmission coincide with the most adverse weather conditions for field work, poor road conditions, the regional tropical cyclone season and consequently difficult accessibility of many remote locations.

Lessons learned to date

Three years into the five-year evaluation, the plan developed by PNGIMR has proven solid and useful, yet ambitious and at times challenging to implement. Indicators on outcome and impact measures were calculated and reported to the Global Fund. First reports of program impact on malaria prevalence were included in the 2012 World Malaria Report (57). Complementary investigations provided valuable insights into barriers to mosquito net use (40), management of febrile illnesses in health facilities (58) and at household level (59), and bio-efficacy of LLINs (60). Some important gaps remain in the area of insecticide resistance monitoring, which is not funded through the Round 8 program, despite being considered crucial nowadays (61). Experience from country-wide surveys shows that the field teams' visits to health facilities and remote villages are generally highly appreciated by the communities and health care workers, particularly in the light of otherwise limited government presence and supervision. On the other hand, villagers and health workers may frequently have expectations from PNGIMR field staff which cannot be fulfilled (eg, distribution of mosquito nets, health service provision). PNGIMR nurses in sentinel surveillance sites have

managed to establish excellent relationships with the hosting health facility and the community. The presence of the PNGIMR staff is seen as a support rather than a burden and patients appreciate the few extra services provided (eg, Hb measurement, which is also provided to non-study patients). Particularly in sentinel surveillance and drug resistance monitoring sites, PNGIMR's presence is now considered normal. In the areas from which drug efficacy patients are recruited, the frequent household visits by the study nurses provide an opportunity for villagers to see a medically trained person without having to travel to a health facility. As a matter of course, patients found to urgently require medical attention are provided transport to a referral facility by PNGIMR. Whenever possible, qualified local people are employed for stationary jobs (eg, nursing staff in surveillance sites) while during surveys local villagers are engaged as porters or field assistants. The involvement of provincial or district health officers during surveys has proven invaluable for establishing initial relationships with communities. The involvement of local people in scientific research and the combination of research and implicit provision of certain services has allowed very favourable relationships with communities, reinforcing the sound experience of PNGIMR working with rural communities from earlier years (18).

On the other hand, implementation of the above program has required capacities that are not easily available in PNG. Firstly, recruitment of suitably qualified scientific staff is a challenge and the training and supervision burden on the few senior staff members is substantial. Financial resources, while considerable with approximately 8% of the overall original grant budget of USD120 million, are challenged by high travel costs which make country-wide surveys an expensive undertaking. Regular supervision of field data collectors is essential, but supervisory capacity is limited. As a consequence, the number of surveillance sites was reduced from a planned 8 sites with demography to 4, plus 3 sites without demography. Drug resistance monitoring, which requires even closer supervision due to good clinical practice (GCP) requirements, was limited to two sites with existing infrastructure and PNGIMR presence. Logistical challenges relate to the reliability of air and sea transport, the condition of roads and the security situation. Law and order problems do not only pose a challenge to research studies, as experienced directly by our team (62), but also to the implementation of health services as reflected in frequent local newspaper reports of violence against health workers.

Scientific capacity at PNGIMR may be compromised by high staff turn-over, but has also been impacted negatively by the uncertainties surrounding the transition from phase 1 to phase 2 of the Round 8 program. The change of Principal Recipient from NDoH to Oil Search Health Foundation at that point resulted in an interruption of fund disbursement to PNGIMR, forcing the Institute to lay off many of its key project staff. Consequently certain activities in sentinel surveillance sites were interrupted and had to be rescheduled once funding was again available (eg, census update rounds and household surveys). Difficulties recruiting PhD-level senior project staff locally or internationally, despite competitive salaries, have resulted in some delays in report writing and adversely impacted on the availability of certain study results for program implementers. On the other hand, the availability of data does not always guarantee that programs are adjusted or adjustments are approved by the donor agency. A prerequisite for this is a regular interaction between PNGIMR and the implementing partners. While a platform for this exchange exists with the Malaria Control Program Technical Working Group, and PNGIMR's work is well appreciated, the geographical distance of PNGIMR's offices in Goroka to the other stakeholders in Port Moresby sometimes makes close interactions difficult. A useful direct contribution in this context was the adjustment of targets following phase 1 review. When the survey data available so far from PNGIMR and Rotarians Against Malaria was taken into consideration, several outcome indicators were adjusted.

The delegation of the overall program evaluation to a national research institution seems a viable arrangement in a setting where such an institution exists and where the NDoH lacks the technical capacity to conduct its own thorough and sound evaluation. In PNG, the division of work between PNGIMR and the implementing partners including NDoH comes naturally, allowing both sides to contribute their expertise in a complementary way. Similar arrangements are also known from more developed countries where program evaluations and operational research activities

are often contracted out to specialized organizations or companies. At the same time, leadership and technical capacity for monitoring and research, including disease surveillance through the NHIS, needs to be strengthened at NDoH. In fact, this capacity is crucial for health planning at national and sub-national levels beyond individual disease-focused programs and projects. External program evaluations can only complement, but not replace, a national health information system. Efforts to strengthen the NHIS at NDoH could tap into the human resource and technical-scientific capacities developed by partner institutions such as PNGIMR. A prerequisite for such arrangements would be good institutional relationships at all levels and mutual understanding of each other's roles and contributions based on trust and open communication.

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REFERENCES

1 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Strategic Investments for Impact: Global Fund Results Summary 2012. Geneva: Global Fund, 2012.
2 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Performance-based Funding at the Global Fund. Geneva: Global Fund, 2009:6.
3 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Monitoring and Evaluation Toolkit: HIV, Tuberculosis, Malaria and Health and Community Systems Strengthening. Fourth edition. Geneva: Global Fund, 2011.
4 **de Savigny D, Adam T.** Systems Thinking for Health Systems Strengthening. Geneva: World Health Organization, 2009.
5 **Garrett L.** The challenge of Global Health. *Foreign Affairs* 2007;56:14-38.
6 **Warren AE, Wyss K, Shakarishvili G, Atun R, de Savigny D.** Global health initiative investments and health systems strengthening: a content analysis of global fund investments. *Global Health* 2013;9:30.
7 **Papua New Guinea Department of Health.** National Malaria Control Program Strategic Plan 2009-2013. Port Moresby: Department of Health, 2009.
8 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Community-based Malaria Prevention and Control in Papua New Guinea – Nationwide Insecticide-treated Nets (ITN) Distribution and Expansion of Confirmed Diagnosis and Appropriate Treatment of Malaria. Grant Number: PNG-304-G01-M. Geneva: Global Fund. www.theglobalfund.org/grantDocuments/PNG-304-G01-M_GA_1_en
9 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Papua New Guinea PNG-304-G01-M Grant Performance Report. Geneva: Global Fund, 2011. www.theglobalfund.org/grantDocuments/PNG-304-G01-M_GPR_0_en
10 **Cibulskis RE, Hiawalyer G.** Information systems for health sector monitoring in Papua New Guinea. *Bull World Health Organ* 2002;80:752-758.
11 **Kurumop SF, Pulford J, Mueller I, Siba PM, Hetzel MW.** Diagnostic capacity and antimalarial availability in Papua New Guinea before the introduction of a revised national malaria treatment protocol. *PNG Med J* 2014;57:59-67.
12 **Hetzel MW, Paul S, Benjamin L, Makita L, Mueller I, Siba PM.** The proportion of fevers attributable to malaria varies significantly between sites in Papua New Guinea. *PNG Med J* 2014;57:39-51.
13 **Bauze AE, Tran LN, Nguyen KH, Firth S, Jimenez-Soto E, Dwyer-Lindgren L, Hodge A, Lopez AD.** Equity and geography: the case of child mortality in Papua New Guinea. *PLoS One* 2012;7:e37861.
14 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.
15 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Malaria Control in Papua New Guinea: Scaling up for Impact. Geneva: Global Fund. www.portfolio.theglobalfund.org/en/Grant/Index/PNG-809-G04-M
16 **Independent State of Papua New Guinea.** Statutes, Chapter 166. Institute of Medical Research Act 1967. Port Moresby: Independent State of Papua New Guinea, 1975.
17 **Reeder JC.** Health research in Papua New Guinea. *Trends Parasitol* 2003;19:241-245.
18 **Reeder JC, Taime J.** Engaging the community in research: lessons learned from the malaria vaccine trial. *Trends Parasitol* 2003;19:281-282.
19 **Vallely A, Betuela I, Greenhill A, Pomat W, Hetzel M, Pulford J, Horwood P, Mueller I, Siba P.** Papua New Guinea Institute of Medical Research: Scientific Report 2006-2011. Goroka: Papua New Guinea Institute of Medical Research, 2012.
20 **Habicht JP, Victora CG, Vaughan JP.** Evaluation designs for adequacy, plausibility and probability of public health programme performance and impact. *Int J Epidemiol* 1999;28:10-18.
21 **National Statistical Office of Papua New Guinea.** PNG 2000 National Census. Port Moresby: National Statistical Office, 2000.
22 **Roll Back Malaria Partnership.** Malaria Indicator Survey (MIS) Toolkit. Geneva: World Health Organization. www.rollbackmalaria.org/toolbox/tool_MISToolkit.html
23 **Temu P, Chen PCY.** The Papua New Guinea vision of Healthy Islands. *Pacific Health Dialog* 1999;6:253-258.
24 **Rosanas-Urgell A, Mueller D, Betuela I, Barnadas C, Iga J, Zimmerman PA, del Portillo HA, Siba P, Mueller I, Felger I.** Comparison of diagnostic methods for the detection and quantification of the four sympatric *Plasmodium* species in field samples from Papua New Guinea. *Malar J* 2010;9:361.
25 **Barnadas C, Kent D, Timinao L, Iga J, Gray LR, Siba P, Mueller I, Thomas PJ, Zimmerman PA.** A new high-throughput method for simultaneous detection of drug resistance associated mutations in *Plasmodium vivax dhfr*, *dhps* and *mdr1* genes. *Malar J* 2011;10:282.
26 **Carnevale EP, Kouri D, DaRe JT, McNamara**

DT, Mueller I, Zimmerman PA. A multiplex ligase detection reaction-fluorescent microsphere assay for simultaneous detection of single nucleotide polymorphisms associated with *Plasmodium falciparum* drug resistance. *J Clin Microbiol* 2007;45:752-761.

27 **World Health Organization, Papua New Guinea Department of Health.** Health Services Delivery Profile Papua New Guinea 2012. Port Moresby: Papua New Guinea Department of Health, 2012.

28 **Kakazo M, Lehmann D, Coakley K, Gratten H, Saleu G, Taime J, Riley ID, Alpers MP.** Mortality rates and the utilization of health services during terminal illness in the Asaro Valley, Eastern Highlands Province, Papua New Guinea. *PNG Med J* 1999;42:13-26.

29 **Belkin JN.** The Mosquitoes of the South Pacific (Diptera: Culicidae). Berkeley, California: University of California Press, 1962.

30 **Lee DJ, Hicks MM, Griffiths M, Debenham ML, Bryan JH, Russell RC, Geary M, Marks EN.** The Culicidae of the Australasian Region. Volume 4. Canberra: Australian Government Publishing Service, 1987.

31 **Beebe NW, Saul A.** Discrimination of all members of the *Anopheles punctulatus* complex by polymerase chain reaction-restriction fragment length polymorphism analysis. *Am J Trop Med Hyg* 1995;53:478-481.

32 **Wirtz RA, Burkot TR, Graves PM, Andre RG.** Field evaluation of enzyme-linked immunosorbent assays for *Plasmodium falciparum* and *Plasmodium vivax* sporozoites in mosquitoes (Diptera: Culicidae) from Papua New Guinea. *J Med Entomol* 1987;24:433-437.

33 **Williams CB.** The use of logarithms in the interpretation of certain entomological problems. *Ann Appl Biol* 1937;24:404-414.

34 **Karunajeewa HA, Mueller I, Senn M, Lin E, Law I, Gomorrai S, Oa O, Griffin S, Kotab K, Suano P, Tarongka N, Ura A, Lautu D, Page-Sharp M, Wong R, Salman S, Siba P, Ilett KF, Davis TME.** A trial of combination antimalarial therapies in children from Papua New Guinea. *N Engl J Med* 2008;359:2545-2557.

35 **Genton B, D'Acremont V, Rare L, Baea K, Reeder JC, Alpers MP, Müller I.** *Plasmodium vivax* and mixed infections are associated with severe malaria in children: a prospective cohort study from Papua New Guinea. *PLoS Med* 2008;5:e127.

36 **Betuela I, Bassat Q, Kiniboro B, Robinson LJ, Rosanas-Urgell A, Stanisic D, Siba PM, Alonso PL, Mueller I.** Tolerability and safety of primaquine in Papua New Guinean children 1 to 10 years of age. *Antimicrob Agents Chemother* 2012;56:2146-2149.

37 **World Health Organization.** Methods for Surveillance of Antimalarial Drug Efficacy. Geneva: World Health Organization, 2009.

38 **Barnadas C, Koepfli C, Karunajeewa HA, Siba PM, Davis TM, Mueller I.** Characterization of treatment failure in efficacy trials of drugs against *Plasmodium vivax* by genotyping neutral and drug resistance-associated markers. *Antimicrob Agents Chemother* 2011;55:4479-4481.

39 **World Health Organization.** Emergency Response to Artemisinin Resistance in the Greater Mekong Subregion. Geneva: World Health Organization, 2013.

40 **Pulford J, Oakiva T, Angwin A, Bryant M, Mueller I, Hetzel MW.** Indifferent to disease: a qualitative investigation of the reasons why some Papua New Guineans who own mosquito nets choose not to use them. *Soc Sci Med* 2012;75:2283-2290.

41 **Hetzel MW, Page-Sharp M, Bala N, Pulford J, Betuela I, Davis TME, Lavu E.** Quality of antimalarials and antibiotics in Papua New Guinea: a survey of the health facility supply chain. *PLoS Med* 2014;9:e96810.

42 **Health Metrics Network, World Health Organization.** Framework and Standards for Country Health Information Systems. Second edition. Geneva: World Health Organization, 2008.

43 **Kitur U.** Health information challenges for Papua New Guinea. *Pac Health Dialog* 2012;18:29-31.

44 **Rudge JW, Phuanakoonon S, Nema KH, Mounier-Jack S, Coker R.** Critical interactions between Global Fund-supported programmes and health systems: a case study in Papua New Guinea. *Health Policy Plan* 2010;25(Suppl1):i48-i52.

45 **Mueller I, Slutsker L, Tanner M.** Estimating the burden of malaria: the need for improved surveillance. *PLoS Med* 2011;8:e1001144.

46 **Whittaker M, Thomason J.** Working together for a better future. *PNG Med J* 2009;52:179-186.

47 **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.

48 **Day B.** The primacy of politics: charting the governance of the Papua New Guinea health system since Independence. *PNG Med J* 2009;52:130-138.

49 **Hetzel MW, Obrist B, Lengeler C, Msechu JF, Nathan R, Dillip A, Makemba AM, Mshana C, Schulze A, Mshinda H.** Obstacles to prompt and effective malaria treatment lead to low community-coverage in two rural districts of Tanzania. *BMC Public Health* 2008;8:317.

50 **Pettersson H.** Design of master sampling frames and master samples for household surveys in developing countries. In: Department of Economic and Social Affairs Statistics Division, United Nations. Household Sample Surveys in Developing and Transition Countries. New York: United Nations, 2005:71-94.

51 **Roll Back Malaria, MEASURE Evaluation, World Health Organization, UNICEF.** Guidelines for Core Population Coverage Indicators for Roll Back Malaria: To Be Obtained from Household Surveys. Calverton, Maryland: MEASURE Evaluation, 2006.

52 **Chromy JR, Abeyasekera S.** Statistical analysis of survey data. In: Department of Economic and Social Affairs Statistics Division, United Nations. Household Sample Surveys in Developing and Transition Countries. New York: United Nations, 2005:389-417.

53 **Allen BJ.** The geography of Papua New Guinea. In: Attenborough RD, Alpers MP, eds. Human Biology in Papua New Guinea: The Small Cosmos. Oxford: Clarendon Press, 1992:36-66.

54 **Lin E, Kiniboro B, Gray L, Dobbie S, Robinson L, Laumaea A, Schöpflin S, Stanisic D, Betuela I, Blood-Zikursh M, Siba P, Felger I, Schofield L, Zimmerman P, Mueller I.** Differential patterns of infection and disease with *P. falciparum* and *P. vivax* in young Papua New Guinean children. *PLoS One* 2010;5:e9047.

55 **Beer N, Ali AS, Eskilsson H, Jansson A, Abdul-Kadir FM, Rotllant-Estelrich G, Abass AK, Wabwire-Mangen F, Björkman A, Källander K.** A qualitative study on caretakers' perceived need of bed-nets after reduced malaria transmission in Zanzibar, Tanzania. *BMC Public Health* 2012;12:606.

56 **Yohannes K, Dulhunty JM, Kourleoutov C,**

Manuopangai VT, Polyn MK, Parks WJ, Williams GM, Bryan JH. Malaria control in central Malaita, Solomon Islands. 1. The use of insecticide-impregnated bed nets. *Acta Trop* 2000;75:173-183.

57 **World Health Organization.** World Malaria Report 2012. Geneva: World Health Organization, 2012.

58 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

59 **Angwin A, Hetzel MW, Mueller I, Siba PM, Pulford J.** A qualitative study of how affected individuals or their caregivers respond to suspected malaria infection in rural Papua New Guinea. *PNG Med J* 2014;57:30-38.

60 **Katusele M, Gideon G, Thomsen EK, Siba PM, Hetzel MW, Reimer LJ.** Long-lasting insecticidal nets remain efficacious after five years of use in Papua New Guinea. *PNG Med J* 2014;57:86-93.

61 **Ochomo EO, Bayoh NM, Walker ED, Abongo BO, Ombok MO, Ouma C, Githeko AK, Vulule J, Yan G, Gimnig JE.** The efficacy of long-lasting nets with declining physical integrity may be compromised in areas with high levels of pyrethroid resistance. *Malar J* 2013;12:368.

62 **Hetzel MW, Pulford J, Tandrapah A, Jamea-Maiasa S; PNGIMR MalCon Team.** Missing in the line of duty. *PNG Med J* 2014;57:94-102.

PNG Med J 2014 Mar-Dec;57(1-4):30-38

A qualitative study of how affected individuals or their caregivers respond to suspected malaria infection in rural Papua New Guinea

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SUMMARY

This paper presents findings from a qualitative study that sought to identify the ways in which affected individuals or their caregivers respond to a suspected malaria infection and to illuminate the rationale underlying the decision-making process. In-depth interviews (IDIs) were conducted with a sub-sample (n = 44) of participants in a country-wide household survey who reported experiencing, or caring for someone who experienced, a suspected malaria infection in the two weeks before the survey. All IDIs were completed between March and July 2011. Analysis was informed by a general inductive methodology. The most commonly reported response involved the use of antimalarial medication and some form of traditional remedy prepared in the home. Reported treatment responses were frequently consistent with a stepped-care approach to disease management, the first step of which was characterized by convenience and was often relatively generic in nature. Seeking assistance from a formal health care provider was the exception amongst study participants, with fewer than half attending a health facility during the target illness episode. A number of barriers to health service access were reported; however, a range of other factors contributed to the decision not to seek formal health care such as perceived severity of illness, positive past experiences using home-based treatments and the aforementioned preference for utilizing convenient 'treatment' options in the first instance. Traditional healers were rarely considered an appropriate treatment option for malaria.

Introduction

A range of treatment options are potentially available to individuals with malaria infection in Papua New Guinea (PNG). Formal health care services are provided through an extensive network of government and church-run health facilities. This health facility network comprises four levels of service provision including hospital, health centre, health subcentre and aid post. The hospital is the largest unit of health service provision and the aid post is the smallest, typically staffed by a single health worker with two years' clinical training. A wide range of medications, including antimalarials, are accessible without prescription from pharmacies and smaller retail stores, even at district or village level. A belief in supernatural causes of disease is still widespread in PNG and for this reason traditional healers are often sought in response to illness and many forms of

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traditional remedy are commonly used (1).

The few studies that have examined treatment seeking for febrile illness or suspected malaria in PNG have reported somewhat inconsistent findings. Treating suspected malaria within the home has been previously identified as the normative response amongst the Nasioi people of Bougainville (2,3), where either traditional remedies, western medicines or both may be employed. If further assistance is needed, then traditional healers are sought more often than formal health care providers in this context, even in instances where formal health care services are available. Conversely, a recent household survey conducted in the Madang and East Sepik Provinces identified utilization of formal health care services as the most common response to suspected malaria in both sites (4). A much earlier study also found that rural householders in Madang Province were less likely to treat suspected malaria at home if they had access to village-based aides trained to presumptively treat fever with a three-day course of antimalarials (5).

Distance to the nearest health facility has been reported as a factor in whether formal treatment is sought for suspected malaria or not (6), although more recent evidence suggests social and family responsibilities may be a bigger obstacle to formal treatment seeking than distance per se (4). Similarly, practical considerations such as cost, availability of transport and convenience have been identified as key determinants in the treatment-seeking decision within PNG (2,3). These findings, although limited in size and scope, suggest that possible regional differences in malaria treatment-seeking behaviour exist in PNG and that multiple factors other than proximity to formal health care may influence an individual's response to malaria infection. Reviews of the international evidence further highlight substantial variation in malaria treatment-seeking behaviours and emphasize the importance of examining the response to malaria in local contexts (7,8).

PNG, with financial support from the Global Fund to Fight AIDS, Tuberculosis and Malaria (GFATM), has substantially strengthened malaria control activities in recent years. Major changes to the national malaria control program include, amongst other things, the expansion of microscopy or malaria rapid diagnostic test (RDT) diagnosis and the introduction of the artemisinin-based combination therapy (ACT) artemether-lumefantrine (AL) for the first-line treatment of uncomplicated malaria (9). These changes represent a substantial improvement in malaria case management, which previously relied on presumptive diagnosis (10) and a first-line treatment of chloroquine and sulphadoxine-pyrimethamine, to which widespread parasite resistance has developed (11). Advancing current understanding of contemporary treatment-seeking behaviour for suspected malaria infection may, therefore, usefully inform strategies to promote greater and/or faster access to diagnostic testing and the highly efficacious antimalarial AL (12). Accordingly, this paper presents findings from a qualitative study of how affected individuals or their caregivers respond to a suspected malaria infection in rural PNG. The reported findings were drawn from in-depth interviews (IDIs) with individuals (or their caregivers) residing in village locations who reported experiencing a suspected malaria episode within the two weeks before their participation in the study. Research questions included the following. What actions, inclusive of self-care strategies, are taken in response to suspected malaria infection? What alternative response options are available but not utilized? What influences whether an available response option is utilized or not?

Methods

Sample size and selection

The study took place in two coastal provinces (Gulf and West Sepik) and one island province (New Ireland) of PNG. The study sample included participants from a country-wide household survey (HHS) of malaria prevalence and treatment-seeking behaviour. The HHS was conducted in 30 randomly selected households in each of four randomly selected villages in every province in PNG. Interviews were sought from a sub-sample of HHS participants. Potential participants were purposively sampled based on age, pregnancy status and gender characteristics. In order to be eligible for inclusion participants must have been included in the HHS sample and, in response to structured survey questions, reported fever or malaria symptoms in the two weeks before the survey inclusive of the survey day. Only one participant per household was interviewed if more than one householder met

the eligibility criteria.

Procedure

A scientific officer trained in in-depth interviewing accompanied the HHS team to most survey villages in the three provinces listed above. The scientific officer was alerted by members of the survey team whenever an HHS participant met the inclusion criteria. The scientific officer then sought verbal informed consent from the respective individual to participate in an IDI. Where appropriate, consent was initially sought from the household head (in cases where the household head was not the individual being sought for interview). Consent from the household head or guardian was also sought in cases where the eligible individual was less than 16 years of age. The interview was conducted with the household head or guardian in cases where the eligible individual was less than 12 years of age. Interviews were 30-60 minutes in length and were conducted on site at the participant's household or preferred location. Participants were not compensated for completing the IDI. The scientific officer conducting the interviews was female, aged in her mid-20s, of Papua New Guinean descent and conversant in two of the three national languages of PNG. Whilst sharing a common social background with the majority of participants, the interviewer considered herself ethnically distinct from all study participants. This study was approved and granted ethical clearance by the Medical Research Advisory Committee of Papua New Guinea (MRAC No 10.12; 26 Feb 2010).

Interview focus

All interviews started with a timeline follow-back (TLFB)-directed review of the most recent fever or suspected malaria episode. The TLFB is a calendar-based method for assessing the occurrence of specified events or behaviours over a specified period and is widely used in behavioural health research (13,14). Once the time period of the physical illness had been identified then all actions taken in response to the fever episode were explored and recorded. This exploration was guided by an interview schedule that covered illness duration, any actions taken over the previous two weeks in response to perceived malaria infection, alternative response actions known or available to the participants that were not utilized and the underlying rationale for reported actions or non-actions in response to the fever episode.

Data analysis

All interviews were recorded on a digital voice recorder, transcribed verbatim, translated into English and entered into NVIVO 9 for subsequent analysis. The data analysis was informed by a general inductive methodology (15). All interview data were independently coded by a scientific officer (BA graduate) and her supervisor (PhD graduate). Coding was conducted over two cycles with the coders meeting to compare and agree upon codes and emerging themes at the end of each cycle, resolving disagreements by consensus opinion or by the creation of new, mutually agreeable, codes/themes. The data model presented below was devised in draft form at the conclusion of the first coding cycle and subsequently refined during the second. Preliminary coding was conducted during the data collection period, thereby allowing emerging themes to be further explored during subsequent interviews and refined or reconfigured as appropriate.

Quotes from participant interviews are presented throughout the results section in italics. To protect participant identities, only the province, status (focus individual/caregiver), gender (of 'patient') and age (of 'patient') of each quoted individual are reported.

Results

Sample characteristics

A total of 44 in-depth interviews were completed in New Ireland (n = 30), West Sepik (n = 8) and Gulf (n = 6) provinces. 33 of these interviews were conducted with caregivers and 11 with the individual who experienced the febrile illness. The focus individuals (those who experienced the febrile illness) were mostly female (n = 24); 19 were aged between 0 and 5 years, 13 between 6 and 15 years and 12 were 16 years of age or older. 2 of the focus individuals were pregnant. Interviews were conducted between March and July 2011.

Illness duration, type and cause

All focus individuals reported experiencing a febrile illness at some point in the 14 days before their interview. The illness duration was 5 days or fewer in 30 cases and between 6

and 11 days in the remaining cases; however, 9 participants reported ongoing illness at the time of the interview. When asked to report what type of illness they or the focus individual had, 39/44 participants reported malaria, 2 reported malaria and pneumonia or cough and 3 reported that they did not know. 16 participants identified the mosquito as the primary cause of their illness. A further 14 participants attributed their illness to lack of personal and/or environmental hygiene, 8 to cold weather or wind, 1 to her spleen and 5 stated that they did not know.

Response options taken

Table 1 presents the reported response options taken by the study participants, by province and overall. As shown, the most common response options reported overall and by province were the use of an antimalarial or other medication (eg, Panadol, amoxycillin) or some form of traditional remedy prepared in the home. Not included in Table 1 were basic self-care strategies employed in the home such as taking bed rest or applying a wet sponge to the head, as these were ubiquitous across the study sample.

Of the 30 participants who consumed an antimalarial, 18 received them from a health facility and 12 utilized an existing home supply, obtained them from neighbours or purchased them from a retail outlet. In 15 cases, the antimalarial was consumed on the first day of illness, in 11 cases on the second day of illness and in 4 cases on the third or later day of illness. Of the 18 participants who visited a health facility, 10 did so on the first day of illness, 5 on the second day and 3 on the third day or later. Only 7 participants reported seeking or receiving assistance from someone living outside of their immediate home (excluding health worker assistance). In 4 of these cases the assistance was from a family member, whilst the other 3 sought assistance from a friend, neighbour and traditional healer, respectively.

Common characteristics of the reported response options taken

A number of common characteristics with respect to fever response were evident across the interview transcripts and across sites. Firstly, most participants reported employing multiple actions in response to the perceived fever episode. In many cases this involved the use of a traditional remedy and modern medicine, either concurrently – "*When he took his first dose [antimalarial medication] on Monday his fever went up so I had to steam bath him to bring his temperature down."* (New Ireland, caregiver, male, 12 years) – or in succession – "*I had treated him with lemon grass on the first day of his illness [Monday], I waited and [given no improvement] gave him medicine on Tuesday.*" (New Ireland,

TABLE 1

Reported fever response options taken by study participants, by province

Province	Reported response options					
	Health facility	**Antimalarial medication**	**Other medication**	**Traditional remedy**	**Traditional healer**	**Prayer**
New Ireland (n = 30)	11	21	22	19	1	3
West Sepik (n = 8)	3	4	4	6	0	0
Gulf (n = 6)	4	5	5	4	0	0
Total (n = 44)	18	30	31	29	1	3

caregiver, male, 10 years). When multiple actions were reported, they were typically done in a manner consistent with a stepped-care approach to disease management. In a stepped-care approach the least intensive/ expensive responses are employed in the first instance and followed up with more intensive/ expensive approaches if the symptoms persist or deteriorate. For example, "*We would use the lemon grass first and if it doesn't help, we would buy medicine for them and if that doesn't help, we would take them to a health facility*." (New Ireland, caregiver, female, 4 years).

The initial fever response frequently involved some form of self-care approach conducted independently within the home environment. A common form of self-care involved taking a cold or warm bath or applying a wet sponge to reduce body temperature: "*When she had fever I washed her with warm water and put this wet towel on her head*." (New Ireland, caregiver, female, 5 months). Traditional remedies involving a herbal drink or herbal bath were also widely employed in the early stages of illness: "*We use the new shoots of the pawpaw tree to steam bath and drink its flower*." (West Sepik, female, 23 years). In other cases, the self-care response involved taking modern medicine purchased from local stores or pharmacies and stored for emergency cases, obtained from neighbours or left over from a previous malaria episode: "*I have some medicines which I have bought at the chemist. I gave her chloroquine medicine for three days*." (New Ireland, caregiver, female, 9 years). Initial response options were often notable for their apparent convenience: "*It is quite far to go to the hospital so we use the herbs*." (West Sepik, caregiver, female, 7 years). Many of the reported responses were also characterized by their relatively generic nature; the same response could be potentially employed to treat a range of common ailments: *"When my child has fever, cough, vomit or diarrhoea, the dada leaf helps to heal him fast*." (New Ireland, caregiver, female, 4 months).

Deciding upon which response option(s) to take

As indicated above, response options were often chosen for their convenience and relatively generic nature; however, a multitude of other factors were evident in the decision-making process over the illness course. Antimalarial medications were widely used by the study participants as they were generally considered an effective and appropriate treatment for malaria. Indeed, a number of participants explicitly stated that antimalarials were *the* most effective and appropriate treatment for malaria: "*We consider medicines as the very first option. Medicine is best*." (Gulf, caregiver, female, 6 months). Nevertheless, a more common view was that antimalarial medication and traditional remedies complemented each other, each playing a different role in resolving the illness episode. Typically, the perceived role of the traditional remedy was symptom reduction whilst the antimalarial provided the cure. The following quote is exemplary in this regard: "*Steam bathing the child with lemon grass is not for curing the sick, but it is just used to cool down the fever and later we will bring the child to the hospital for proper medication. Only medicine will cure the sick completely*." (West Sepik, caregiver, male, 1 year). For other participants traditional remedies were utilized as they were less expensive and/or more convenient than antimalarial medication; in effect, they were a less preferable, but more accessible 'treatment' option: "*When there is no time to go to the health facility or there is no medicine, we just use the herbs*." (New Ireland, female, 20 years).

A few participants considered traditional remedies to be a more effective treatment for malaria than medication, although they were in the minority. More frequently reported was the belief that traditional remedies were more appropriate (than antimalarials) for febrile illness resulting from sorcery, or offered protection against the possibility that the illness resulted from such: *"We use herbs because it is our traditional belief or in case the sickness is caused by some superstitious actions. Therefore, we apply both the herbs and the medicines*." (Gulf, caregiver, female, 6 months).

Whilst the majority of participants across sites used or were open to using traditional remedies, very few consulted a traditional healer, even though one or more traditional healers were available to all. When asked why this was the case, it was suggested that traditional healers are not used to treating malaria infection: "*They [traditional healers] do not cure fever. Medicine helps in curing fever or sickness*." (West Sepik, caregiver, female, 5 months). Nevertheless, one participant

sought help from a traditional healer before seeking assistance from a health worker and a number of participants stated that they would seek assistance from a traditional healer if antimalarials proved ineffective: "*When we go to the hospital and there's nothing the doctors can do then we take our children and go see her [the traditional healer].*" (New Ireland, caregiver, male, 12 years).

Health facilities were utilized by fewer than half of the study participants even though all reported the existence of a health facility that could be accessed if required. Participants identified a range of barriers to treatment access when queried why this was the case. Commonly reported barriers included issues pertaining to cost, unreliable drug supply, health worker attitudes and attendance, quality of care and health facility proximity, as indicated by the following interview excerpts:

> "*I don't like to go quickly to the hospital because it costs money for this purpose: like paying for medicine, hiring a vehicle to get to the hospital. So I must make use of what is available at home to relieve the malaria."* (New Ireland, caregiver, male, 5 months);
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> "*Most times when we go to [hospital name] they tell us that there is shortage of drugs and they tell us to go and buy amoxycillin and Panadol from the trade stores.*" (New Ireland, female, 20 years);
>
> "*They [health workers] complain and argue with us all the time while giving out treatment. They have a poor relationship with the patients.*" (Gulf, female, 25 years);
>
> "*One thing, the health worker is not always present at the aid post. He is always out in the bush.*" (New Ireland, caregiver, female, 9 years);
>
> "*They do not make proper examinations. Therefore we the parents sometimes argue with them.*" (West Sepik, female, 23 years);
>
> "*We do not live close to town where we can seek help quickly.*" (West Sepik, caregiver, female, 1 year).

Cost was not uniformly prohibitive as many participants reported making a payment of some kind during the recent illness episode, whether in the form of transport costs to the nearest health centre – "*We pay K9.00 and K1.00 to go to [health centre name]. So that's K10 for getting there and a total of K20 for return."* (West Sepik, caregiver, male, 1 year); outpatient fees at the local health centre – "*The nurses give us medication, when we pay the fees.*" (New Ireland, caregiver, male, 6 months); or for medication purchased at a local store – *"The hospital referred me to buy the medicine from Chemcare, which cost K50 for a full packet.*" (West Sepik, female, 35 years). Concern regarding quality of care was not solely directed towards formal health service provision. For example, when asked why she did not seek antimalarials from friends or family members known to have a supply, the following participant replied: "*I don't go and ask because I fear the medicine they give might be expired."* (Gulf, female, 25 years).

The perceived severity of illness was also found to influence participant decision-making. For example, many caregivers or focus individuals recognized the need to treat fever promptly when life-threatening symptoms were noticed: "*If it is a serious illness we go to the hospital for proper examination like blood testing.*" (West Sepik, caregiver, female, 7 years). However, mild symptoms were treated with less urgency, often by traditional remedy: *"I knew that my illness was not that serious that's why I did not go to the hospital.*" (Gulf, female, 25 years). Seeking formal health care or more intensive treatment responses was also often given a low priority, further suggesting that a perceived malaria infection was not always considered a severe illness: "*I was just being lazy and could not do those things [response options available to the participant, but not taken] to treat the child's fever."* (New Ireland, caregiver, female, 6 months). Response decisions were also influenced by past experience in managing the same or similar illnesses. Past response options that had proven effective were trusted and employed again: "*I personally wanted to use lemon grass because I have used it before and it has helped to cure my child's illness.*" (New Ireland, caregiver, female, 2 years). Conversely, past response options that proved ineffective or were in some way undesirable were avoided: "*The tree leaves I mentioned before were shown to me by my mother and I drank it once and it was bitter so now I only drink lemon [grass]."* (New Ireland, female, 50 years).

Discussion

The reported findings suggest that medical pluralism, or the use of various treatment types, is a normative response to perceived malaria episodes in rural locations across PNG. The pluralistic response most common to participants in this study involved the use of antimalarial medication and some form of traditional remedy prepared in the home. Reported treatment responses were also frequently consistent with a stepped-care approach to disease management, the first step of which was characterized by convenience and was often relatively generic (rather than disease specific) in nature. The findings further suggest that antimalarial medication is widely considered the 'best' treatment for perceived malaria infection, with traditional remedies typically valued as a complement to antimalarial medication or a viable alternative when antimalarial medication cannot be obtained. Traditional healers were rarely considered an appropriate option for malaria, although may potentially be consulted if the suspected illness aetiology shifts towards sorcery or the spiritual realm. Seeking assistance from a formal health care provider was the exception amongst study participants, with fewer than half attending a health facility during the target illness episode. A number of barriers to health service access were reported, which undoubtedly deterred formal treatment seeking for many participants. However, a range of other factors contributed to the decision not to seek formal health care such as perceived severity of illness, positive past experiences using home-based treatments and the aforementioned preference for utilizing convenient 'treatment' options in the first instance. Thus, improved access to formal health care services alone (at least not without simultaneous improvement in quality of care) might not have greatly increased health service utilization among study participants.

These findings should be considered in light of the limitations inherent in the study methodology. Study data were collected via in-depth interview from a relatively small number of participants collectively residing in only 3 of the 20 (at the time of data collection) provinces in PNG. Thus, the reported findings (as is consistent with a qualitative methodology) are not readily generalizable beyond the study participants themselves. The 30-day TLFB method has not been validated as a method for documenting treatment-seeking behaviour in a PNG context and participant recall may have been inaccurate at times and/or participant responses may have been subject to some form of social desirability bias. Assurance of participant confidentiality, the practice of reviewing documented treatment-seeking behaviours at the conclusion of each TLFB and the relatively non-threatening nature of the research topic are likely to have protected against these methodological concerns, although they cannot be ruled out entirely.

Despite these limitations, the reported findings extend current understanding of malaria treatment-seeking behaviour in PNG. The apparent preference for convenient, home-based responses to perceived malaria infection in the first instance (including the use of antimalarials obtained from an existing or easily accessible source) is consistent with earlier reports of disease management in PNG (16) and the reported malaria treatment-seeking behaviour of the Nasioi people of Bougainville (2,3). However, if the home-based response was unsuccessful, Nasioi were most likely to seek assistance from a traditional healer as a next step, a trend not evident among participants in this study. This discrepancy is possibly explained by potential cultural differences in the perceived role of sorcery or spirits in malaria aetiology, with traditional healers more likely to be sought if an illness is thought to be grounded in sorcery or the spiritual realm (1). Alternatively, the greater use of traditional healers among the Nasioi people may be a consequence of the breakdown of formal health care service provision during the armed conflict in Bougainville in the 1990s (3). The findings reported above also differ from the study of malaria treatment seeking completed by Davy and colleagues (4), which identified utilization of formal health care services as the most common response to perceived malaria infection in villages within the Madang and East Sepik Provinces of PNG. Self-help or home-based remedies did not appear to be a defined 'treatment' option in the Davy and colleagues study, which may account for this difference in part. In addition, many participants in the Davy and colleagues study lived within a 30-minute walk of a health centre, which may further account for the apparent difference in health service access.

One area in which past and present studies

of treatment seeking for suspected malaria in PNG agree is that perceived illness severity, or the prioritization of tasks/considerations not related to the illness episode, exerts considerable influence on the treatment-seeking process. For example, earlier studies have reported 'illness severity', 'lack of motivation' and 'the condition was not serious' as the most common reason for not seeking treatment (3,4). These collective findings suggest that formal help seeking for suspected malaria infection may be considered unnecessary in many cases due to a perception of low disease risk. This possibility is supported by a recent qualitative study examining reasons for mosquito net use among net-owning individuals in PNG. Pulford and colleagues (17) identified indifference, grounded in a relative lack of concern regarding the risk or consequences of malaria infection, as a primary barrier to mosquito net use. Studies of malaria treatment-seeking behaviour from other parts of Melanesia and elsewhere have also reported that febrile illness is often perceived as low risk and formal health care unnecessary (18-21).

Initial reluctance to seek help for suspected malaria infection may also be related to lay understanding, and use, of the term 'malaria'. Participant responses suggest that they may not always have a clear understanding of malaria aetiology or, perhaps more likely, that the term 'malaria' may be commonly applied to a wide range of symptoms that include, but are not exclusive to, a biomedical definition of malaria infection. Thus various symptom profiles may collectively be described as malaria, but formal health care is only sought when a particular set of symptoms (presumably considered more severe) are present. Similar issues with malaria (or febrile illness) classification have been reported elsewhere (19,22), including among the Nasioi of Bougainville (2) and in Tafea Province, Vanuatu (20). The common health worker practice of presumptively treating all fever cases with antimalarials may further confuse lay understanding of malaria classification in PNG (10), especially as fewer than 50% of fever cases presenting to health facilities in PNG test positive for malaria parasitaemia (23).

Home-based management of malaria (HMM) programs may hold particular appeal as an alternative to formal health services for the treatment of malaria as antimalarial medication is a desired treatment response and would likely be sought out if readily available in the local community. HMM programs have proven highly acceptable in a range of contexts (24) and have been a demonstrably popular treatment option in Madang Province, PNG in the past (5). Furthermore, as the treatment options reported by participants in this study frequently required financial expenditure at some point, a fee-based model of HMM may be as acceptable as a free (yet potentially less sustainable) alternative. One such fee-based model has recently been established in PNG with considerable success (25). Nevertheless, as participants in this study expressed concerns with quality of medical care and frustrations when health care access was limited due to such things as antimalarial stock-outs or staff unavailability, then HMM programs may flounder if they are not perceived to be a trustworthy and reliable treatment source. Health promotion/ behaviour change messages that encourage seeking a diagnostic test and AL (if positive) within 24 hours of symptom onset may further bolster support for community-based HMM programs or encourage greater utilization of existing health services. A health promotion campaign of this nature will likely require continuous and extensive exposure to achieve lasting behaviour change and could potentially benefit by promoting traditional remedies as a complement to AL, rather than a first step following symptom onset.

Finally, investing in improved quality of health care service provision (given participant concerns) might result in more frequent attendance at formal health care facilities. Improvements in health service infrastructure, resourcing, clinical supervision and support are likely to promote greater rates of formal treatment seeking and, as has previously been suggested, increase job satisfaction among the health care work force (26).

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REFERENCES

1 **Macfarlane J.** Common themes in the literature on traditional medicine in Papua New Guinea. *PNG Med J* 2009;52:44-53.

2 **Macfarlane JE.** A focused ethnography about treatment-seeking behaviour and traditional medicine in the Nasioi area of Bougainville. *PNG Med J* 2008;51:29-42.

3 **Macfarlane JE, Alpers MP.** Treatment-seeking behavior among the Nasioi people of Bougainville: choosing between traditional and western medicine. *Ethnic Health* 2009;14:147-168.

4 **Davy CP, Sicuri E, Ome M, Lawrence-Wood E, Siba P, Warvi G, Mueller I, Conteh L.** Seeking treatment for symptomatic malaria in Papua New Guinea. *Malar J* 2010;9:268.

5 **Moir JS, Tulloch JL, Vrbova H, Jolley DJ, Heywood PF, Alpers MP.** The role of voluntary village aides in the control of malaria by presumptive treatment of fever. 2. Impact on village health. *PNG Med J* 1985;28:267-278.

6 **Müller I, Smith T, Mellor S, Rare L, Genton B.** The effect of distance from home on attendance at a small rural health centre in Papua New Guinea. *Int J Epidemiol* 1998;27:878-884.

7 **McCombie SC.** Treatment seeking for malaria: a review of recent research. *Soc Sci Med* 1996;43:933-945.

8 **McCombie SC.** Self-treatment for malaria: the evidence and methodological issues. *Health Policy Plan* 2002;17:333-344.

9 **Papua New Guinea Department of Health.** National Malaria Treatment Policy. Port Moresby: Department of Health, Sep 2009.

10 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

11 **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;77:947-954.

12 **Karunajeewa HA, Mueller I, Senn M, Lin E, Law I, Gomorrai PS, Oa O, Griffin S, Kotab K, Suano P, Tarongka N, Ura A, Lautu D, Page-Sharp M, Wong R, Salman S, Siba P, Ilett KF, Davis TME.** A trial of combination antimalarial therapies in children from Papua New Guinea. *N Engl J Med* 2008;359:2545-2557.

13 **Lewis-Esquerre JM, Colby SM, Tevyaw TO, Eaton CA, Kahler CW, Monti PM.** Validation of the timeline follow-back in the assessment of adolescent smoking. *Drug Alcohol Depend* 2005;79:33-43.

14 **Weinhardt LS, Carey MP, Maisto SA, Carey KB, Cohen MM, Wickramasinghe SM.** Reliability of the timeline follow-back sexual behavior interview. *Ann Behav Med* 1998;20:25-30.

15 **Thomas DR.** A general inductive approach for analyzing qualitative evaluation data. *Am J Eval* 2006;27:237-246.

16 **Welsch RL.** The distribution of therapeutic knowledge in Ningerum: implications for primary health care and the use of aid posts. *PNG Med J* 1985;28:205-210.

17 **Pulford J, Oakiva T, Angwin A, Bryant M, Mueller I, Hetzel MW.** Indifferent to disease: a qualitative investigation of the reasons why some Papua New Guineans who own mosquito nets choose not to use them. *Soc Sci Med* 2012;75:2283-2290.

18 **Kamat VR.** "I thought it was only ordinary fever!": cultural knowledge and the micropolitics of therapy seeking for childhood febrile illness in Tanzania. *Soc Sci Med* 2006;62:2945-2959.

19 **Pilkington H, Mayombo J, Aubouy N, Deloron P.** Malaria, from natural to supernatural: a qualitative study of mothers' reactions to fever (Dienga, Gabon). *J Epidemiol Community Health* 2004;58:826-830.

20 **Tynan A, Atkinson JA, Toaliu H, Taleo G, Fitzgerald L, Whittaker M, Riley I, Schubert M, Vallely A.** Community participation for malaria elimination in Tafea Province, Vanuatu: Part II. Social and cultural aspects of treatment-seeking behavior. *Malar J* 2011;10:204.

21 **Yadav SP.** A study of treatment seeking behavior for malaria and its management in febrile children in rural part of desert, Rajasthan, India. *J Vector Borne Dis* 2010;47:235-242.

22 **O'Connell KA, Samandari G, Phok S, Phou M, Dysoley L, Yeung S, Allen H, Littrell M.** "Souls of the ancestor that knock us out" and other tales. A qualitative study to identify demand-side factors influencing malaria case management in Cambodia. *Malar J* 2012;11:355.

23 **Hetzel MW, Paul S, Benjamin L, Makita L, Mueller I, Siba PM.** The proportion of fevers attributable to malaria varies significantly between sites in Papua New Guinea. *PNG Med J* 2014:57:39-51.

24 **Ajayi IO, Browne EN, Garshong B, Bateganya F, Yusuf B, Agyei-Baffour P, Doamekpor L, Balyeku A, Munguti K, Cousens S, Pagnoni F.** Feasibility and acceptability of artemisinin-based combination therapy for the home management of malaria in four African sites. *Malar J* 2008;7:6.

25 **McKay J, Lepani K.** Health System Strengthening in Papua New Guinea: Exploring the Role of Demand-Responsive Mechanisms. Sydney: Lowy Institute for International Policy, 2010.

26 **Jayasuriya R, Whittaker M, Halim G, Martineau T.** Rural health workers and their work environment: the role of inter-personal factors on job satisfaction of nurses in rural Papua New Guinea. *BMC Health Serv Res* 2012;12:156.

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The proportion of fevers attributable to malaria varies significantly between sites in Papua New Guinea

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SUMMARY

Malaria is endemic across lowland Papua New Guinea (PNG) and case management has been based on symptomatic diagnosis and presumptive treatment of fever cases with an antimalarial. This study aimed to investigate the prevalence of malaria infection among fever cases presenting to 5 purposely selected sentinel health facilities in order to estimate the proportion of patients requiring antimalarial drugs. A total of 1807 fever patients were screened. Overall, 45% of fever patients had a positive malaria blood slide; 35% were infected with *Plasmodium falciparum,* 9% with *P. vivax* and 2% with *P. malariae*. Slide positivity was highest in Dreikikir (75%) and lowest in Wipim (2%). Among patients aged 1-4 years, 22% had moderate to severe anaemia (Hb <8 g/dl) and 21% of children 2-9 years of age showed signs of splenomegaly (Hackett score 1-5). Comorbidity differed significantly between study sites and was not closely correlated with malaria infection. Clinical diagnosis by health facility staff was malaria for 67% of all fever cases, including 89% of slide-positive and 48% of slide-negative patients. 70% of rapid diagnostic test-negative cases were treated with an antimalarial. It is estimated that due to the lack of parasitological diagnosis the selected health facilities reported an excess of 18% (Dreikikir) to 98% (Wipim) malaria patients on average each month. In consideration of the significant differences in malaria-attributable fevers between study sites, the implementation of parasitological diagnosis in health facilities and administration of antimalarials only to test-positive patients has the potential to significantly improve the management of fever cases and reporting of malaria. A better tailoring to different settings may increase the effectiveness of malaria control interventions.

Introduction

Malaria is endemic across most parts of Papua New Guinea (PNG) but varying levels of endemicity have been observed in different areas of the country (1,2). Variations in endemicity have been attributed to climatic factors (2), which influence mosquito ecology, development of the *Plasmodium* spp. parasite in the mosquito and consequently transmission of malaria parasites (3), abundance of alternative hosts such as dogs and pigs (4), historic and current control activities including the use of mosquito nets, and treatment-seeking behaviours in the community (5). Significant differences in morbidity have been

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reported even between nearby villages (4).

Malaria treatment, however, has been largely uniform across PNG with most cases of febrile illness being considered malaria and treated with an antimalarial drug. A country-wide household survey conducted in 2008-2009 found that 55.4% of self-reported fever cases in the community received some sort of drug and 38.8% were treated with an antimalarial. Amongst patients attending a health facility during the course of their illness, 74.2% received an antimalarial (6). A country-wide health facility survey undertaken in 2010 found that 96.4% of fevers presenting to health facilities were treated with an antimalarial drug; patients in the more malarious lowland areas were more likely to receive the correct antimalarial prescription than patients in the Highlands Region (7). Malaria was generally diagnosed presumptively based on clinical signs and symptoms – although clinical assessments were generally considered superficial. Even in facilities with available microscopy or rapid diagnostic tests (RDTs), less than 50% of the fever patients were tested, with patients in the Highlands Region being tested more frequently. The malaria test result rarely influenced antimalarial prescription and most of the negative cases still received an antimalarial drug (7).

The present study aimed at investigating the prevalence of malaria infection among fever cases presenting to health facilities in order to estimate the proportion of fevers warranting treatment with antimalarials. The study was part of the evaluation of the Global Fund to Fight AIDS, Tuberculosis and Malaria (GFATM) Round 3 malaria grant to PNG. The study protocol was granted ethical clearance by the Institutional Review Board of the PNG Institute of Medical Research (IMR IRB No 0803) and the PNG Medical Research Advisory Committee (MRAC No 07.30).

Methods

Study sites

Surveillance of malaria cases was established starting in October 2008 in five health facilities in purposely selected sentinel sites across PNG. Selection of the sites was based on operational and epidemiological considerations and included only locations that had not yet been covered with the free distribution campaign of insecticide-treated mosquito nets (ITNs) (8). This resulted in no site being located in the Islands Region provinces, which were all covered with the ITN distribution campaign before mid-2008. Endemic malaria, a functioning and collaborative health centre or health subcentre and reasonable accessibility were prerequisites for selection. The location of the sentinel sites is shown in Figure 1. Selected facilities included three health centres, one health subcentre and one hospital (Table 1). All sites, except Wipim, were located in the Momase Region and all Momase sites, except Mumeng, were in areas considered highly malaria endemic (1,9). Sausi is located in the Ramu Valley, home to industrial sugar and oil palm estates, and nearby areas are regularly flooded by the Ramu River and its tributaries. Dreikikir has been a centre for lymphatic filariasis research (10) and malaria is considered highly endemic in the area (11,12). Braun Memorial Hospital in Gagidu village, in the Finschhafen area, is a mission hospital located on the Morobe coastline. Mumeng is situated at an intermediate altitude in the hills of Bulolo District and Wipim in Southern Region is a remote government station in the drainage basin of the mighty Fly River (13).

Data collection

A research nurse was based in each site for the duration of approximately two months coinciding roughly with the main malaria transmission season. All outpatients attending the health facility were screened for a history of fever within the previous three days. A capillary blood sample was collected from self-reported fever cases of all ages and an RDT (ICT Malaria Combo, ICT Diagnostics, South Africa) was performed for on-the-spot diagnosis of malaria. From the same blood sample, thick and thin films were prepared on one slide and haemoglobin (Hb) levels were measured using a HemoCue Hb 201+ Analyser (HemoCue AB, Ängelholm, Sweden). Axillary temperature was measured using an electronic thermometer and spleen palpation was performed on patients aged 2-9 years. Demographic information of the patient was recorded in a one-page form. Results of the above assessment were recorded in the patient's clinic book, after which the patient would undergo routine diagnostic procedures and obtain treatment by the health facility clinicians following their standard practice. The final clinical diagnosis and treatment of

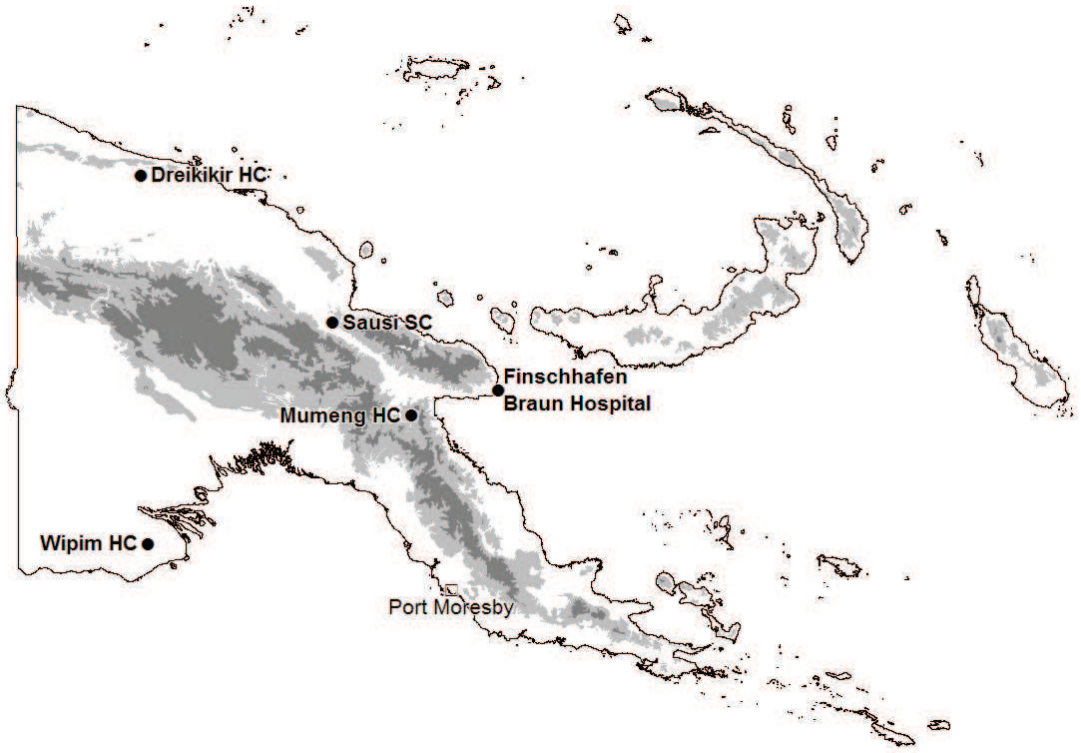

Figure 1. Location of study sites. Grey shaded areas indicate mountain ranges.
HC = health centre, SC = health subcentre

TABLE 1

STUDY SITES

Province	Health facility	Altitude (metres)	Catchment population (villages)*	Surveillance period	Total surveillance days
East Sepik	Health Centre, Dreikikir	420	8300 (41)	30/10/2008-19/12/2008	47
Madang	Health Subcentre, Sausi	160	6700 (21)	20/10/2008-18/12/2008	44
Morobe	Health Centre, Mumeng	940	17000 (10)	28/01/2009-31/03/2009	54
Morobe	Braun Memorial Hospital, Finschhafen	6	14000 (30)	08/06/2009-21/08/2009	50
Western	Health Centre, Wipim	52	9000 (22)	16/04/2009-19/06/2009	37

* Self-reported by health facility

the patient was performed independently by the health facility's clinicians but recorded on the patient's study form. Participation in the study was voluntary and based on oral informed consent. Refusing patients were offered the diagnostic tests without enrolment in the study. Surveillance was repeated after one year in all sites with the exception of Wipim.

Laboratory procedures

Thin smears were first fixed with methanol by the research nurse based in the health facility. Thick and thin smears were stained with Giemsa (4% for 30 minutes) and read by light microscopy independently by two Papua New Guinea Institute of Medical Research (PNGIMR) microscopists. Discordant reads were confirmed by a senior microscopist. A minimum of 200 thick film fields were read before a slide was declared negative. The number of parasites was counted until reaching 200 white blood cells (WBC).

Data analysis

Data were double-entered into a Microsoft FoxPro or Microsoft Access database at PNGIMR Goroka and analysed with Stata (StataCorp LP, College Station, USA) software. Differences in binary and categorical variables between study sites were assessed by Chi-squared test and logistic regression and a non-parametric test for differences in median age. Random effect regression models were applied to allow for clustering at the sentinel site level. The accuracy of comorbidity indicators in predicting slide positivity was based on analysing the area under the receiver operating characteristic (ROC) curve (sensitivity versus 1 − specificity) and comparison of the ROC areas between sites was made using the 'roccomp' command in Stata (14).

Results

Study population

A total of 1807 patients with self-reported history of fever were screened in the five health facilities before the large-scale free distribution of long-lasting insecticide-treated nets (LLINs). The number of fever patients presenting to the facilities differed substantially between the sites, ranging from 4/day in Wipim to 11/day in Sausi. A total of 49% of the patients were female; 11% were under the age of 1 year and 41% below 5 years. The age distribution differed significantly between the study sites as did the self-reported use of mosquito nets on the night before visiting the health facility (Table 2).

Malaria infection

Microscopy results were available for 1800 patients, out of which 45.4% (95% confidence interval [CI] 43.1-47.8) were positive for malaria. Across all sites and age groups, *Plasmodium falciparum* was the dominant species, accounting for 35.2% (95% CI 33.0-37.4) of all infections, followed by 9.3% *P. vivax* (95% CI 8.0-10.8) and 2.1% *P. malariae* (95% CI 1.5-2.8). This included 1.9% (95% CI 1.3-2.6) mixed infections of *P. falciparum* and *P. vivax* and 0.3% (95% CI 0.1-0.6) mixed *P. falciparum* and *P. malariae*. No infection with *P. ovale* was found.

Slide positivity rates varied significantly between the study sites, being lowest in Wipim (2.2%; 95% CI 0.5-6.3), followed by Finschhafen (24.5%; 95% CI 20.0-29.4), Mumeng (43.3%; 95% CI 38.8-47.9), Sausi (51.1%; 95% CI 46.4-55.8) and Dreikikir (74.9%; 95% CI 70.3-79.1) (χ^2 = 306.26, df = 4, p <0.001). The most obvious difference was observed in infections with *P. falciparum* (Figure 2). With the exception of Wipim, where only 3 *P. falciparum*-positive cases were found (in the age group 20+ years), overall and age-specific patterns of species composition were very similar in all sites. Differences between sites were significant for all species independently (*P. falciparum* χ^2 = 264.41, p <0.001; *P. vivax* χ^2 = 32.87, p <0.001; *P. malariae* χ^2 = 17.55, p = 0.002; *P. falciparum* mixed infections χ^2 = 12.83, p = 0.012; all df = 4). When Wipim was excluded, differences between the other sites remained clearly significant for all species (*Plasmodium* spp. χ^2 = 193.78, p <0.001; *P. falciparum* χ^2 = 188.31, p <0.001; *P. vivax* χ^2 = 16.52, p = 0.001; *P. malariae* χ^2 = 13.40, p = 0.004; *P. falciparum* mixed infections χ^2 = 8.95, p = 0.030; all df = 3).

Independent of the study site, slide positivity was strongly correlated with age groups (*P. falciparum* Wald χ^2 = 107.07, p <0.001, study site random effect rho = 0.360; *P. vivax* Wald χ^2 = 60.87, p <0.001, rho = 0.073; *P. malariae* Wald χ^2 = 13.32, p = 0.010, rho = 0.113; *P. falciparum* mixed infections Wald χ^2 = 14.25,

TABLE 2

STUDY POPULATION CHARACTERISTICS BY SENTINEL SITE

	Dreikikir	Finschhafen	Mumeng	Sausi	Wipim	Total
Number of fever patients	390	345	473	463	136	1807
Patients/day	8	7	9	11	4	-
Female (%)	51.7	52.2	48.8	44.9	52.9	49.4
Age group in years (%)*						
<1	2.8	4.9	17.6	14.9	8.1	10.6
1-4	55.1	29.6	24.3	21.6	6.6	29.9
5-9	24.4	18.6	14.6	18.1	14.7	18.4
10-19	5.9	13.0	12.7	12.5	21.3	11.9
20+	10.8	33.9	30.9	28.7	49.3	28.0
missing	1.0	0	0	4.1	0	1.3
Median age (years)**	4	9	7	6	18	6
Mosquito net use last night (%)*	56.1	80.7	35.2	93.1	67.7	65.5
Comorbidity (%)***						
Fever*	74.7	41.5	40.6	26.0	44.9	44.7
Anaemia*	-	22.7	8.0	9.3	0.7	11.3
Splenomegaly*	-	17.2	24.2	50.6	0.0	20.9

* Chi-squared test p <0.001
** Non-parametric K-sample test of equal medians p <0.001
*** Fever = axillary temperature ≥37.5°C; Anaemia = Hb <8 g/dl; Splenomegaly assessed in children aged 2-9 years

p = 0.007, rho = 0.014; all df = 4). Infections with *P. falciparum* peaked in the age group 5-9 years and *P. vivax* in the age group 1-4 years. In Sausi, where *P. malariae* was most common (4.3% of fever patients), a peak was observed in the age group 5-9 years. Mixed infections of *P. falciparum* and any other species (mostly *P. vivax)* tended to peak in the age group 1-4 years (Figure 2).

Predictors of malaria infection

Of all 1807 self-reported febrile illness cases included in this study, 44.7% had acute fever (axillary temperature ≥37.5°C) at the time of presenting to the health facility (Table 2). Acute fever was age dependent (χ^2 = 111.07, p <0.001) and overall most common in the age groups 1-4 (59.4%) and 5-9 (50.8%) years. Haemoglobin levels were measured in all sites except Dreikikir. A total of 11.3% of 1368 patients had moderate to severe anaemia (Hb <8 g/dl). Most cases of anaemia were found in Finschhafen. Anaemia was most prevalent in patients aged 1-4 years (22.4%) and least in patients 10-19 (5.8%) and 20 years and

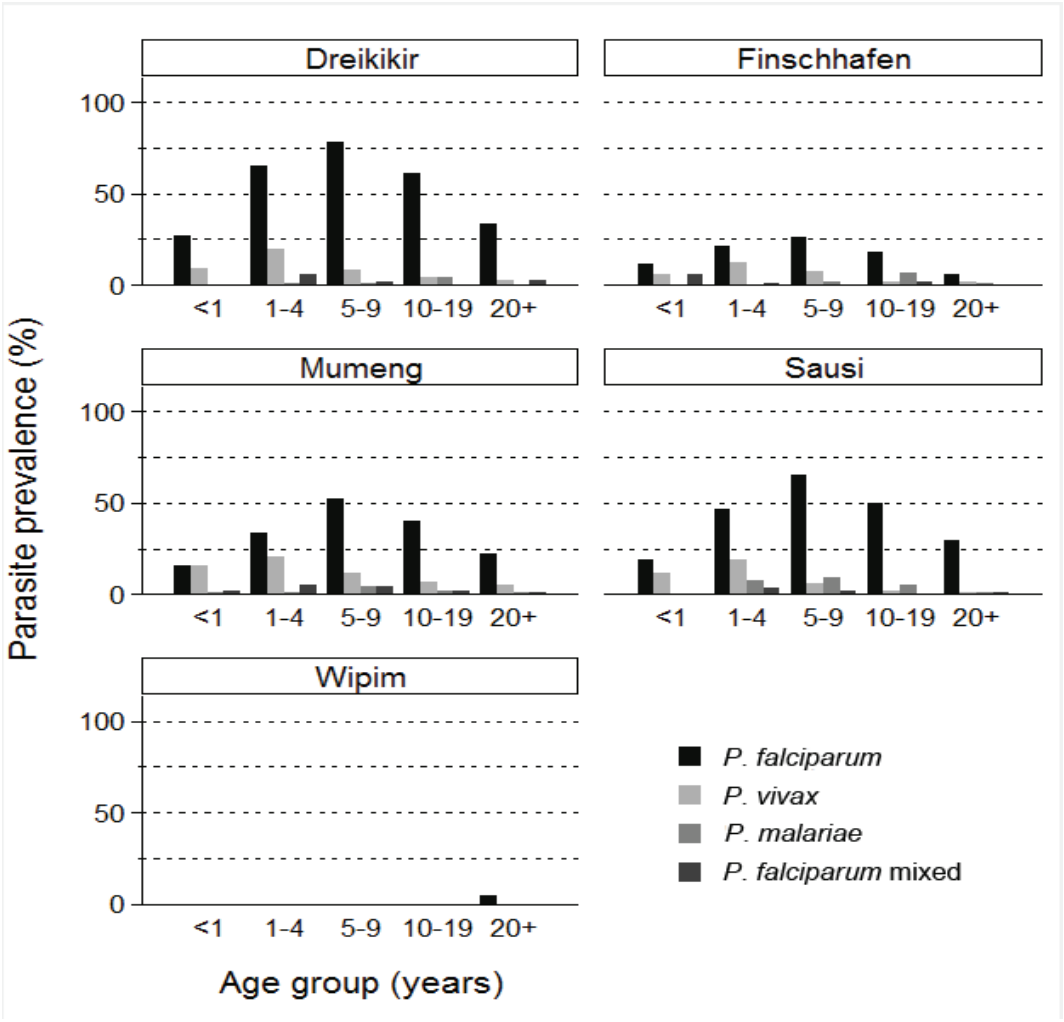

Figure 2. Slide positivity of fever patients by age group and study site.

older (5.7%) (χ^2 = 58.75, df = 4, p <0.001). Spleens were not palpated in Dreikikir. Out of 494 patients 2-9 years of age screened in the other sites, 20.9% were found with signs of splenomegaly (Hackett score 1-5). Detected splenomegaly increased slightly with each year of age (OR = 1.17, 95% CI 1.05-1.29, p = 0.003, with study site as random effect). Splenomegaly was most common in Sausi (50.6%).

None of the above clinical signs were accurate predictors of malaria infection as determined by microscopy. The sensitivity of acute fever in predicting slide positivity was 54.3% (95% CI 50.8-57.7), ranging from 79.5% in Dreikikir to 32.9% in Sausi. Anaemia (Hb <8 g/dl) showed a sensitivity of 16.7% (95% CI 13.5-20.2), ranging from 37.8% in Finschhafen to 0% in Wipim, and that of splenomegaly in children 45.4% (95% CI 39.4-51.4), ranging from 58.7% in Sausi to 29.4% in Finschhafen. The combination of fever and anaemia or fever and splenomegaly did not improve the accuracy of the clinical predictors (data not shown). While the accuracy of clinical predictors (assessed by comparing the areas under the ROC curve) varied to different degrees between study sites (Table 3), none of the clinical signs was as accurate as an RDT. RDTs had a 90.0% (95% CI 87.7-92.0) sensitivity of detecting a malaria infection and a negative predictive value (proportion of negative RDTs that were slide negative) of 91.1% (95% CI 89.0-92.9) for any *Plasmodium* species and 96.1% (95%

TABLE 3
ACCURACY OF PREDICTORS OF MALARIA INFECTION

	Fever % (95% CI)	Anaemia* % (95% CI)	Splenomegaly*/** % (95% CI)	RDT % (95% CI)
***Plasmodium* spp.**				
Sensitivity	54.3 (50.8-57.7)	16.7 (13.5-20.2)	45.4 (39.4-51.4)	90.0 (87.7-92.0)
Specificity	63.5 (60.4-66.6)	91.8 (89.8-93.6)	90.0 (85.2-93.7)	84.4 (82.0-86.6)
ROC	0.59 (0.57-0.61)	0.54 (0.52-0.56)	0.68 (0.64-0.71)	0.87 (0.86-0.89)
PPV	55.4 (51.8-58.8)	54.5 (46.3-62.5)	85.8 (79.1-91.0)	82.7 (80.0-85.1)
NPV	62.5 (59.4-65.5)	65.3 (62.5-67.9)	55.4 (50.0-60.7)	91.1 (89.0-92.9)
Equality of ROC areas between sites	χ^2 = 5.82, df = 4, p = 0.213	χ^2 = 28.13, df = 3, p <0.001	χ^2 = 1.53, df = 2, p = 0.466	χ^2 = 7.09, df = 4, p = 0.131
P. falciparum				
Sensitivity	56.6 (52.6-60.5)	17.0 (13.4-21.2)	53.3 (46.1-60.4)	94.4 (92.2-96.0)
Specificity	62.0 (59.1-64.7)	90.8 (88.9-92.5)	85.6 (81.1-89.4)	75.0 (72.4-77.5)
ROC	0.59 (0.57-0.62)	0.54 (0.52-0.56)	0.69 (0.65-0.74)	0.85 (0.83-0.86)
PPV	44.6 (41.2-48.2)	41.0 (33.2-49.2)	71.6 (63.6-78.7)	67.1 (63.9-70.2)
NPV	72.4 (69.6-75.2)	74.5 (72.0-76.9)	72.9 (67.9-77.5)	96.1 (94.6-97.3)
Equality of ROC areas between sites	χ^2 = 7.69, df = 4, p = 0.104	χ^2 = 19.78, df = 3, p <0.001	χ^2 = 1.78, df = 2, p = 0.410	χ^2 = 19.01, df = 4, p <0.001
P. vivax**				
Sensitivity	55.4 (47.5-63.0)	16.4 (10.0-24.6)	21.5 (12.3-33.5)	81.3 (74.6-86.9)
Specificity	56.7 (54.1-59.2)	87.9 (85.9-89.8)	66.3 (61.5-71.0)	50.1 (47.5-52.7)
ROC	0.56 (0.52-0.60)	0.52 (0.49-0.56)	0.44 (0.38-0.50)	0.66 (0.63-0.69)
PPV	12.6 (10.3-15.2)	11.6 (7.0-17.7)	9.5 (5.3-15.4)	15.6 (13.2-18.2)
NPV	91.9 (89.9-93.6)	91.6 (89.7-93.1)	83.8 (79.3-87.7)	96.0 (94.3-97.2)
Equality of ROC areas between sites	χ^2 = 11.26, df = 3, p = 0.010	χ^2 = 1.51, df = 2, p = 0.470	χ^2 = 1.94, df = 2, p = 0.380	χ^2 = 62.00, df = 3, p <0.001

CI = confidence interval
RDT = rapid diagnostic test
ROC = receiver operating characteristic (sensitivity versus 1− specificity)
PPV = positive predictive value
NPV = negative predictive value
*Excluding Dreikikir
**Excluding Wipim

CI 94.6-97.3) for *P. falciparum* (Table 3). The accuracy of RDTs in detecting malaria infection did not differ between study sites ($\chi^2 = 7.09$, df = 4, p = 0.131); however, lower accuracy levels were observed for species-specific detection rates in Sausi (*P. falciparum* 65.7%, 95% CI 47.8-80.9) and Wipim (*P. vivax* 66.7%, 95% CI 9.4-99.2), resulting in significantly different species-specific accuracy between sites (Table 3). The combination of individual clinical predictors with the RDT result did not increase accuracy levels.

Diagnosis and treatment

The final clinical diagnosis made by staff of the health facilities was malaria in 66.6% of the 1807 cases including 89.2% of slide-positive (95.7% RDT-positive) and 47.7% of slide-negative (38.1% RDT-negative) patients. Another 9.4% of patients were diagnosed with pneumonia, 5.8% with flu or common cold, 0.2% with typhoid fever and 7.8% with any other infection (multiple diagnoses were possible). The remaining patients had other or no diagnoses recorded. After malaria, the most common diagnosis in point-of-care RDT-negative patients was pneumonia (16.2% overall and 29.2% in children under 5 years of age). For 28.7% of all patients diagnosed with malaria by the health facility clinician, a negative RDT test result was available at the time of diagnosis and 39.1% were later confirmed malaria negative by light microscopy.

A total of 84.5% of all 1807 fever patients, including 74.2% of slide-negative and 69.7% of point-of-care RDT-negative cases, were treated with an antimalarial drug. The most commonly administered drug was sulphadoxine-pyrimethamine (SP), in 66.8% of all antimalarial treatments, which was usually administered in combination with chloroquine (23.4%), amodiaquine (16.5%), quinine (10.7%) or an artemisinin monotherapy, usually artemether (16.2%). In total, 76.8% (1173/1527) of the antimalarial treatments consisted of two or three antimalarials. Only 23.2% of all antimalarial treatments were monotherapies, most commonly chloroquine (9.0% of all antimalarial treatments), amodiaquine (7.2%) or an artemisinin derivative (4.2%). Primaquine, the only drug against *P. vivax* hypnozoites, was provided to 322 patients (21.1%), including 25% of patients with a *P. vivax*-positive and 17.0% with a *P. vivax*-negative blood slide. A total of 40.7% of patients with a point-of-care RDT test result of 'non-*P. falciparum* malaria' received primaquine, as well as 26.6% of those with '*P. falciparum* or mixed malaria'. Dosages of prescribed drugs were not recorded in the frame of this study and it is thus not possible to ascertain how many patients received primaquine as a 14-day anti-hypnozoite treatment and how many as a single-dose, anti-gametocyte treatment.

Comparison with routine statistics

For the three years before we conducted our sentinel site surveillance, the National Health Information System (NHIS) reported a monthly average of 579 outpatient malaria cases for Dreikikir (period November-December every year), 203 for Finschhafen (June-August), 224 for Mumeng (February-March), 223 for Sausi (October-December) and 206 for Wipim (April-June). None of the sentinel site facilities had implemented routine use of parasitological diagnosis (with the possible exception of Finschhafen, where on occasions slides were collected for microscopy) and the above numbers are therefore largely presumptive diagnoses. Based on the slide positivity rates of those sentinel site surveillance cases with the final diagnosis 'malaria', the proportion and absolute monthly number of 'real' (ie, slide-confirmed) malaria cases can be estimated (Table 4). This shows that in the sentinel site health facilities, an average excess number of 63 (Sausi) to 201 (Wipim) malaria patients was reported every month for the time of the year in which surveillance was conducted – or an excess of 18% (Dreikikir) to 98% (Wipim).

Discussion

This study highlights a number of issues that are important for understanding the clinical epidemiology of malaria in PNG and for addressing the burden of malaria through targeted control activities, particularly in a health centre setting. Firstly, the data confirm that a large number (in some settings even the vast majority) of fever patients presenting to health facilities are not infected with malaria parasites. As in an earlier study based on >10 years of morbidity surveillance in the Wosera area, East Sepik Province (15), children aged between 5 and 9 years had the highest parasite rates. Parasitaemia was less common in febrile children under 1 year of age and in adults over 20 years. Considering

TABLE 4

Estimated difference between NHIS reported and 'real' (slide-positive) malaria cases

	Dreikikir	Finschhafen	Mumeng	Sausi	Wipim
Reference months	November - December	June - August	February - March	October - December	April - June
Monthly NHIS reported malaria cases*	579	203	224	223	206
Estimated slide positivity among 'malaria' cases**	81.5%	31.8%	69.5%	71.8%	2.3%
Estimated monthly number of confirmed malaria cases	472	65	156	160	5
Average monthly overestimation of malaria cases for reference months	107	138	68	63	201

NHIS = National Health Information System

*Monthly average in the three years before surveillance based on time periods indicated in the text

**Based on proportion with a positive slide out of all cases with the final health facility diagnosis 'malaria'

that not all malaria infections cause clinical disease (16), the real malaria-attributable fraction among the fever patients would be even lower than the proportion of cases with parasites. According to a study conducted in the vicinity of the Dreikikir study site, the clinical tolerance is similar for all malaria species, which would translate into a lower malaria-attributable fraction among infections with all species (16).

Interestingly, the Wipim study site in Western Province showed an almost complete absence of malarial fevers, which was supported by findings from entomological surveys in the health centre's catchment area (Lisa Reimer, PNGIMR, Madang, personal communication). The absence of malaria cases could be attributed to seasonal fluctuations due to heavy rains during the surveillance period (which may have washed away mosquito larvae and reduced transmission) as well as the low population density in the area. However, the population prevalence rates in three nearby villages were between 0 and 1.5%, suggesting that the situation observed at Wipim Health Centre may indeed reflect very low endemicity (6). The data thus emphasize that differences in malaria-attributable fevers can be striking, even within lowland areas considered malaria endemic. It should be noted that all sites, except Mumeng, were located in areas previously considered hyperendemic (17).

While the new national malaria treatment protocol, now the National Malaria Treatment Policy (18), emphasized the need for parasitological confirmation before the administration of antimalarials, the majority of fever cases in rural health facilities are still treated presumptively and mostly with antimalarial drugs. A study conducted in 2010 across PNG found that only 15% of 79 randomly sampled health facilities had operational parasitological diagnosis (RDT or microscopy) available (19). Provider observations conducted during the same study in 54 of these facilities found that only 15% of observed fever cases were diagnosed with an RDT and only 3.6% by light microscopy. Even in those facilities with RDT or microscopy available, these tools were used for less than 50% of the fever patients (7). The same study also found that an antimalarial was prescribed to 96.4% of fever cases, including 82% of patients who were tested negative (7). Overdiagnosis of malaria and overtreatment of malaria-negative patients with antimalarial drugs have been reported from many places (20-22) and a similar tendency was found in this study, where all fever cases were diagnosed by RDT by a study nurse but the majority of RDT-negative patients were subsequently diagnosed with malaria and prescribed an antimalarial drug by the health facility's clinical staff in spite of the available RDT result. Many clinicians continue to rely on clinical signs and symptoms for the diagnosis of malaria. However, the results from this study underline that neither a history of fever (inclusion criterion for this study) nor acute fever, nor any of the other assessed clinical signs, are reliable predictors of a malaria infection. A thorough clinical assessment is a central component of providing adequate clinical care to a patient and there are indisputable benefits of measuring body temperature, Hb levels and palpating a patient's abdomen. However, some of these assessments cannot be or are for other reasons not routinely performed in PNG health facilities (7). The results from this study therefore clearly support the calls for scaling-up the diagnostic capacity of health facilities across PNG and basing all malaria treatments on parasitological rather than clinical diagnosis, particularly in a health centre/aid post setting where the diagnostic capacities are limited. RDT results proved to be by far the most sensitive diagnostic tool with high sensitivity and only a small proportion of missed infections. The latter is reflected in the high negative predictive values, which represent the proportion of negative RDTs that are true non-malarial fevers. This should give confidence to health workers to trust in both positive as well as negative RDT results and is in line with findings from a study conducted elsewhere in PNG (23).

While positive malaria test results are simple to interpret, negative tests may leave clinicians in the dilemma as to how non-malarial fevers should – and can, in the local context – be treated. Studies in tropical countries found dengue, influenza and rickettsials to be major aetiological agents of non-malarial febrile illness; yet the disease agent remained unidentified in 20-50% of cases (24-26). In neighbouring West Papua, Indonesia, an inpatient fever study identified typhoid fever, pneumonia, leptospirosis, urinary tract infections, rickettsioses, dengue and meningitis/encephalitis as the most common confirmed or suspected diagnoses; yet 64% of the fevers remained undiagnosed

(27). Very few studies have investigated the aetiology of non-malarial fevers in PNG. Urinary tract infections were diagnosed in 9% of children with unspecific febrile illness in Port Moresby (28) and dengue was diagnosed based on serological tests in 8% of outpatient fever cases (14% of RDT-negative fevers) in Madang Province (29). Stored dried blood spot samples from the fever patients in Wipim were analysed by polymerase chain reaction (PCR) and 15 cases of dengue type 1 (11%) were found among the 136 fever patients sampled (30).

Despite these doubts, withholding antimalarial medicines from children with a negative malaria RDT was proven to be safe in a prospective study conducted in children aged 3-27 months in high-endemicity settings in East Sepik and Madang Provinces. Of all 3942 RDT-negative children who were not treated with an antimalarial, less than one percent was found to be parasitaemic in the following seven days, the majority of whom were then treated as outpatients. The 26 RDT-negative children who returned with a severe illness had other underlying conditions, mostly lower respiratory tract infections (23).

This study presents the situation before the rollout of large-scale malaria control interventions including insecticide-treated mosquito nets (ITNs) and artemisinin-based combination therapy (ACT). It is expected that the interventions, which significantly increased ITN coverage across PNG (8), will result in reductions in health facility malaria cases. Both the observed differences in parasitaemia rates between sites and the expected changes due to intensified control warrant the consistent application of parasitological diagnosis of malaria across PNG. The Global Fund Round 8 malaria grant supports the rollout of RDTs and improvement in microscopy services alongside training of health workers in the new treatment protocol (31). The scaling-up of malaria diagnostic tools (and its proper application) is one of several urgently required measures which can improve case management and save unnecessary treatments (32). However, a major shift in the current malaria case management practice is required to conform to the new guidelines (7) and an improved understanding of the aetiology and treatment of non-malarial fevers is necessary to adequately manage those patients with a negative malaria test. The strong evidence for the essential first step, confirming the reliability of RDTs in PNG settings, particularly in case of negative results, should be encouraging to clinicians (23).

The application of findings from this study to estimate over-reporting of health facility malaria cases by the NHIS highlights another problem with presumptive diagnosis (and reporting). Health information systems should provide evidence of the disease burden for health planning, including quantification of medical supplies such as diagnostic tests and medicines. In order to be optimally useful for planning, such surveillance data need to be accurate, timely and actionable (33). A consistent application of parasitological diagnosis of malaria and ideally the application of point-of-care tests for other febrile illnesses and subsequent test-based reporting could therefore significantly improve reporting through the NHIS, which in turn would allow for better planning and targeting of malaria control interventions. A positive secondary effect would be an expected reduction in health expenditures at provider and patient levels. Improved surveillance of malaria is therefore a key component of intensifying malaria control efforts and a prerequisite for a potential future malaria elimination campaign (34,35).

While the above results highlight principal areas of relevance for the national malaria control program, the relatively brief two-month period of surveillance (which was determined by financial and time factors) is a primary limitation of this study. Fortunately, in several settings additional data sources were available to support the key findings. The presence of a PNGIMR nursing officer in the sentinel sites may have influenced patient attendance and health facility staff behaviour (Hawthorne effect). Yet changes in staff behaviour related to malaria diagnosis and treatment could only have been minimal since no significant differences were seen from the findings in a country-wide survey on malaria case management (7). The estimates of excess malaria reporting were based on the assumptions that the case load and age structure of patients in the three years before the surveillance were similar to those in the surveillance period, and that NHIS reports were mainly based on presumptive diagnosis. The latter may not have been the case in all instances (particularly in Finschhafen) and hence over-reporting would be lower

than indicated. However, this might again be outweighed by the added value of RDT results in sentinel site facilities. Lastly, the selection of study sites was a result of the ITN distribution schedule and the sites do not necessarily allow for an extrapolation of the results to the rest of PNG, particularly as all sites, except Wipim, were located in the Momase Region. While these shortcomings need to be acknowledged, the key results and conclusions of this study remain unchanged.

Conclusions

Significant differences in malaria-attributable fevers between sites located in malaria-endemic regions of PNG highlight the need to change from previously universal presumptive treatment of malaria to parasitological diagnosis. Local and international evidence supports a test-and-treat policy based on RDTs or microscopy. The differences also highlight that malaria control may need to be tailored better to different settings within PNG. Non-malarial fevers may present a challenge to health workers in remote health facilities and further research and training are required to address the management of other causes of fever.

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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 **Betuela I, Maraga S, Hetzel MW, Tandrapah A, Sie A, Yala S, Kundi J, Siba P, Reeder JC, Mueller I.** Epidemiology of malaria in the Papua New Guinean highlands. *Trop Med Int Health* 2012;17:1181-1191.

3 **Cooper RD, Waterson DGE, Frances SP, Beebe NW, Pluess B, Sweeney AW.** Malaria vectors of Papua New Guinea. *Int J Parasitol* 2009;39:1495-1501.

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 **Mueller I, Tulloch J, Marfurt J, Hide R, Reeder JC.** Malaria control in Papua New Guinea results in complex epidemiological changes. *PNG Med J* 2005;48:151-157.

6 **Hetzel MW, Gideon G, Mueller I, Siba PM.** Papua New Guinea/The Global Fund Round 3 Malaria Control Programme Evaluation 2008/2009: Results from Cross-Sectional Surveys and Sentinel Sites. Goroka: Papua New Guinea Institute of Medical Research, 2010.

7 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

8 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.

9 **Kazura JW, Siba PM, Betuela I, Mueller I.** Research challenges and gaps in malaria knowledge in Papua New Guinea. *Acta Trop* 2012;121:274-280.

10 **Bockarie M, Kazura J, Alexander N, Dagoro H, Bockarie F, Perry R, Alpers M.** Transmission dynamics of *Wuchereria bancrofti* in East Sepik Province, Papua New Guinea. *Am J Trop Med Hyg* 1996;54:577-581.

11 **Bockarie MJ, Dagoro H.** Are insecticide-treated bednets more protective against *Plasmodium falciparum* than *Plasmodium vivax*-infected mosquitoes? *Malar J* 2006;5:15.

12 **Bockarie MJ, Alexander N, Bockarie F, Ibam E, Barnish G, Alpers M.** The late biting habit of parous *Anopheles* mosquitoes and pre-bedtime exposure of humans to infective female mosquitoes. *Trans R Soc Trop Med Hyg* 1996;90:23-25.

13 **Reagan DP, Rau MT, Barrie B.** Fauna and food webs of the Fly River Basin. In: Bolton BR, ed. The Fly River, Papua New Guinea: Environmental Studies in an Impacted Tropical River System. Oxford: Elsevier, 2008:549-574.

14 **Sullivan Pepe M.** The Statistical Evaluation of Medical Tests for Classification and Prediction. Oxford: Oxford University Press, 2004.

15 **Genton B, D'Acremont V, Rare L, Baea K, Reeder JC, Alpers MP, Müller I.** *Plasmodium vivax* and mixed infections are associated with severe malaria in children: a prospective cohort study from Papua New Guinea. *PLoS Med* 2008;5:e127.

16 **Müller I, Genton B, Rare L, Kiniboro B, Kastens W, Zimmerman P, Kazura J, Alpers M, Smith TA.** Three different *Plasmodium* species show similar patterns of clinical tolerance of malaria infection. *Malar J* 2009;8:158.

17 **Parkinson AD.** Malaria in Papua New Guinea 1973. *PNG Med J* 1974;17:8-16.

18 **Papua New Guinea Department of Health.** National Malaria Treatment Policy. Port Moresby: Department

of Health, Sep 2009.
19 **Kurumop SF, Pulford J, Mueller I, Siba PM, Hetzel MW.** Diagnostic capacity and antimalarial availability in Papua New Guinea before the introduction of a revised national malaria treatment protocol. *PNG Med J* 2014;57:59-67.
20 **Joshi R, Colford JM Jr, Reingold AL, Kalantri S.** Nonmalarial acute undifferentiated fever in a rural hospital in central India: diagnostic uncertainty and overtreatment with antimalarial agents. *Am J Trop Med Hyg* 2008;78:393-399.
21 **Reyburn H, Mbakilwa H, Mwangi R, Mwerinde O, Olomi R, Drakeley C, Whitty CJ.** Rapid diagnostic tests compared with malaria microscopy for guiding outpatient treatment of febrile illness in Tanzania: randomised trial. *Br Med J* 2007;334:403.
22 **Reyburn H, Mbatia R, Drakeley C, Carneiro I, Mwakasungula E, Mwerinde O, Saganda K, Shao J, Kitua A, Olomi R, Greenwood BM, Whitty CJ.** Overdiagnosis of malaria in patients with severe febrile illness in Tanzania: a prospective study. *Br Med J* 2004;329:1212.
23 **Senn N, Rarau P, Manong D, Salib M, Siba P, Robinson LJ, Reeder J, Rogerson S, Mueller I, Genton B.** Rapid diagnostic test-based management of malaria: an effectiveness study in Papua New Guinean infants with *Plasmodium falciparum* and *Plasmodium vivax* malaria. *Clin Infect Dis* 2012;54:644-651.
24 **Leggat PA.** Assessment of febrile illness in the returned traveller. *Aust Fam Physician* 2007;36:328-332.
25 **Wilson ME, Freedman DO.** Etiology of travel-related fever. *Curr Opin Infect Dis* 2007;20:449-453.
26 **de Lavaissiere M, D'Ortenzio E, Dussart P, Fontanella JM, Djossou F, Carme B, Marchou B.** Febrile illness at the emergency department of Cayenne Hospital, French Guiana. *Trans R Soc Trop Med Hyg* 2008;102:1055-1057.
27 **Punjabi NH, Taylor WR, Murphy GS, Purwaningsih S, Picarima H, Sisson J, Olson JG, Baso S, Wangsasaputra F, Lesmana M, Oyofo BA, Simanjuntak CH, Subekti D, Corwin AL, Richie TL.** Etiology of acute, non-malaria, febrile illnesses in Jayapura, northeastern Papua, Indonesia. *Am J Trop Med Hyg* 2012;86:46-51.
28 **Morris CM, Tefuarani N, Ripa P, Laki R, Vince JD.** Urinary tract infection in infants and young children presenting with fever without a focus in Port Moresby. *PNG Med J* 2007;50:145-151.
29 **Senn N, Luang-Suarkia D, Manong D, Siba PM, McBride WJH.** Contribution of dengue fever to the burden of acute febrile illnesses in Papua New Guinea: an age-specific prospective study. *Am J Trop Med Hyg* 2011;85:132-137.
30 **Bande G, Hetzel MW, Iga J, Barnadas C, Mueller I, Siba PM, Horwood PF.** An investigation into febrile illnesses of unknown aetiology in Wipim, Papua New Guinea. *PNG Med J* 2014;57:52-58.
31 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Malaria Control in Papua New Guinea: Scaling up for Impact. Principal Recipient Department of Health of Papua New Guinea. Geneva: Global Fund. http://portfolio.theglobalfund.org/en/Grant/Index/PNG-809-G04-M
32 **Pang T, Peeling RW.** Diagnostic tests for infectious diseases in the developing world: two sides of the coin. *Trans R Soc Trop Med Hyg* 2007;101:856-857.
33 **Mueller I, Slutsker L, Tanner M.** Estimating the burden of malaria: the need for improved surveillance. *PLoS Med* 2011;8:e1001144.
34 **malERA Consultative Group on Diagnoses and Diagnostics.** A research agenda for malaria eradication: diagnoses and diagnostics. *PLoS Med* 2011;8:e1000396.
35 **malERA Consultative Group on Integration Strategies.** A research agenda for malaria eradication: cross-cutting issues for eradication. *PLoS Med* 2011;8:e1000404.

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An investigation into febrile illnesses of unknown aetiology in Wipim, Papua New Guinea

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SUMMARY

In Papua New Guinea the aetiology of febrile illnesses remains poorly characterized, mostly due to poor diagnostic facilities and the inaccessibility of much of the rural areas of the country. We investigated the aetiological agents of febrile illnesses for 136 people presenting to Wipim Health Centre in Western Province, Papua New Guinea. Arboviral and rickettsial real-time polymerase chain reaction (PCR) assays, malaria blood smears and a malaria PCR test were used to identify pathogens associated with a history of fever. In 13% (n = 18) of cases an aetiological agent was identified. Dengue virus type 1 was detected in 11% (n = 15) of the samples tested and malaria in 2% (n = 3). None of the other arboviral or rickettsial pathogens tested for were detected in any of the samples. Although dengue viruses have been identified in Papua New Guinea using serological methods, this study represents the first direct detection of dengue in the country. The detection of malaria, on the other hand, was surprisingly low considering the previous notion that this was a hyperendemic region of Papua New Guinea.

Introduction

Febrile illnesses are one of the most common causes of morbidity and mortality in developing countries. They can be caused by many different aetiological agents and often present with an unspecific symptomatology. In resource-poor settings, the clinical management of such illnesses is challenging due to limitations in diagnostic facilities (1) which in turn also result in poorly characterized causes of disease (2). In Papua New Guinea malaria has long been regarded as one of the most important causes of morbidity and mortality, particularly in lowland regions (3). However, endemicity levels have been shown to vary greatly between sites even in areas previously considered hyperendemic (4,5). In addition, the large-scale free distribution of insecticide-treated bednets has the potential to impact significantly on the burden of malaria across the country (6). Nevertheless, malaria remains the most common clinical diagnosis and antimalarials the most popular treatment of fever cases at most health centres (7). Alternative aetiologies are rarely considered unless treatment failure with antimalarial medication occurs. Although only limited studies have been conducted to investigate the aetiology of non-malarial febrile illnesses, a large range of pathogens have been implicated as causes of disease,

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including typhoid (8), dengue (9) and various other arboviruses (10). The current research investigated the causes of febrile illnesses with a focus on arthropod-borne pathogens in patients presenting to Wipim Health Centre in Western Province, Papua New Guinea. The study was approved by the Papua New Guinea Institute of Medical Research Institutional Review Board (IMR IRB No 1027) and the Papua New Guinea Medical Research Advisory Committee (MRAC No 11.14).

Materials and Methods

Wipim Health Centre is located in the South Fly District of Western Province, Papua New Guinea (PNG) and serves approximately 9000 people in 22 villages (5). Active malaria surveillance was conducted as part of an evaluation of the National Malaria Control Program by the Papua New Guinea Institute of Medical Research (PNGIMR) covering a period of 37 working days between April and June 2009. During this period, 136 patients presented to the clinic with a self-reported history of fever within the previous three days. A malaria blood slide was completed and a dried blood spot sample on filter paper was sent to the PNGIMR laboratory in Goroka, Eastern Highlands Province, for further analysis. Blood samples were collected from eligible patients following oral informed consent.

Malaria blood slides were stained with Giemsa and read by light microscopy independently by two microscopists; discordant reads were confirmed by a third senior microscopist. A minimum of 200 thick film fields were read before a slide was declared negative.

Genomic nucleic acids were extracted from dried blood spot samples using the Favorgen 96-Well Genomic DNA (deoxyribonucleic acid) Kit (Biotech. Corp., Taiwan) according to the manufacturer's instructions. The presence of a malaria infection was confirmed using a semi-quantitative post-PCR (polymerase chain reaction) ligase detection reaction-fluorescent microsphere assay (11). This assay combines PCR amplification of a fragment of the small subunit ribosomal RNA (ribonucleic acid) gene using *Plasmodium* genus-specific primers, followed by a multiplex species-specific ligation detection reaction. The design and sensitivity of this assay have been described previously (11,12). The positivity of the reactions for each species was determined using a polar transformation of the data (13,14).

Previously published real-time PCR primers and probes were used to test for a large range of arboviral and rickettsial species (Table 1). The individual arboviral real-time reverse transcription (RT)-PCR assays were conducted in 20 µl reaction mixes containing 2 µl of the nucleic acid sample, 200 nM of each primer, 200 nM of each probe, 1× QuantiTect Multiplex RT-PCR Master Mix, 1× QuantiTect RT (Qiagen) and sterile deionized water. Viral positive controls and negative controls of nuclease-free water were included with each assay to ensure consistency of the assays. All reactions were conducted with a Bio-Rad CFX-96 Real-Time System with the following cycling parameters: 50°C for 20 minutes and 95°C for 15 minutes, followed by 40 cycles of 94°C for 45 seconds and 60°C for 75 seconds.

The samples were also screened for all known rickettsial species using universal real-time PCR primers that were designed to detect the three rickettsial groups (scrub typhus group, typhus group and spotted fever group). The individual real-time PCR assays were conducted in 20 µl reaction mixes containing 2 µl of the nucleic acid sample, 200 nM of each primer, 1× Platinum SYBR Green qPCR Supermix (Invitrogen) and sterile deionized water. Rickettsial positive controls and negative controls of nuclease-free water were included with each assay to ensure consistency of the assays. All reactions were conducted with a Bio-Rad CFX-96 Real-Time System with the following cycling parameters: 95°C for 2 minutes, followed by 40 cycles of 95°C for 15 seconds, 54°C for 15 seconds and 60°C for 30 seconds. Melt curve analysis was also performed with increments of 1°C/30 seconds at 70-95°C to determine the peak fluorescence change over time (df/dT).

Results

All of the clinical samples were tested for regionally important arboviruses and rickettsials (Table 1) to detect the aetiological agents of fever in the area. Dengue virus 1 (DENV-1) was detected in 11.0% (n = 15) of the samples. Malaria was detected in 3 patients by both light microscopy and PCR. No other dengue type and none of the other pathogens of interest were detected in any of the samples.

TABLE 1

Primer and probe sequences used in this study for the detection of arboviruses and rickettsials

Virus/Bacteria	Primer/Probe	Sequence	Reference
Flaviviruses			
DENV	NS5F	GGAAGGAGAAGGACTGCACA	15
	NS5R	ATTCTTGTGTCCCATCCTGCT	
DENV-1	DSQ1	[FAM]CTCAGAGACATATCAAAGATTCCCGGG[BHQ1]	
DENV-2	DSQ2	[TexasRed]TAAGAGACGTGAGCAAGAAAGAGGGAGGAG[BHQ2]	
DENV-3	DSQ3	[Cy5]ACATTTCCAAGATACCCGGAGGAG[BHQ3]	
DENV-4	DSQ4	[HEX]CCTAGAGGACATAGACAAAAAGGAAGGAGACC[BHQ1]	
JEV	JEVF	ATCTGGTGYGGYAGTCTCA	16
	JEVR	CGCGTAGATGTTCTCAGCCC	
	JEVP	[FAM]CGGAACGCGATCCAGGGCAA[BHQ1]	
KUNV	KUNF	AACCCCAGTGGAGAAGTGGA	16
	KUNR	TCAGGCTGCCACACCAAA	
	KUNP	[Texas Red]CGATGTTCCATACTCTGGCAAACG[BHQ2]	
MVEV	MVEF	ATCTGGTGYGGAAGYCTCA	16
	MVER	CGCGTAGATGTTCTCAGCCC	
	MVEP	[Cy5]ATGTTGCCCTGGTCCTGGTCCCT[BHQ3]	
Togaviruses			
CHIKV	CHIKF	TCGACGCGCCCTCTTTAA	17
	CHIKR	ATCGAATGCACCGCACACT	
	CHIKP	[FAM]ACCAGCCTGCACCCATTCCTCAGAC[BHQ1]	

RRV	RRVF	ACGGAAGAAGGGATTGAGTACCA	18
	RRVR	TCGTCAGTTGCGCCCATA	
	RRVP	[Texas Red]CAACAACCCGCCGGTCCGC[BHQ2]	
BFV	BFF3	TGGATAACACAGTGTGGCAGT	This study
	BFR3	GGCACATGGATCTTTCCTTT	
	BFP3	[Cy5]CCAATACGTGCCCAGGTCCGA[BHQ3]	
Rickettsials			
TG/SFG	gltA-F	TCGCAAATGTTCACGGTACTTT	19
	gltA-R	TCGTGCATTTCTTTCCATTGTG	
SFG	OmpB-F	CGACGTTAACGGTTTCTCATTCT	
	OmpB-R	ACCGGTTTCTTTGTAGTTTTCGTC	
STG	47kDa-F	AACTGATTTTATTCAAACTAATGCTGCT	
	47kDa-R	TATGCCTGAGTAAGATACRTGAATRGAATT	

DENV = dengue virus
JEV = Japanese encephalitis virus
KUNV= kunjin virus
MVEV = Murray Valley encephalitis virus
CHIKV = chikungunya virus
RRV = Ross River virus
BFV = Barmah Forest virus
TG = typhus group
SFG = spotted fever group
STG = scrub typhus group

There was no significant difference between the mean age of dengue-infected patients (12.5 years) and non-infected patients (19.0 years). Likewise, there was no statistical significance in the sex distribution of dengue-infected patients. However, we found that a significant proportion (86.7%; Fisher's Exact test, p = 0.011) of dengue patients were residents in Wipim, whereas Wipim residents constituted only 54.4% of all screened patients (Table 2).

Discussion

In the present study DENV-1 was the most important pathogen detected in febrile patients presenting to the Wipim Health Centre in the South Fly District of Western Province. Although it is generally accepted that dengue is endemic and an important cause of febrile illnesses throughout the lowlands of PNG, very few studies have been conducted to investigate the prevalence and distribution of this important pathogen. Dengue outbreaks have been reported in PNG in the past (20,21) and a recent study at Yagaum rural hospital and Jumba town clinic, Madang Province detected DENV in 8% of febrile cases enrolled into the study (9). Dengue fever has been widely reported in other neighbouring countries such as Fiji, French Polynesia and New Caledonia, where it has become a major concern among health authorities (21). In these Pacific island nations dengue fever normally occurs in an epidemic pattern of disease. In this study, the geographical distribution of cases suggests that during the specimen collection period there was an outbreak of DENV-1 in the village of Wipim. The majority of dengue cases were concentrated around Wipim (87%) and there was no significant difference in the age distribution of dengue-infected and other patients, which suggests an outbreak pattern of disease rather than endemic transmission.

Malaria was detected in 3 cases by both microscopy and PCR. The low prevalence (2%) of malaria at Wipim was a surprise, as these samples were collected before large-scale, free bednet distribution and this area had previously been categorized as hyperendemic for malaria (22). There were no malaria-dengue concurrent infections detected in this study, which is consistent with the paucity of studies reporting co-infections between these two pathogens (23).

An aetiological agent was detected in only 13% of febrile patients, with DENV-1 and malaria the only pathogens identified. We did not conduct an exhaustive search for the aetiological agents of fever in the clinical samples that were analysed in this study. Previous research has shown that typhoid, influenza, *Streptococcus pneumoniae* and other pathogens are important causes of febrile illnesses in the country (8,24,25). The purpose of this study was to screen samples for arthropod-borne pathogens which have not been extensively studied in PNG.

This study was an initial effort to determine which pathogens were circulating in the Western Province area and contributing to febrile illnesses. However, the current study has a number of limitations that should be

TABLE 2

Analysis of variables associated with dengue at Wipim Health Centre

Variable	DENV positive	DENV negative	p value
Number of patients	15	121	
Number (%) female	7 (46.7)	65 (53.7)	0.785[a]
Mean age (years)	12.5	19.0	0.712[b]
Number (%) residents of Wipim	13 (86.7)	61 (50.4)	**0.011**[a]

DENV = dengue virus
[a] Determined using Fisher's Exact test
[b] Determined using median test

mentioned. Dried blood spots have been used previously to detect dengue viruses (26,27) and malaria (28,29); however, the detection of other pathogens that were targeted in this survey have not been evaluated. In particular, arboviruses such as Japanese encephalitis virus (JEV) produce a low viraemia in humans which may be difficult to detect from dried blood spots. The nucleic acid extraction methods utilized in this study may not have been optimal for the detection of many arboviral species with an RNA genome targeted in this study. DNA extractions from blood spots were optimized for the detection of *Plasmodium* spp., as malaria surveillance was the primary objective of the study. The low detection rate of pathogenic organisms may be due to the suboptimal specimens and extraction methods utilized in this study. However, the low detection rate may also reflect the large range of potential pathogens that may cause febrile illnesses in PNG and other low-income settings. The selection of patients was based upon a self-reported history of fever, which may not reflect the true burden of febrile illnesses. In addition, patients reporting a history of fever may have been convalescent and no longer had pathogens in their bloodstream. Notwithstanding these limitations, dengue was detected in a high proportion of patients, which shows that this pathogen can be an important cause of febrile illness in PNG and should be considered by clinicians during disease outbreaks. It is evident that much more surveillance and research is required to better understand the distribution, epidemiology and burden of dengue in PNG. In addition, appropriate diagnostic tools are required at point-of-care in order to enable clinicians to diagnose and manage patients more appropriately.

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REFERENCES

1 **Animut A, Mekonnen Y, Shimelis D, Ephraim E.** Febrile illnesses of different etiology among outpatients in four health centers in Northwestern Ethiopia. *Jpn J Infect Dis* 2009;62:107-110.
2 **Murdoch DR, Woods CW, Zimmerman MD, Dull PM, Belbase R, Keenan AJ, Scott RM, Basnyat B, Archibald LK, Reller LB.** The etiology of febrile illness in adults presenting to Patan hospital in Kathmandu, Nepal. *Am J Trop Med Hyg* 2004;70:670-675.
3 **Müller I, Bockarie M, Alpers M, Smith T.** The epidemiology of malaria in Papua New Guinea. *Trends Parasitol* 2003;19:253-259.
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 **Hetzel MW, Paul S, Benjamin L, Makita L, Mueller I, Siba PM.** The proportion of fevers attributable to malaria varies significantly between sites in Papua New Guinea. *PNG Med J* 2014;57:39-51.
6 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.
7 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.
8 **Siba V, Horwood PF, Vanuga K, Wapling J, Sehuko R, Siba PM, Greenhill AR.** The evaluation of serological diagnostic tests for typhoid fever in Papua New Guinea using a composite reference standard. *Clin Vaccine Immunol* 2012;19:1833-1837.
9 **Senn N, Luang-Suarkia D, Manong M, Siba PM, McBride WJH.** Contribution of dengue fever to the burden of acute febrile illnesses in Papua New Guinea: an age-specific prospective study. *Am J Trop Med Hyg* 2011;85:132-137.
10 **Jonduo MH, Bande G, Horwood PF.** Arboviruses of human health significance in Papua New Guinea. *PNG Med J* 2012;55:35-44.
11 **McNamara DT, Thomson JM, Kasehagen LJ, Zimmerman PA.** Development of a multiplex PCR-ligase detection reaction assay for diagnosis of infection by the four parasite species causing malaria in humans. *J Clin Microbiol* 2004;42:2403-2410.
12 **Kasehagen LJ, Mueller I, McNamara DT, Bockarie MJ, Kiniboro B, Rare L, Lorry K, Kastens W, Reeder JC, Kazura JW, Zimmerman PA.** Changing patterns of *Plasmodium* blood-stage infections in the Wosera region of Papua New Guinea monitored by light microscopy and high throughput PCR diagnosis. *Am J Trop Med Hyg* 2006;75:588-596.
13 **DaRe JT, Kouri DP, Zimmerman PA, Thomas PJ.** Differentiating *Plasmodium falciparum* alleles by transforming Cartesian X,Y data to polar coordinates. *BMC Genet* 2010;11:57.
14 **Barnadas C, Kent D, Timinao L, Iga J, Gray LR, Siba P, Mueller I, Thomas PJ, Zimmerman PA.** A new high-throughput method for simultaneous detection of drug resistance associated mutations in *Plasmodium vivax dhfr*, *dhps* and *mdr1* genes. *Malar J* 2011;10:282.
15 **Kong YY, Thay CH, Tim TC, Devi S.** Rapid detection, serotyping and quantitation of dengue viruses by TaqMan real-time one-step RT-PCR. *J Virol Methods* 2006;138:123-130.
16 **Pyke AT, Smith IL, van den Hurk AF, Northill JA,**

Chuan TF, Westacott AJ, Smith GA. Detection of Australasian Flavivirus encephalitic viruses using rapid fluorogenic TaqMan RT-PCR assays. *J Virol Methods* 2004;117:161-167.
17 **Edwards CJ, Welch SR, Chamberlain J, Hewson R, Tolley H, Cane PA, Lloyd G.** Molecular diagnosis and analysis of Chikungunya virus. *J Clin Virol* 2007;39:271-275.
18 **Hall RA, Prow NA, Pyke AT.** Ross River virus. In: Liu D, ed. Molecular Detection of Human Viral Pathogens. Boca Raton, Florida: CRC Press, 2010.
19 **Paris DH, Blacksell SD, Stenos J, Graves SR, Unsworth NB, Phetsouvanh R, Newton PN, Day NPJ.** Real-time multiplex PCR assay for detection and differentiation of rickettsiae and orientiae. *Trans R Soc Trop Med Hyg* 2008;102:186-193.
20 **Zigas V, Doherty RL.** An outbreak of dengue in the Rabaul community. *PNG Med J* 1973;16:42-45.
21 **Kiedrzynski T, Souarès Y, Stewart T.** Dengue in the Pacific: an updated story. *Pacific Health Dialog* 1998;5:129-136.
22 **Parkinson AD.** Malaria in Papua New Guinea 1973. *PNG Med J* 1974;17:8-16.
23 **Epelboin L, Hanf M, Dussart P, Ouar-Epelboin S, Djossou F, Nacher M, Carme B.** Is dengue and malaria co-infection more severe than single infections? A retrospective matched-pair study in French Guiana. *Malar J* 2012;11:142.
24 **Kelso A, Reading PC.** Influenza in the Pacific. *PNG Med J* 2010;53:180-190.
25 **Anga G, Barnabas R, Kaminiel O, Tefuarani N, Vince J, Ripa P, Riddell M, Duke T.** The aetiology, clinical presentations and outcome of febrile encephalopathy in children in Papua New Guinea. *Ann Trop Paediatr* 2010;30:109-118.
26 **Matheus S, Meynard JB, Lacoste V, Morvan J, Deparis X.** Use of capillary blood samples as a new approach for diagnosis of dengue virus infection. *J Clin Microbiol* 2007;45:887-890.
27 **Cao-Lormeau VM, Roche C, Aubry M, Teissier A, Lastere S, Daudens E, Mallet HP, Musso D, Aaskov J.** Recent emergence of dengue virus serotype 4 in French Polynesia results from multiple introductions from other South Pacific Islands. *PLoS One* 2011;6:e29555.
28 **Taylor BJ, Martin KA, Arango E, Agudelo AM, Maestre A, Yanow SK.** Real-time PCR detection of *Plasmodium* directly from whole blood and filter paper samples. *Malar J* 2011;10:244-251.
29 **Hwang J, Jaroensuk J, Leimanis ML, Russel B, McGready R, Day N, Snounou G, Nosten F, Imwong M.** Long-term storage limits PCR-based analyses of malaria parasites in archival dried blood spots. *Malar J* 2012;11:339.

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Diagnostic capacity and antimalarial availability in Papua New Guinea before the introduction of a revised national malaria treatment protocol

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SUMMARY

***Background*: Papua New Guinea (PNG) introduced a revised national malaria treatment protocol (NMTP) in late 2011. Successful implementation of the revised protocol requires all health facilities in PNG to have reliable access to microscopy or malaria rapid diagnostic kits as well as a reliable supply of all recommended first-line medications. This paper presents findings from a study that sought to assess the availability of microscopy, malaria rapid diagnostic kits and recommended first-line antimalarial medication in Papua New Guinean health facilities across the country before the introduction of the revised treatment protocol. *Methods*: A country-wide cross-sectional survey of 79 randomly selected health centres, health subcentres and aid posts. Data were collected via an interviewer-administered questionnaire completed with the officer in charge of participating health facilities. *Results*: Overall, 15% of surveyed health facilities had unexpired rapid diagnostic test (RDT) in stock or working microscopy available. A recommended first-line antimalarial for uncomplicated malaria was available in 85% of health facilities. The preferred first-line antimalarial combination for treating severe malaria was present in 42% of health facilities, although 68% had the capacity to provide either the preferred or recommended substitute first-line medication for severe malaria. The total number of health workers employed in the 79 surveyed health facilities was 443, only 3 of whom were medical doctors. *Conclusions*: Our findings indicate that diagnostic capacity was low in Papua New Guinean health facilities before the introduction of the new NMTP and that access to recommended first-line antimalarial medication was variable. Substantial improvements in diagnostic capacity and antimalarial procurement and distribution will need to be made if the revised protocol is to be adhered to.**

Background

Papua New Guinea (PNG) implemented a revised national malaria treatment protocol (NMTP), later entitled the National Malaria Treatment Policy (1), in all government and church-run health facilities in late 2011. The revised NMTP stipulates that all fever or suspected malaria cases should be tested for malaria infection by rapid diagnostic test (RDT) or microscopy and that antimalarials should only be prescribed upon confirmation of malaria parasitaemia (1). This contrasts with the former protocol that stipulated the provision of antimalarials to all children with fever and, where microscopy was not available, presumptive diagnosis in febrile adults (2,3). The revised NMTP also heralded

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a substantial change in recommended antimalarial medication regimens. In the obsolete protocol amodiaquine (AQ) plus sulphadoxine-pyrimethamine (SP) or chloroquine (CQ) plus SP was recommended for the treatment of uncomplicated malaria. Artemether injection followed by artesunate tablets plus SP or, if artemether and artesunate were unavailable, quinine injection followed by quinine tablets plus SP, was recommended for the treatment of severe malaria. The revised NMTP introduced artemether-lumefantrine (AL) as the new first-line treatment for uncomplicated falciparum malaria, AL plus primaquine as the new first-line treatment for uncomplicated vivax malaria and artesunate injection followed by AL for the first-line treatment of severe malaria, whether falciparum or vivax (1).

The new NMTP promises an improvement in malaria case management for a number of reasons. These include: the well-established sensitivity and specificity of malaria RDTs (4); the proven utility of malaria RDTs in health services across the world (5), including PNG (6); the demonstrated efficacy of artemisinin-based combination therapies (ACTs) in treating malaria (7-9); and the widespread resistance to the current recommended first-line antimalarial medications (7,10). Nevertheless, if the revised NMTP is to improve clinical practice and treatment outcome then adequate supplies of RDTs and ACTs will need to be reliably distributed across an extensive health facility network and, once available, incorporated into routine malaria case management practice. Malaria RDT kits were present in the PNG health system before the introduction of the revised NMTP, as was the capacity for malaria diagnosis via microscopy. A Round 3 grant from the Global Fund to Fight AIDS, Tuberculosis and Malaria (GFATM) had specifically aimed to improve microscopic diagnosis in health centres and introduce RDTs for malaria in health facilities without microscopy during the period 2004 to 2009 (11). However, the availability of microscopy, RDT and first-line antimalarials, inclusive of the recommended ACT regimen for severe malaria, has not been thoroughly examined in PNG. For example, it is not reliably known how comprehensive the coverage with malaria RDT kits is in health facilities across the country or how often diagnostic tests are utilized before the administration of specific treatment. Equally unknown is the extent to which first-line antimalarial medications are available in health facilities across the country.

This paper addresses this gap in part by presenting findings from a country-wide health facility survey of malaria case management capacity and practice. The survey was conducted as part of the monitoring and evaluation component of a Round 8 GFATM grant supporting the implementation of the NMTP. Specifically, this paper presents country-wide data on the availability of malaria diagnostic tools and first-line antimalarial medication current at the time of data collection as well as selected characteristics of the health work force. A companion paper presents data on malaria case management practice (12). The reported findings will provide some insight into diagnostic capacity before the introduction of the new NMTP as well as some indication of how effective the antimalarial procurement and distribution system is in maintaining reliable medication supplies across the health facility network.

Methods

Study setting

PNG comprises 20 provinces divided into four regions (Highlands, Momase, Southern and Islands) according to their geographical location. Malaria endemicity ranges from holoendemic transmission in large parts of the Momase, Southern and Islands Regions to unstable transmission with localized epidemics in many areas of the Highlands Region (13). In total, over 90% of the population are considered at risk of malaria infection (14). The majority of health services in PNG are delivered through government and church providers via an extensive health facility network. This network comprises four levels of service provision including hospital, health centre, health subcentre and aid post. The hospital is the largest unit of health service provision in terms of the number and range of clinical staff employed and the number and range of clinical services provided. The aid post is the smallest unit of health service provision and is typically staffed by a single health worker with two years' clinical training. In theory, all cases of uncomplicated malaria are treatable at the aid post level with severe cases referred to a health centre or hospital. Current clinical positions in the PNG health system include medical doctors, health extension officers (HEOs), nursing officers, community health workers (CHWs) and rural/

medical laboratory assistants (RLA/MLAs).

Study sites

This study was carried out country-wide (including all 20 provinces) in areas with endemic or potentially epidemic malaria. The study sample consisted of two health centres or health subcentres and up to four aid posts selected from each province using a simple random sampling procedure with province stratification. The sampling frame included all health centres/subcentres operational in March 2010 from an inventory provided by the National Department of Health (n = 689). Aid posts were randomly selected on site at participating (ie, randomly selected and consenting) health centres/subcentres. The sampling frame for aid posts was all operational aid posts under the supervision of the health centre/subcentre at the time of survey. Hospitals were excluded from the sampling frame as they are few in number and serve only a minority of the PNG population. Conversely, health centres, health subcentres and aid posts are widely available across the country and are the main providers of primary care. The term 'health facility' as used in this paper therefore includes health centres, health subcentres and aid posts, but excludes hospitals.

Procedure

The survey was carried out from June to November 2010 by three trained field teams. Each team consisted of two research assistants and a graduate (Bachelor's level) scientific officer. Each team spent a maximum of five days in each health centre/subcentre and up to one day in each aid post. Before any health facility visit the respective provincial health authorities were informed of the study objectives, sites and timetable. Each Provincial Health Authority was asked to commission a health officer to accompany the field team. Oral informed consent was sought from the officer in charge at all participating health facilities and from all participating health workers. This study was approved and granted ethical clearance by the Medical Research Advisory Committee of Papua New Guinea (MRAC No 10.12; 26 Feb 2010).

Instruments

This paper presents data collected from a checklist completed with the officer in charge of each participating health facility. The checklist assessed the human resource capacity and the availability of supplies relevant to malaria case management. Key questions included the number of clinical staff employed and their respective qualifications, the quantity of RDT in stock, the quantity of microscopes and availability of essential microscopy supplies, and the availability of a range of antimalarial medications. Recorded numbers of clinical staff were based on figures provided by the officer in charge. All reported RDT stock, microscopes, including their essential supplies of Giemsa stain, slides and power for a light bulb microscope, antimalarials and other reported medical equipment or supplies were observed by the respective Papua New Guinea Institute of Medical Research (PNGIMR) field team leaders.

Data analysis

All data were double entered into DMSys version 5.1 (Sigma Soft International). Data analysis was performed using Intercooled Stata version 9. Univariable analysis was performed to describe the characteristics of the various samples. Between-group differences on selected variables were examined by Chi-squared statistics.

Results

Study sample

A total of 79 health facilities were visited in 20 provinces. Of these, 40 were health centres or subcentres and 39 were aid posts. The distribution of health facility type in each region is shown in Table 1.

Availability of diagnostic tools

As shown in Table 2, a total of 15% of surveyed health facilities had unexpired RDT in stock or working microscopy available. Working microscopy was defined as the presence of a functional microscope, all essential supplies – Giemsa stain, slides and (in the case of electric microscopes) power – and a trained RLA or MLA in employment. All unexpired RDT kits and working microscopy were observed in health centres/subcentres (nil observed in aid posts). However, it should be noted that microscopy would not normally be expected to be present in an aid post setting. No health centre/subcentre had both unexpired RDT kits and working microscopy

TABLE 1

Surveyed health facilities by type and region

Health facility type	Region				Total
	Southern No (%)	Highlands No (%)	Momase No (%)	Islands No (%)	
Health centre/subcentre	12 (48)	10 (59)	8 (50)	10 (48)	40 (51)
Aid post	13 (52)	7 (41)	8 (50)	11 (52)	39 (49)
Total	25	17	16	21	79

TABLE 2

Percentage of health facilities with unexpired RDT in stock, working microscopy available, or either unexpired RDT or working microscopy

Diagnostic test	Health centre/ subcentre (n = 40)		Aid post (n = 39)		Overall (n = 79)	
	%	(95% CI)	%	(95% CI)	%	(95% CI)
RDT	17.5	(7.3-32.8)	0	-	8.9	(3.6-17.4)
Microscopy[a]	12.5	(4.2-26.8)	0[b]	-	6.3	(2.1-14.2)
RDT or microscopy	**30.0**	**(16.6-46.5)**	**0**	**-**	**15.2**	**(8.1-25.0)**

RDT = rapid diagnostic test
CI = confidence interval
[a] Working microscopy was defined as the presence of a functional microscope, all essential supplies – Giemsa stain, slides and (in the case of electric microscopes) power – and a trained rural laboratory assistant or medical laboratory assistant in employment
[b]Working microscopy was not expected in aid post settings (ie, '0' was the expected result)

at the time of survey. A greater percentage of health facilities in the less malarious Highlands Region had RDT or microscopy capacity available than in the combined (malaria-endemic) Southern, Momase and Islands Regions (23.5% vs 12.9%), although this difference did not reach a level that was considered significant ($\chi^2 = 1.169$, $p = 0.280$), and was primarily attributable to variance in the availability of working microscopy (11.8% vs 4.8%).

The total number of unexpired RDT kits across the 8.9% (7/79) of health facilities in which they were present was 4089. The median number of RDT kits per health facility in which they were present was 368 (range 38-2500). An additional 8.9% (7/79) of health

facilities (five health centres/subcentres and two aid posts) had expired RDT in stock. Five of the health facilities with expired RDT in stock were in provinces with endemic malaria transmission. The total number of expired RDT kits in these additional seven health facilities was 1024 (median per health facility 167; range 19-375).

Similarly, an additional 8.9% (7/79) of health facilities (in addition to those reported in Table 2) were observed to have a microscope available; however, none of these additional seven health facilities had a trained RLA or MLA in employment, two had no Giemsa stain in stock and one (with an electric microscope) had no power supply. Overall, a total of 13 microscopes were observed by the PNGIMR field team in the 79 health facilities surveyed (6 functional). Only one health facility had more than one microscope.

Availability of antimalarial medication

Table 3 lists the percentage of health facilities with a range of antimalarial drugs in stock. As can be seen, the most commonly available antimalarial was amodiaquine, followed by chloroquine and SP (present in 90%, 89% and 86% of surveyed health facilities, respectively). Table 4 lists the percentage of health facilities with the recommended first-line antimalarial combinations (current in the country at the time of data collection and as described above in Background) in stock. As shown in Table 4, 85% of health

TABLE 3

PERCENTAGE OF HEALTH FACILITIES WITH SELECTED ANTIMALARIALS IN STOCK*

Medication type[a]	%	95% CI
Amodiaquine (Camoquin)	89.9	(81.0-95.5)
Artemisinin-naphthoquine (Arco)	2.5	(0.3-8.8)
Artemether injections	49.4	(37.9-60.9)
Artemether tablets	54.4	(42.8-65.7)
Artesunate injections	24.1	(15.1-35.0)
Artesunate suppositories	2.5	(0.3-8.8)
Chloroquine (Nivaquine)	88.6	(79.5-94.7)
Dihydroartemisinin-piperaquine	2.5	(0.3-8.8)
Doxycycline (Vibramycin)	70.9	(59.6-80.6)
Sulphadoxine-pyrimethamine (Fansidar)	86.1	(76.5-92.8)
Atovaquone-proguanil (Malarone)	3.9	(0.8-11.0)
Primaquine	73.1	(61.8-82.5)
Quinine injections	62.0	(50.4-72.7)
Quinine tablets	82.3	(72.1-90.0)

*The quantity of each medication was not accounted for in this analysis; rather, the data represent the percentage of health facilities that had at least one vial or container (inclusive of a single, opened container) of the respective antimalarial in stock
[a]Popular trade names are indicated in parentheses
CI = confidence interval

TABLE 4

PERCENTAGE OF HEALTH FACILITIES WITH CURRENT FIRST-LINE ANTIMALARIALS IN STOCK*

Diagnosis	Medication	%	95% CI
Uncomplicated	AQ + SP	82.3	(72.1-90.0)
	CQ + SP	81.0	(70.6-89.0)
	AQ + SP or CQ + SP	84.8	(75.0-91.9)
Severe	AI + AT + SP	41.8	(30.8-53.4)
	QI + QT + SP	51.9	(40.4-63.3)
	AI + AT + SP or QI + QT + SP	68.4	(56.9-78.4)

*The quantity of each medication was not accounted for in this analysis; rather, the data represent the percentage of health facilities that had at least one vial or container (inclusive of a single, opened container) of the respective antimalarial in stock
CI = confidence interval
AQ = amodiaquine
SP = sulphadoxine-pyrimethamine
CQ = chloroquine
AI = artemisinin injection
AT = artemisinin tablet
QI = quinine injection
QT = quinine tablet

facilities had the capacity to provide one of the two recommended first-line regimens for uncomplicated malaria (both combination regimens were available in 78.5% of the surveyed facilities). The preferred first-line antimalarial combination for treating severe malaria (artemether injection plus artesunate tablets plus SP) was present in 42% of health facilities and 68% had the capacity to provide artemether injection plus artesunate tablets plus SP or quinine injection plus quinine tablets plus SP. Oral artemisinin combination therapies were found in 2.5% of health facilities surveyed (artemisinin-naphthoquine), whilst oral artemisinin monotherapies were located in 54% of health facilities (Table 3).

A higher percentage of health centres/subcentres than aid posts had at least one of the two recommended antimalarials for uncomplicated malaria available (92.5% vs 76.9%), a difference approaching significance (χ^2 = 3.719, p = 0.054). Differences by health facility type in the availability of the recommended artemisinin-based combination therapy for severe malaria were marginal and did not reach significance (42.5% in health centre/subcentre vs 41.0% in aid post; χ^2 = 0.018, p = 0.894). However, the quinine-based combination for treating severe malaria was available in a greater number of health centres/subcentres than in aid posts (67.5% vs 35.9%) and this difference was significant (χ^2 = 7.900, p <0.01). Differences between the less malarious Highlands Region and combined (malaria-endemic) Southern, Momase and Islands Regions were also evident in terms of the availability of all three of these antimalarial combinations (AQ + SP or CQ + SP 88.2% vs 83.9%, ACT for severe malaria 52.9% vs 38.7%, quinine for severe malaria 70.6% vs 46.8%), although the reported differences did not reach a level that was considered significant (χ^2 = 0.197, p = 0.657, χ^2 = 1.111, p = 0.292, χ^2 = 3.031, p = 0.082, respectively).

Human resource capacity

Among the 443 health workers employed

in all the health facilities, 0.7% (3/443) were doctors, 3.6% (16/443) were HEOs, 32.5% (144/443) were nurses, 59.4% (263/443) were CHWs and 3.8% (17/443) were RLAs or MLAs. A mean of 9.98 (median = 8) health workers were employed in health centres/ subcentres at the time of survey and a mean of 1.13 (median = 1) in aid posts. No doctors or HEOs were employed at any of the 39 aid posts included in the survey. A total of 2/40 surveyed health centres/subcentres had one or more doctors in employment and 13/40 had at least one HEO in employment.

Discussion and Conclusions

This paper sought to assess the availability of microscopy, RDTs and first-line antimalarials in PNG health facilities and to provide some insight into human resource capacity with regard to malaria case management. The findings indicate that parasitological diagnosis of malaria is rarely available in health facilities across PNG. Microscopy is usually dysfunctional due to the absence of microscopes, reagents and skilled laboratory technicians. As an alternative to microscopy, an RDT is a reliable, cost-effective, fast and easy-to-perform means to determine the presence of malaria infection. However, RDTs were also largely unavailable with only 8.9% of surveyed health facilities having unexpired kits in stock. Aid posts, the lowest level of health service provision, are usually the facilities closest to the patient's home and hence most easily accessible. These facilities totally lacked any form of diagnostic test for malaria. It was of note that as many health facilities had expired RDTs in stock as had unexpired RDTs, suggesting that available diagnostic tools are underutilized even if they are easy to use. Microscopy has been present in the health system for decades being considered the gold standard for malaria diagnosis. Nevertheless, most health facilities are unable to maintain the capacity for microscopy services. Considering the lack of skilled laboratory staff in health facilities and apparent problems in the delivery of essential microscopy supplies, RDT diagnosis for malaria may be more realistic in an operational context than increasing the utilization of microscopes.

The availability of current first-line antimalarials varied. The preferred first-line medication recommended for use in cases of severe malaria in the treatment guideline current at the time of survey (artemether injection + artesunate tablets + SP) was only available in 42% of surveyed health facilities. However, the first-line medications for uncomplicated malaria were more widely available with at least one of the two recommended regimens present in 85% of health facilities. The widespread availability of chloroquine, amodiaquine and SP might suggest a reasonably well-working drug distribution. On the other hand, the relative scarcity of potentially life-saving treatment for severe malaria may indicate a problem in the medical supplies system. Having said this, as the quantity of antimalarial supply was not taken into account, it remains possible that many of the health facilities with the first-line antimalarials in stock may still have been considered in short supply. In addition, the quality of the stocked medication as well as their expiry date was not examined as part of this study, raising the possibility that available (and possibly prescribed) medications may not have met the required quality standard in at least some cases. Of obvious additional concern are the substantial levels of resistance to chloroquine and amodiaquine that have been reported in PNG for the past 20 to 30 years (15,16) and have been increasing ever since (10). Hence, the revised treatment protocol for malaria in PNG recommends the use of different artemisinin-based combination regimens for the treatment of all cases of malaria (1).

Regional differences in diagnostic capacity and antimalarial availability were also of note. Health facilities in the PNG Highlands Region had a greater diagnostic capacity relative to health facilities in the other, more malarious, regions of PNG. Similarly, a greater percentage of surveyed health facilities in the Highlands Region had the recommended first-line medications for severe malaria in stock than did health facilities elsewhere. Thus, at the time of survey, health facilities in the region of PNG with the lowest levels of malaria transmission had the greatest capacity for diagnosis and treatment of severe malaria. It is not possible to determine why this was the case based on the study findings presented in this paper. The discrepancy may have resulted from a greater demand on available malaria diagnostic and treatment resources in the malaria-endemic Southern, Momase and Islands Regions. Whatever the cause, this finding does suggest that the distribution of malaria diagnostics and medicines in the

malaria-endemic regions of PNG warrants closer scrutiny, in order to correct possible underestimates in supply or inefficiencies in the procurement and distribution system.

There were a number of challenges associated with the collection of data and therefore some potential limitations of the study we have described. The final sample size, and especially the number of aid posts included in the sample, was lower than anticipated. This was largely due to the inaccessibility of aid posts or, more frequently, the absence of any functional aid post to survey. The study was conducted during a period of typically low malaria transmission (June-November, 2010). Thus the number of malaria patients presenting to health facilities and the subsequent pressure on resources (eg, RDT kits, antimalarial medication) may have been lower during the survey period than during peak transmission periods. Nevertheless, the officer in charge of any health facility in PNG can request additional medical supplies anytime as required (although the requested medical supplies may not always be available for delivery). Despite these limitations, it may be reasonably concluded that parasitological diagnosis of malaria was not widely available in health facilities across the country during the survey period, whilst the availability of first-line antimalarials varied.

The renewed effort to reduce the burden of malaria in PNG supported by grants from the Global Fund requires improvements in access to prompt and appropriate diagnosis and treatment for all cases of malaria (17). The universal availability of diagnostic tools and efficacious medicines is a prerequisite for any control program. In accordance with international recommendations (18) the new malaria treatment protocol specifies that all suspected malaria patients must be tested for malaria infection by RDT or microscopy and only positive patients prescribed an ACT (1). To successfully implement this protocol, the current medical supplies system will need to be strengthened in order to ensure constant supplies of diagnostics and antimalarial drugs. As improving malaria diagnostic capacity was one aim of the Round 3 GFATM grant concluded before the completion of this survey, the reported findings suggest that any such gains were likely to have been modest at best or difficult to sustain (eg, a continuous RDT supply). Ensuring that a high proportion of health facilities have either RDT or microscopy diagnostic capacity available during the course of the Round 8 GFATM grant therefore presents a particularly challenging undertaking. A recent evaluation of the mosquito net distribution component of the Round 3 GFATM grant further suggests that PNG is a particularly challenging environment in which to implement malaria control programs and recommends that program success be assessed against locally appropriate targets rather than the often more ambitious international norms (19). Innovative applications to monitor RDT and drug stocks at health facilities in real time may contribute to improved supply if reports of shortages trigger immediate action (20). In addition, other aspects of access to malaria treatment need to be considered and included in a comprehensive control strategy (21), such as the proper application of the malaria treatment protocol by health workers. This aspect was investigated in a companion paper which showed serious shortcomings in the treatment of malaria in PNG (12).

The importance of implementing a test-and-treat policy with the new treatment protocol extends beyond its clinical benefit for individual patients. Data collected from five health centres across PNG showed large variations in the proportion of fever cases tested positive for malaria by light microscopy (2% to 75%) and significant reductions of these proportions after the distribution of mosquito nets (22). In order to adequately assess the burden of malaria in PNG and the impact of interventions on malaria incidence, case reporting should be based on a parasitological rather than a clinical diagnosis. Despite imperfect sensitivity and specificity of some RDTs (4), their application can significantly improve malaria diagnosis, leading to a reduction in overtreatment and more adequate reporting of malaria through routine reporting channels such as PNG's National Health Information System (23).

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REFERENCES

1 **Papua New Guinea Department of Health.** National Malaria Treatment Policy. Port Moresby: Department of Health, Sep 2009.

2 **Papua New Guinea Department of Health.** Standard Treatment for Common Illnesses of Children in Papua New Guinea: A Manual for Nurses, Community Health Workers, Health Extension Officers and Doctors. Eighth edition. Port Moresby: Department of Health, 2005.

3 **Papua New Guinea Department of Health.** Standard Treatment for Common Illnesses of Adults in Papua New Guinea: A Manual for Nurses, Community Health Workers, Health Extension Officers and Doctors. Port Moresby: Department of Health, 2005.

4 **World Health Organization.** Malaria rapid diagnostic test performance: results of WHO product testing of malaria RDTs: round 1 (2008). Geneva: World Health Organization, 2009.

5 **World Health Organization.** The role of laboratory diagnosis to support malaria disease management. Focus on the use of rapid diagnostic tests in areas of high transmission. Report of a WHO Technical Consultation, 25-26 October 2004. WHO/HTM/MAL/2006.1111. Geneva: World Health Organization, 2006.

6 **Senn N, Rarau P, Manong D, Salib M, Siba P, Robinson LJ, Reeder J, Rogerson S, Mueller I, Genton B.** Rapid diagnostic test-based management of malaria: an effectiveness study in Papua New Guinean infants with *Plasmodium falciparum* and *Plasmodium vivax* malaria. *Clin Infect Dis* 2012;54:644-651.

7 **Karunajeewa HA, Mueller I, Senn M, Lin E, Law I, Gomorrai PS, Oa O, Griffin S, Kotab K, Suano P, Tarongka N, Ura A, Lautu D, Page-Sharp M, Wong R, Salman S, Siba P, Ilett KF, Davis TME.** A trial of combination antimalarial therapies in children from Papua New Guinea. *N Engl J Med* 2008;359:2545-2557.

8 **Sinclair D, Zani B, Donegan S, Olliaro P, Garner P.** Artemisinin-based combination therapy for treating uncomplicated malaria. *Cochrane Database Syst Rev* 2009;CD007483.

9 **Sinclair D, Gogtay N, Brand F, Olliaro P.** Artemisinin-based combination therapy for treating uncomplicated *Plasmodium vivax* malaria. *Cochrane Database Syst Rev* 2011;CD008492.

10 **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;77:947-954.

11 **Global Fund to Fight AIDS, Tuberculosis and Malaria.** Community-based Malaria Prevention and Control in Papua New Guinea – Nationwide Insecticide-treated Nets (ITN) Distribution and Expansion of Confirmed Diagnosis and Appropriate Treatment of Malaria. Geneva: Global Fund, 2004. http://portfolio.theglobalfund.org/en/Grant/Index/PNG-304-G01-M

12 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

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

14 **World Health Organization.** World Malaria Report 2010. Geneva: World Health Organization, 2010.

15 **Grimmond TR, Donovan KO, Riley ID.** Chloroquine resistant malaria in Papua New Guinea. *PNG Med J* 1976;19:184-185.

16 **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.

17 **Hetzel MW.** An integrated approach to malaria control in Papua New Guinea. *PNG Med J* 2009;52:1-7.

18 **World Health Organization.** Guidelines for the Treatment of Malaria. Geneva: World Health Organization, 2006.

19 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.

20 **Barrington J, Wereko-Brobby O, Ward P, Mwafongo W, Kungulwe S.** SMS for Life: a pilot project to improve anti-malarial drug supply management in rural Tanzania using standard technology. *Malar J* 2010;9:298.

21 **Obrist B, Iteba N, Lengeler C, Makemba A, Mshana C, Nathan R, Alba S, Dillip A, Hetzel MW, Mayumana I, Schulze A, Mshinda H.** Access to health care in contexts of livelihood insecurity: a framework for analysis and action. *PLoS Med* 2007;4:1584-1588.

22 **Hetzel MW, Gideon G, Mueller I, Siba PM.** Papua New Guinea/The Global Fund Round 3 Malaria Control Programme Evaluation 2008/2009: Results from Cross Sectional Surveys and Sentinel Sites. Goroka: Papua New Guinea Institute of Medical Research, 2010.

23 **Ishengoma DS, Francis F, Mmbando BP, Lusingu JP, Magistrado P, Alifrangis M, Theander TG, Bygbjerg IC, Lemnge MM.** Accuracy of malaria rapid diagnostic tests in community studies and their impact on treatment of malaria in an area with declining malaria burden in north-eastern Tanzania. *Malar J* 2011;10:176.

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Primaquine treatment for *Plasmodium vivax* – an essential tool for malaria control and elimination in Papua New Guinea

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SUMMARY

***Plasmodium vivax* is a major cause of malarial infection and disease in young children in Papua New Guinea. Recent increase in funding for malaria control has improved accessibility to preventive measures, diagnosis and artemisinin combination therapies. Yet the current treatment and control measures are more effective against *P. falciparum* than against *P. vivax* and *P. ovale* due to the biological differences in the liver stage life-cycle of these parasites. *P. vivax* and *P. ovale* have a dormant liver stage called a hypnozoite. The artemisinin combination therapies, while highly effective against the blood stages of all plasmodium species causing human malarial illness, have no effect upon the hypnozoites in the liver and the stage V gametocytes of *P. falciparum*. Currently, primaquine is the only licensed drug shown to be effective against both the hypnozoites of *P. vivax* and *P. ovale* in the liver and the stage V gametocytes of *P. falciparum*. Primaquine has a high associated risk of life-threatening haemolytic anaemia when administered to glucose-6-phosphate dehydrogenase (G6PD)-deficient persons. The lack of cheap, reliable point-of-care testing for the diagnosis of G6PD deficiency remains a major obstacle to the widespread use of primaquine in clinical and public health practice. Furthermore, there is a paucity of primaquine safety and tolerability data, especially in young children with the highest *P. vivax* disease burden. For malaria control and elimination efforts to be effective, interventions such as mass drug administration must include primaquine. This opinion paper highlights the need to eradicate hypnozoites in the liver of the human host with primaquine treatment for radical cure of malarial illness and discusses the challenges in the use of primaquine as a public health tool for malaria control and elimination programs in countries such as Papua New Guinea.**

Introduction

Papua New Guinea (PNG) exhibits one of the highest endemicity levels of *Plasmodium vivax*, the most widely distributed plasmodium species, with an estimated 2.5 billion at-risk population globally (1). The current treatment regimens and malaria control measures are more effective against *Plasmodium falciparum* than *P. vivax,* as shown in countries like Brazil (2) and Thailand (3), where sustained malaria control and case management programs

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have led to a proportional increase in the prevalence of *P. vivax,* replacing *P. falciparum* as the predominant species. *P. vivax* is more difficult to control due to the dormant liver stage in its life-cycle called a hypnozoite.

Biology of hypnozoite relapses in *P. vivax* and *P. ovale*

In contrast to *P. falciparum*, *P. vivax* and *P. ovale* both have the ability to develop into hypnozoites that can remain dormant in the parenchymal cells of the host liver following an acute infection. After a period of time, which varies in duration depending on the geographical area, hypnozoites can spontaneously reactivate, causing the release of new merozoites into the blood stream triggering a new reproduction cycle. *P. vivax* strains from tropical regions cause relapses more frequently at approximately three-weekly intervals and more often than strains from temperate regions, which take about 8 to 10 months after the initial infection (4). The risk of relapse within a month of the primary parasitaemia often exceeds 50% and multiple relapse episodes (three or more) can occur following the first relapse episode (5). The latent phase of *P. vivax* in the liver is therefore an important source of new arising infections, even more so as the current control measures and treatment regimens may not effectively address this particular source of new clinical episodes. Little is known of *P. ovale,* often associated with mixed infections in PNG (6). *P. ovale* malaria cases of African origin have been seen in travellers following relapse almost five months after returning from Africa (7). Clinically, relapses may present as a new malaria episode, indistinguishable from a new infection, and with the potential, through the development of gametocytes, to further transmit the infection to a mosquito and eventually to a new human host.

Primaquine hypnozoiticidal therapy for *P. vivax* and *P. ovale*

Historical context

The radical cure of *P. vivax* and *P. ovale* infections requires the treatment of both blood and liver stages of the parasite. For over 60 years, the only drug known to have any effect in eradicating the liver stages of both *P. vivax* and *P. ov*ale has been primaquine (PQ).

PQ is an analogue of pamaquine (plasmoquine), an 8-aminoquinoline drug produced in Germany during the 1920s (8). It is the only licensed drug currently available for radical cure (elimination) of hypnozoites. The current recommendation for PQ, requiring a long, 14-day treatment course, was adopted from work by Sinton and Bird in 1928 (9) on pamaquine, which in combination with quinine seemed to adequately cure *P. vivax* infections, provided the treatment duration was sufficiently long. Studies in healthy non-immune volunteers have shown that, provided an adequate total dose was delivered, the dosing schedule did not affect the overall efficacy of PQ (10). The administration of 60 mg (base) of PQ daily for 7 days was as effective as the 30 mg daily for 14 days in preventing relapses in glucose-6-phosphate dehydrogenase (G6PD)-normal adult volunteers infected with the Chesson strain of *P. vivax* (10). The Chesson strain is an isolate from PNG which is relatively resistant to PQ and thus requires higher doses of PQ to prevent relapses (11). Overall, these past observations and recent studies and reviews support the so-called total dose effect, with the curative activities of PQ being equally effective at the same total dose over 7 and 14 days (5,10). The total dose effect is probably a correlate of the key pharmacokinetic index of area under the curve (AUC).

Toxicity: the problem of G6PD deficiency

There is an important major drawback of implementation of PQ in routine clinical practice for the treatment of hypnozoites. PQ has a high associated risk of side-effects, particularly among people with G6PD deficiency, an X-chromosome-linked hereditary disorder (more common in males than in females) due to mutations in the G6PD gene (12). There are many biochemical and clinical phenotypes due to the functional variants arising from the mutations in the G6PD gene. This enzymatic deficiency, of which about 140 different variants exist, is an absolute contraindication for the use of PQ when the enzyme's activity is below the threshold of 5%, but the drug can be used in milder cases with the provision of spreading the treatment on a weekly basis during a two-month-long 0.75 mg/kg schedule. G6PD deficiency is frequent in malaria-endemic areas such as PNG. Other drugs known to have an association with haemolysis in persons with G6PD deficiency, and which are widely used in PNG, are sulfamethoxazole,

dapsone, nitrofurantoin and cotrimoxazole. The rarity of reported cases of haemolytic anaemia associated with the use of these drugs in PNG suggests that this clinical phenotype is either rare or subclinical. Further work is needed in this area.

Dosing: pharmacological considerations

Experimental challenges carried out in the 1950s showed synergy between blood schizonticides, such as chloroquine (CQ) or quinine, with PQ in preventing relapses, while CQ or quinine administered alone have been shown to have no effect upon the hypnozoites (13). Furthermore, the therapeutic efficacy of weekly administration of PQ was increased when used concurrently with CQ on healthy G6PD-deficient subjects infected with the Chesson strain of *P. vivax* (14). CQ has been the choice of treatment for *P. vivax* infection for a long time but the emergence of drug resistance has forced changes to treatment regimens, and research is needed to find effective combination antimalarial therapies. The risk of relapse after treatment with CQ alone begins after 35 days, reaching 58% by 60 days, while with quinine therapy relapses are encountered earlier, starting at 17 days, with 60% of patients relapsing by day 35 (5). This effect is due to CQ's slower elimination profile leading to more prolonged therapeutic concentrations of CQ, and to the fact that CQ has a longer half-life than quinine (30-60 days vs a few hours) (15).

Primaquine for gametocyte therapy

Besides its activity against hypnozoites, PQ is highly effective in killing the sexual forms of all *Plasmodium* spp. parasites (ie, the gametocytes) (16). This is particularly important in the treatment of *P. falciparum*, whose stage V gametocytes are relatively resistant to treatment with most other blood-stage antimalarials (17,18). Consequently, *P. falciparum* gametocytes are commonly seen for up to 4 weeks after successful treatment of asexual forms and can contribute both to transmission and to potentially faster spread of drug resistance (19). For these reasons, a single dose of 0.75 mg/kg PQ was included in the PNG national treatment guidelines until 2000, when the PQ single dose was dropped in conjunction with the switch from CQ or amodiaquine (AQ) monotherapy to CQ or AQ plus sulfadoxine-pyrimethamine (SP) combination therapy. Following a large consultative process, the World Health Organization (WHO) has recently issued a recommendation for the inclusion of a single dose of 0.75 mg/kg of PQ for the treatment of *P. falciparum* malaria irrespective of G6PD status in places in which there is a threat of artemisinin resistance or where elimination programs are in place (20). PNG should therefore consider reintroducing such a single PQ dose in its national treatment guidelines.

Primaquine in Papua New Guinea and the Melanesian Western Pacific

To date, several molecular studies to characterize G6PD deficiency have been conducted in PNG (21,22), Solomon Islands (23) and Vanuatu (24). There are also reports of population-based screening of G6PD deficiency in the Solomon Islands (25). However, epidemiological screening of markers of G6PD deficiency in any given population is of only limited value (12) if it does not allow an estimation of the prevalence of clinically relevant phenotypes (allelic mutations associated with haemolysis) present at the individual level. Studies are therefore needed to identify the allelic mutations in Melanesian populations that are associated with clinically significant risks of severe/life-threatening haemolysis. The absence of such evidence presents a major obstacle for the implementation of a PQ treatment policy and the use of PQ in mass drug administration for malaria elimination in *P. vivax*-endemic countries such as PNG. For these reasons, the National Malaria Control Program Strategic Plan (26) clearly states the need to test for G6PD deficiency at the hospital level and for adopting the use of only a low dose of PQ (0.25 mg/kg) as treatment for confirmed *P. vivax* cases in all health facilities until further information is available. However, a review of 18 studies published since 1950 (27) showed that the effectiveness of PQ ≤0.25 mg/kg was no different from that of no PQ treatment.

Efficacy data of *P. vivax* treatment and the contribution of hypnozoites to *P. vivax* infection

For a long time, the effective treatment for *P. vivax* was CQ. *P. vivax* resistance to CQ, first reported in 1989 from PNG (28), has later become widespread throughout the island of New Guinea (29,30), requiring new studies to reassess the effectiveness of the old and

new treatment regimens against relapses. Following reports of increasing resistance of *P. falciparum* and *P. vivax* against CQ and sulfadoxine-pyrimethamine (31), a trial of combination antimalarial therapies in children was carried out in Madang (32). The standard treatment for uncomplicated malaria in PNG was changed from AQ/CQ + SP to artemether-lumefantrine (AL) in 2009, in the wake of global trends to move towards artemisinin-based combination therapy (ACT). However, the efficacy of AL for preventing recurrent *P. vivax* infections was not substantially better than that of CQ + SP, with 87.0% and 69.7% of participants in the CQ + SP and the AL groups, respectively, showing recurrent *P. vivax* infections during 6 weeks of follow-up (p = 0.06) (32). Recently, genotyping of the same samples showed that most of the infections in the AL group were of different genotypes, suggesting new *P. vivax* infections rather than recrudescence of the initial infection from the same genotype observed at baseline (33).

The observations from these studies suggest that the new standard treatment of AL, while effective against acute clinical *P. vivax* malaria episodes, does not prevent late treatment failures (34) and therefore may have limited effect on the prevalence and transmission of *P. vivax.* Indeed, both new infections and/or relapses from hypnozoites in the liver allow *P. vivax* to re-establish blood stage infection very rapidly following treatment with AL, particularly because the half-life of lumefantrine (the only drug remaining in the bloodstream after the disappearance of the short-lived artemisinin component) is relatively short (4-5 days), and thus the post-treatment prophylactic effect conferred by the use of this drug combination is shorter than that of other combinations. As a consequence, the new PNG standard treatment protocol for confirmed (or suspected) *P. vivax* malaria adopted in 2009 includes the prescription of PQ at 0.25 mg/kg daily for 14 days after 3 days of AL (35).

Only recently, studies on PQ safety and tolerability and its effect on hypnozoites were performed in cohorts of PNG children aged 1 to 10 years old living in areas of high transmission and thus with high reinfection risk (36,37). The results show PQ to be safe and effective when used in combination with artesunate in G6PD-normal children. Pretreatment with artesunate plus PQ (14 days, 0.5 mg/kg) reduced the incidence of *P. vivax* malaria by 49% for the initial 3 months (p = 0.031) and 19% for months 4-9 (p = 0.25), and reduced the risk of *P. vivax* infections diagnosed by light microscopy and PCR (polymerase chain reaction) by 67% and 44%, respectively (p <0.001), when compared to a group treated only with artesunate (37). The effect of artesunate + PQ was limited to the first 3 months of follow-up and 30% of the children in the artesunate + PQ group had reinfection by 2 weeks of follow-up. Even though the artesunate + PQ combination may not be optimally efficacious in eradicating hypnozoites completely, most likely due to the short half-life of artesunate, PQ use had a significant impact in reducing the incidence and burden of relapse malarial disease when compared to artesunate alone.

Alternatives to primaquine for hypnozoiticidal therapy

Tafenoquine

Although the improved use of PQ could result in a significant improvement of current treatment of *P. vivax* malaria and reduce transmission of any *Plasmodium* spp., the need for G6PD testing as well as problems with adherence to the long 14-day treatment schedule are considerable obstacles to a large-scale rollout of PQ therapy. Consequently, there is a great need to develop alternative anti-hypnozoite drugs. One such novel drug is tafenoquine (38,39). As another 8-aminoquinoline it shares with PQ the problem of potential haemolysis in G6PD-deficient individuals; however, due to its long half-life, it can be given as a single dose or 3-day-long treatment and can thus potentially be combined with standard 3-day blood-stage regimens. Tafenoquine is currently undergoing phase II/III testing.

Research priorities

Even though tafenoquine will address the problem of poor compliance with current PQ regimens, additional anti-hypnozoite drugs that can be safely given in G6PD-deficient individuals are a high priority on the malaria elimination research agenda (40). It is also essential to carry out studies that may contribute to a better understanding of the role of relapses as sources of newly arising infections, and also in this context the potential efficacy and safety of PQ in preventing or delaying such relapses, particularly in children.

A safety, tolerability and pilot efficacy study of short-course, high-dose PQ treatment for *P. vivax* in children 5 to 10 years old is currently in progress in PNG, comparing the standard 14-day to a 7-day regimen. If shown to be safe, proceeding to a 3½-day dose regimen will be considered.

Health policy considerations for PNG

The clinical implementation of PQ alongside a blood-schizonticidal drug in primary health care settings is challenging. Uncertainties remain around the appropriate application of PQ in PNG, particularly in the presence of G6PD deficiency. Potentially as a consequence of ambiguous recommendations, the prescription of PQ to patients has been inconsistent in recent years (41).

Point-of-care testing for G6PD

The available methods of testing for G6PD deficiency, such as the Motulsky dye decolouration test (42), NADPH (nicotinamide adenine dinucleotide phosphate) fluorescent spot test (43,44) and variations of the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) formazan methods (45-48), require specialized equipment and have therefore not been successfully implemented in clinical settings in malaria-endemic areas (45,49). More recently, the United States Food and Drug Administration (FDA) approved the BinaxNOW® G6PD test (Inverness Medical, Switzerland) and the Dojindo G6PD WST-8 Assay Kit (Dojindo Molecular Technologies, Japan) (25,45,50), which have provided a more suitable alternative, though challenges to their implementation in malaria-endemic areas remain. These are qualitative diagnostic tests, dependent on visual interpretation of colour change within a specified time and temperature range. The optimal temperature for BinaxNOW® G6PD ranges from 18 to 25ºC. Both tests are temperature/light sensitive, rather expensive (approximately US$8 and US$5 per test, respectively), not stand-alone kits (ie, requiring additional equipment) and require a higher level of training to perform and interpret than the commonly utilized malaria rapid diagnostic tests (RDTs). An alternative would be the new G6PD test kit called CareStart™ (Access Bio, New Jersey, USA), which is still undergoing development. It was recently tested for the first time under field conditions to assess its performance (51). The CareStart™ is an RDT-format test which could be used together with current RDT testing for malaria diagnosis once it is fully developed and approved as a point-of-care, easy-to-use diagnostic tool for G6PD deficiency testing.

Until such challenges are overcome and routine G6PD screening is implemented at outpatient health services in PNG, *P. vivax* malaria and relapses from the dormant stages in the liver will remain a challenge for the PNG National Malaria Control Program as the low-dose PQ treatment recommended in the absence of G6PD testing is unlikely to be effective against circulating vivax strains present in PNG (5,27).

The hypnozoites in the liver represent an important source of reinfection, disease and transmission of *P. vivax*. In order for malaria prevention and control programs to be effective, treatment options for eradication of the liver stages of *P. vivax* must be evaluated and implemented together with other control measures such as ACTs and insecticide-impregnated bednets.

Recommendations

For elimination of malaria to become an eventual reality, all confirmed cases of malaria, including *P. falciparum* monoinfections, will need to be treated with hypnozoiticidal doses of PQ 0.5 mg/kg or higher. A single dose of 0.75 mg/kg PQ that was included in the PNG national treatment guidelines before 2000 should be reintroduced as routine gametocytocidal treatment for clinical cases of *P. falciparum* and the treatment policy should include PQ for asymptomatic infections of all plasmodium species. Asymptomatic carriers of all human *Plasmodium* spp. contribute to disease, transmission and development of resistance to antimalarial drugs (52). Mass drug administration for malaria control and elimination must include PQ (53) with an ACT such as dihydroartemisinin-piperaquine (54) that has a partner drug with a long terminal elimination phase (55). Finally, knowledge of the distribution of G6PD deficiency variants throughout PNG and the Melanesian Pacific that are associated with clinically significant risk of severe haemolytic anaemia, and a point-of-care, easy-to-use diagnostic test for G6PD deficiency will be needed to eliminate malaria.

REFERENCES

1 **Gething PW, Elyazar IRF, Moyes CL, Smith DL, Battle KE, Guerra CA, Patil AP, Tatem AJ, Howes RE, Myers MF, George DB, Horby P, Wertheim HFL, Price RN, Mueller I, Baird JK, Hay SI.** A long neglected world malaria map: *Plasmodium vivax* endemicity in 2010. *PLoS Negl Trop Dis* 2012;6:e1814.

2 **Chaves SS, Rodrigues LC.** An initial examination of the epidemiology of malaria in the state of Roraima, in the Brazilian Amazon basin. *Rev Inst Med Trop Sao Paulo* 2000;42:269-275.

3 **Luxemburger C, Thwai KL, White NJ, Webster HK, Kyle DE, Maelankirri L, Chongsuphajaisiddhi T, Nosten F.** The epidemiology of malaria in a Karen population on the western border of Thailand. *Trans R Soc Trop Med Hyg* 1996;90:105-111.

4 **White NJ.** Determinants of relapse periodicity in *Plasmodium vivax* malaria. *Malar J* 2011;10:297.

5 **Baird JK, Hoffman SL.** Primaquine therapy for malaria. *Clin Infect Dis* 2004;39:1336-1345.

6 **Mueller I, Zimmerman PA, Reeder JC.** *Plasmodium malariae* and *Plasmodium ovale* – the "bashful" malaria parasites. *Trends Parasitol* 2007;23:278-283.

7 **Gallien S, Taieb F, Schlemmer F, Lagrange-Xelot M, Atlan A, Sarfati C, Molina JM.** Failure of atovaquone/proguanil to prevent *Plasmodium ovale* malaria in traveler returning from Cameroon. *Travel Med Infect Dis* 2008;6:128-129.

8 **Carson PE.** 8-Aminoquinolines. In: Peters W, Richards WHG, eds. Antimalarial Drugs II. Handbook of Experimental Pharmacology, Part 68/2. Berlin: Springer-Verlag, 1984:83-121.

9 **Sinton JA, Bird W.** Studies in malaria, with special reference to treatment. IX. Plasmoquine in the treatment of malaria. *Indian J Med Res* 1928;16:159-177.

10 **Clyde DF, McCarthy VE.** Radical cure of Chesson strain vivax malaria in man by 7, not 14, days of treatment with primaquine. *Am J Trop Med Hyg* 1977;26:562-563.

11 **Ehrman FC, Ellias JM, Young MD.** *Plasmodium vivax* Chesson strain. *Science* 1945;101:377.

12 **Cappellini MD, Fiorelli G.** Glucose-6-phosphate dehydrogenase deficiency. *Lancet* 2008;371:64-74.

13 **Alving AS, Arnald J, Hockwald RS, Clayman CB, Dern RJ, Beutler E, Flanagan CL.** Potentiation of the curative action of primaquine in vivax malaria by quinine and chloroquine. *J Lab Clin Med* 1955;46:301-306.

14 **Alving AS, Johnson CF, Tarlov AR, Brewer GJ, Kellermeyer RW, Carson PE.** Mitigation of the haemolytic effect of primaquine and enhancement of its action against exoerythrocytic forms of the Chesson strain of *Plasmodium vivax* by intermittent regimens of drug administration: a preliminary report. *Bull World Health Organ* 1960;22:621-631.

15 **Krishna S, White NJ.** Pharmacokinetics of quinine, chloroquine and amodiaquine. Clinical implications. *Clin Pharmacokinet* 1996;30:263-299.

16 **White NJ.** Primaquine to prevent transmission of falciparum malaria. *Lancet Infect Dis* 2013;13:175-181.

17 **White NJ.** The role of anti-malarial drugs in eliminating malaria. *Malar J* 2008;7(Suppl1):S8.

18 **Peatey CL, Leroy D, Gardiner DL, Trenholme KR.** Anti-malarial drugs: how effective are they against *Plasmodium falciparum* gametocytes? *Malar J* 2012;11:34.

19 **Bousema T, Drakeley C.** Epidemiology and infectivity of *Plasmodium falciparum* and *Plasmodium vivax* gametocytes in relation to malaria control and elimination. *Clin Microbiol Rev* 2011;24:377-410.

20 **World Health Organization.** Updated WHO policy recommendation: Single dose primaquine as a gametocytocide in *Plasmodium falciparum* malaria. Geneva: World Health Organization, 2012.

21 **Chockkalingam K, Board PG, Nurse GT.** Glucose-6-phosphate dehydrogenase deficiency in Papua New Guinea. The description of 13 new variants. *Hum Genet* 1982;60:189-192.

22 **Yenchitsomanus P, Summers KM, Chockkalingam C, Board PG.** Characterization of G6PD deficiency and thalassaemia in Papua New Guinea. *PNG Med J* 1986;29:53-58.

23 **Hirono A, Ishii A, Kere N, Fujii H, Hirono K, Miwa S.** Molecular analysis of glucose-6-phosphate dehydrogenase variants in the Solomon Islands. *Am J Hum Genet* 1995;56:1243-1245.

24 **Ganczakowski M, Town M, Bowden DK, Vulliamy TJ, Kaneko A, Clegg JB, Weatherall DJ, Luzzatto L.** Multiple glucose-6-phosphate dehydrogenase-deficient variants correlate with malaria endemicity in the Vanuatu archipelago (southwestern Pacific). *Am J Hum Genet* 1995;56:294-301.

25 **Kuwahata M, Wijesinghe R, Ho MF, Pelecanos A, Bobogare A, Landry L, Bugora H, Vallely A, McCarthy J.** Population screening for glucose-6-phosphate dehydrogenase deficiencies in Isabel Province, Solomon Islands, using a modified enzyme assay on filter paper dried bloodspots. *Malar J* 2010;9:223.

26 **Papua New Guinea Department of Health.** National Malaria Control Program Strategic Plan 2009-2013. Port Moresby: Department of Health, 2009:25.

27 **John GK, Douglas NM, von Seidlein L, Nosten F, Baird JK, White NJ, Price RN.** Primaquine radical cure of *Plasmodium vivax*: a critical review of the literature. *Malar J* 2012;11:280.

28 **Rieckmann KH, Davis DR, Hutton DC.** *Plasmodium vivax* resistant to chloroquine? *Lancet* 1989;2:1183-1184.

29 **Price RN, Douglas NM, Anstey NM.** New developments in *Plasmodium vivax* malaria: severe disease and the rise of chloroquine resistance. *Curr Opin Infect Dis* 2009;22:430-435.

30 **Karyana M, Burdarm L, Yeung S, Kenangalem E, Wariker N, Maristela R, Umana KG, Vemuri R, Okoseray MJ, Penttinen PM, Ebsworth P, Sugiarto P, Anstey NM, Tjitra E, Price RN.** Malaria morbidity in Papua Indonesia, an area with multidrug resistant *Plasmodium vivax* and *Plasmodium falciparum*. *Malar J* 2008;7:148.

31 **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;77:947-954.

32 **Karunajeewa HA, Mueller I, Senn M, Lin E, Law I, Gomorrai PS, Oa O, Griffin S, Kotab K, Suano P, Tarongka N, Ura A, Lautu D, Page-Sharp M, Wong R, Salman S, Siba P, Ilett KF, Davis TME.** A trial of combination antimalarial therapies in children from Papua New Guinea. *N Engl J Med* 2008;359:2545-2557.

33 **Barnadas C, Koepfli C, Karunajeewa HA, Siba PM, Davis TME, Mueller I.** Characterization of treatment failure in efficacy trials of drugs against *Plasmodium vivax* by genotyping neutral and drug resistance-

associated markers. *Antimicrob Agents Chemother* 2011;55:4479-4481.

34 **Senn N, Rarau P, Manong D, Salib M, Siba P, Reeder JC, Rogerson SJ, Genton B, Mueller I.** Effectiveness of artemether/lumefantrine for the treatment of uncomplicated *Plasmodium vivax* and *P. falciparum* malaria in young children in Papua New Guinea. *Clin Infect Dis* 2013;56:1413-1420.

35 **Papua New Guinea Department of Health.** National Malaria Treatment Policy. Port Moresby: Department of Health, Sep 2009:28.

36 **Betuela I, Bassat Q, Kiniboro B, Robinson LJ, Rosanas-Urgell A, Stanisic D, Siba PM, Alonso PL, Mueller I.** Tolerability and safety of primaquine in Papua New Guinean children 1 to 10 years of age. *Antimicrob Agents Chemother* 2012;56:2146-2149.

37 **Betuela I, Rosanas-Urgell A, Kiniboro B, Stanisic DI, Samol L, de Lazzari E, Del Portillo HA, Siba P, Alonso PL, Bassat Q, Mueller I.** Relapses contribute significantly to the risk of *Plasmodium vivax* infection and disease in Papua New Guinean children 1-5 years of age. *J Infect Dis* 2012;206:1771-1780.

38 **Fernando D, Rodrigo C, Rajapakse S.** Primaquine in vivax malaria: an update and review on management issues. *Malar J* 2011;12:351.

39 **Elmes NJ, Nasveld PE, Kitchener SJ, Kocisko DA, Edstein MD.** The efficacy and tolerability of three different regimens of tafenoquine versus primaquine for post-exposure prophylaxis of *Plasmodium vivax* malaria in the Southwest Pacific. *Trans R Soc Trop Med Hyg* 2008;102:1095-1101.

40 **malERA Consultative Group on Drugs.** A research agenda for malaria eradication: drugs. *PLoS Med* 2011;8:e1000402.

41 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

42 **Bernstein RE.** A rapid screening dye test for the detection of glucose-6-phosphate dehydrogenase deficiency in red cells. *Nature* 1962;194:192-193.

43 **Beutler E.** A series of new screening procedures for pyruvate kinase deficiency, glucose-6-phosphate dehydrogenase deficiency and glutathione reductase deficiency. *Blood* 1966;28:553-562.

44 **Beutler E, Mitchell M.** Special modifications of the fluorescent screening method for glucose-6-phosphate dehydrogenase deficiency. *Blood* 1968;32:816-818.

45 **Tantular IS, Kawamoto F.** An improved, simple screening method for detection of glucose-6-phosphate deficiency. *Trop Med Int Health* 2003;8:569-574.

46 **Iwai K, Matsuoka H, Kawamoto F, Arai M, Yoshida S, Hirai M, Ishii A.** A rapid single-step screening method for glucose-6-phosphate dehydrogenase deficiency in field applications. *Jpn J Trop Med Hyg* 2003;31:93-97.

47 **Fairbanks VF, Lampe LT.** A tetrazolium-linked cytochemical method for estimation of glucose-6-phosphate dehydrogenase activity in individual erythrocytes: applications in the study of heterozygotes for glucose-6-phosphate dehydrogenase deficiency. *Blood* 1968;31:589-603.

48 **Padilla C, Nishiyama K, Shirakawa T, Matsuo M; Newborn Screening Study Group.** Screening for glucose-6-phosphate dehydrogenase deficiency using a modified formazan method: a pilot study on Filipino male newborns. *Pediatr Int* 2003;45:10-15.

49 **Jalloh A, Tantular IS, Pusarawati S, Kawilarang AP, Kerong H, Lin K, Ferreira MU, Matsuoka H, Arai M, Kita K, Kawamoto F.** Rapid epidemiologic assessment of glucose-6-phosphate dehydrogenase deficiency in malaria-endemic areas in Southeast Asia using a novel diagnostic kit. *Trop Med Int Health* 2004;9:615-623.

50 **Tinley KE, Loughlin AM, Jepson A, Barnett ED.** Evaluation of a rapid qualitative enzyme chromatographic test for glucose-6-phosphate dehydrogenase deficiency. *Am J Trop Med Hyg* 2010;82:210-214.

51 **Kim S, Nguon C, Guillard B, Duong S, Chy S, Sum S, Nhem S, Bouchier C, Tichit M, Christophel E, Taylor WR, Baird JK, Menard D.** Performance of the CareStart™ G6PD deficiency screening test, a point-of-care diagnostic for primaquine therapy screening. *PLoS One* 2011;6:e28357.

52 **Laishram DD, Sutton PL, Nanda N, Sharma VL, Sobti RC, Carlton JM, Joshi H.** The complexities of malaria disease manifestations with a focus on asymptomatic malaria. *Malar J* 2012;11:29.

53 **Shanks GD.** Control and elimination of *Plasmodium vivax*. *Adv Parasitol* 2012;80:301-341.

54 **Lwin KM, Phyo AP, Tarning J, Hanpithakpong W, Ashley EA, Lee SJ, Cheah P, Singhasivanon P, White NJ, Lindegårdh N, Nosten F.** Randomized, double-blind, placebo-controlled trial of monthly versus bimonthly dihydroartemisinin-piperaquine chemoprevention in adults at high risk of malaria. *Antimicrob Agents Chemother* 2012;56:1571-1577.

55 **Karunajeewa HA, Ilett KF, Mueller I, Siba P, Law I, Page-Sharp M, Lin E, Lammey J, Batty KT, Davis TME.** Pharmacokinetics and efficacy of piperaquine and chloroquine in Melanesian children with uncomplicated malaria. *Antimicrob Agents Chemother* 2008;52:237-243.

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How molecular epidemiology studies can support the National Malaria Control Program in Papua New Guinea

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SUMMARY

Papua New Guinea (PNG) is undertaking intensified efforts to control malaria. The National Malaria Control Program aims to reduce the burden of disease by large-scale distribution of insecticide-treated bednets, improved diagnosis and implementation of new treatments. A scientific program monitoring the effect of these interventions, including molecular epidemiology studies, closely accompanies the program. Laboratory assays have been developed in (or transferred to) PNG to measure prevalence of infection and intensity of transmission as well as potential resistance to currently used drugs. These assays help to assess the impact of the National Malaria Control Program, and they reveal a much clearer picture of malaria epidemiology in PNG. In addition, analysis of the geographical clustering of parasites aids in selecting areas where intensified control will be most successful. This paper gives an overview of current research and recently completed studies in the molecular epidemiology of malaria conducted in Papua New Guinea.

Introduction

The National Malaria Control Program has adopted a comprehensive strategy to reduce the burden of malaria in Papua New Guinea (PNG) through distribution of insecticide-treated nets, enhanced diagnosis using rapid diagnostic tests (RDTs) and deploying new, effective treatments such as artemisinin combination therapy (ACT) (1). Measuring the impact of these interventions is complex and requires several malaria indicators. Basic indicators include the incidence of disease (number of new malaria cases in a defined period of time), the prevalence of infections (proportion of the community infected with malaria parasites, including asymptomatic carriers) and the intensity of transmission. It is also critical to monitor the emergence of drug resistance country-wide and over time.

Over the past 30 years, the Papua New Guinea Institute of Medical Research (PNGIMR), in close collaboration with research institutes in different countries, has developed profound expertise to conduct malaria research (2,3). Numerous cohorts have been designed to monitor parasite prevalence and incidence (4-6), to assess efficacy of drugs and to validate novel treatment strategies such as intermittent preventive treatment in infants (IPTi) (7) or using ACT as new first-line treatment for malaria infections (8). Laboratory analyses have complemented clinical observations in the field. Molecular assays have been developed which detect the prevalence of various malaria parasite species simultaneously (9-11), estimate the number of parasite strains concurrently infecting a human host (12-15) and assess the frequency of mutations associated with

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antimalarial drug resistance (16,17).

This paper provides a brief overview of the molecular tools available to support the PNG National Malaria Control Program in assessing the impact of malaria control interventions and gives examples of findings from recently completed studies.

Diagnosis of malaria infections

There are currently three distinct methods to diagnose malaria infections. These are based on direct visualization of parasites in a blood smear (microscopy), detection of parasite antigens circulating in the blood stream (RDT) and detection of parasite deoxyribonucleic acid (DNA) by molecular assays.

For routine diagnosis in the field, light microscopy or RDT remain the methods of choice. These diagnostic methods provide sufficiently high levels of sensitivity for the current management of *P. falciparum* and *P. vivax* clinical cases in children in PNG (18).

However, once successful malaria control will have drastically reduced malaria prevalence, a high proportion of asymptomatic infections with low parasitaemia is expected as many individuals in the population will remain immune or semi-immune to malaria (19). These parasite carriers potentially contribute to transmission and therefore it will be important to perform active surveillance of low-density infections. Microscopy or RDT will no longer be the most suitable methods to detect infected individuals. It is commonly known that detection of malaria parasites in blood smears by light microscopy misses a substantial proportion of infected samples. In particular, infections with low parasitaemia and mixed infections are underdiagnosed. Similarly, RDT's performance decreases for low parasitaemia infections, in particular in detecting *P. vivax* (20). A recent review of studies conducted in several countries, including PNG, revealed that between 20 and 80% of all infected blood samples (49% on average in PNG) were misclassified as infection negative by standard microscopic diagnosis (21). In this epidemiological setting, polymerase chain reaction (PCR)-based methods can be used, such as ligation detection reaction-fluorescent microsphere assay (LDR-FMA) (9) or quantitative PCR (qPCR). These assays will improve the sensitivity of the diagnosis and, when using qPCR, provide an estimate of the number of parasites in the sample (11). Detection of as low as 1-10 parasites per microlitre of blood can be achieved. The improved sensitivity of malaria diagnosis using molecular techniques has already been demonstrated in PNG samples (Table 1). These molecular assays have been adapted to high-throughput in appropriately equipped laboratories such as those set up at PNGIMR. They are currently being used to investigate the proportion of submicroscopic infections in country-wide surveys to complement the assessment of the impact of the ongoing control strategies. The established capacity for molecular diagnosis in-country will be essential to strengthen the National Malaria Control Program once transmission is reduced substantially.

Genotyping malaria parasites: identifying individual parasite clones

Individuals are often infected with genetically different parasites of the same species, known as 'clones'. Multiclone infections occur either due to repeated bites of infective mosquitoes or multiple clones in a single mosquito inoculum. Knowledge of the number of clones infecting a human host at a particular time point or over a period of time can give insight into changes in transmission. Several molecular assays are available for genotyping parasites, ie, to distinguish individual parasite clones. They are based on highly polymorphic genetic markers (12-14,24). For *P. falciparum* and *P. vivax* the genetic diversity was found to be very high in PNG, ie, a large number of genetically distinct variants were detected (13,14). With currently available markers, the probability that two unrelated parasites carry the same alleles ranges from less than 1% to 8%. Genotyping thus allows confident assumptions to be made about (i) whether two humans are infected with the same parasite clone, and (ii) whether different blood samples collected from the same individual at different time points contain the same or different parasite clones.

Measuring the multiplicity of infection and the force of infection

By applying parasite genotyping to a cohort study conducted in Ilaita (East Sepik Province, PNG) (5), it was shown that despite a similar prevalence of *P. falciparum* and *P. vivax* infections (50% vs 53%), the multiplicity of infection (MOI) – the number of parasite

clones per human host – differed substantially: on average, *P. vivax* infections contained 2.7 clones, while *P. falciparum* infections contained only 1.5 clones (25).

The number of clones present within a human host depends on factors such as naturally acquired immunity against malaria, transmission intensity and the amount of exposure to malaria treatment. For *P. vivax,* relapses (reactivation of latent liver stages) also contribute to new infections detected in the bloodstream. Therefore, as a measurement of transmission, the number of new infections acquired over time, known as the molecular force of blood-stage infection (molFOB), represents a more specific indicator than MOI at a single time point.

MOI and molFOB analyses are not feasible by microscopy, since these measures are based on molecular differences amongst morphologically identical clones of the same species. Study participants can remain infection-positive for many months, and it is impossible to determine based on microscopy whether a single parasite clone is persisting or whether new infections have replaced those previously detected. Genotyping of the nearly 5000 blood samples collected in the above-mentioned Ilaita study revealed a complex pattern of infections over time, with interesting differences between *P. falciparum* and *P. vivax*. Most importantly, despite a comparable prevalence of the two species (5), individuals acquired around 15 *P. vivax* infections per year (26) compared to only 6 *P. falciparum* infections (27). For *P. falciparum*, the use of a bednet dramatically reduced the number of infections and as a consequence the number of episodes. For *P. vivax,* bednet use led to a reduction in infections but not to a reduction in clinical episodes (26). Given that bednet use and number of infections are highly correlated, molFOB can be used as a surrogate marker to measure the impact of bednet distribution and other vector control interventions in research studies.

Distinguishing between treatment failure and new infections in drug trials

The efficacy of malaria treatment is critical for reducing morbidity and mortality. However, resistance to commonly used drugs is a major concern in the global efforts to control malaria. Drug efficacy trials, with genotyping correction, are therefore regularly conducted in PNG, and are part of the evaluation of the National Malaria Control Program, to assess whether the antimalarial drugs in use will successfully clear parasites from infected hosts (1).

In these trials, individuals are regularly checked for recurring infections after drug administration (28). If *P. falciparum* parasites reappear during follow-up this could be because of a persistence of the original infection (ie, recrudescence of the same clone) or it could be due to a newly acquired infection (ie, a different clone). Only the first situation, ie, recrudescence, constitutes a true treatment failure. Given the very high diversity of *Plasmodium* populations in PNG, it is highly unlikely that a new infection would carry the same allele of the genotyping marker as the one at baseline before drug administration (14). The World Health Organization (WHO) therefore recommends using PCR-corrected treatment failure rates as a primary endpoint for *P. falciparum* treatment efficacy trials as this provides a more accurate assessment of drug efficacy (28).

As an example, in 2005, the efficacy of four treatments against *P. falciparum* malaria (chloroquine plus sulfadoxine-pyrimethamine (CQ-SP), artesunate plus SP (ART-SP), dihydroartemisinin-piperaquine (DHA-PIP) and artemether-lumefantrine (AL)) was assessed in PNG (8). Before correction of treatment failure rates for new infections by genotyping, no significant difference between treatments was observed by day 42 (observed treatment failure rates ranging from 31.1% to 37.4%). However, after correction by genotyping, the AL treatment group showed the lowest rate of treatment failure (4.8% vs 12%, 14.6% and 18.4%, for DHA-PIP, ART-SP and CQ-SP respectively). This led to the adoption of AL as first-line treatment for all malaria cases in PNG.

For *P. vivax,* the situation is complicated by relapsing hypnozoites from the liver that cause additional blood-stage infections. Nevertheless, genotyping is possible in *P. vivax* drug trials that assess efficacy of drugs targeting blood stages (such as ACT, SP and CQ). In high transmission areas, relapses are usually of a different genotype from the last blood-stage infection (29), which results in them being classified as new infection and not as treatment failure. Efficacy estimates before and after genotyping can differ substantially. For example, in the above-cited study in PNG,

TABLE 1

Comparison of the diagnostic sensitivity of light microscopy and PCR for *Plasmodium* spp. in malaria surveys in PNG

PNG study site	Type of survey	Sample size	Age of study participants	Number of malaria infections (prevalence %)				Reference
				Plasmodium species	Light microscopy	PCR-LDR-FMA	qPCR	
Wosera, East Sepik	Cross-sectional	1182	All ages	Pf	244 (20.6)	389 (32.9)	-	(22)
				Pv	150 (12.7)	320 (27.1)	-	
				Pm	47 (4)	147 (12.4)	-	
				Po	3 (0.25)	65 (5.5)	-	
				Mixed infection	28 (2.4)	199 (16.8)	-	
Maprik and Wosera-Gawi, East Sepik	Cross-sectional	2527	All ages	Pf	756 (29.9)	1387 (54.9)	-	(23)
				Pv	368 (14.6)	902 (35.7)	-	
				Pm	99 (3.9)	339 (13.4)	-	
				Po	0	121 (4.8)	-	
Maprik, East Sepik	Cohort	264	0.9-4.5 years	Pf	86 (32.6)	131 (49.6)	-	(5)
				Pv	117 (44.3)	140 (53.0)	-	
				Pm	11 (4.2)	26 (9.8)	-	
				Po	0	7 (2.7)	-	
				Mixed infection	45 (17.0)	85 (32.2)	-	
Maprik, East Sepik	Cohort	452	1-5 years	Pf	115 (25.4)	213 (47.1)	185 (40.9)	(11)
				Pv	248 (54.9)	305 (67.5)	297 (65.7)	

Pm	11 (2.4)	35 (7.7)	21 (4.6)
Po	0	20 (4.4)	33 (7.3)

PCR = polymerase chain reaction
PNG = Papua New Guinea
LDR = ligation detection reaction
FMA = fluorescent microsphere assay
qPCR = quantitative polymerase chain reaction
Pf = *Plasmodium falciparum*
Pv = *Plasmodium vivax*
Pm = *Plasmodium malariae*
Po = *Plasmodium ovale*

out of 33 children positive for *P. vivax*, 13 were parasite positive 28 days after treatment with AL (30). However, genotyping revealed that only 6 of these 13 children were infected with the same parasite clone as before treatment, and in the remaining 7 cases a new infection or a relapse (different clone) was found. In summary, genotyping demonstrated that the drug was much more efficient (only 6 treatment failures in 33 children) than it appeared to be when only the parasite positivity at day 28 was measured (13 treatment failures). Figure 1 shows an example of genotyping results of a treatment failure versus new infection.

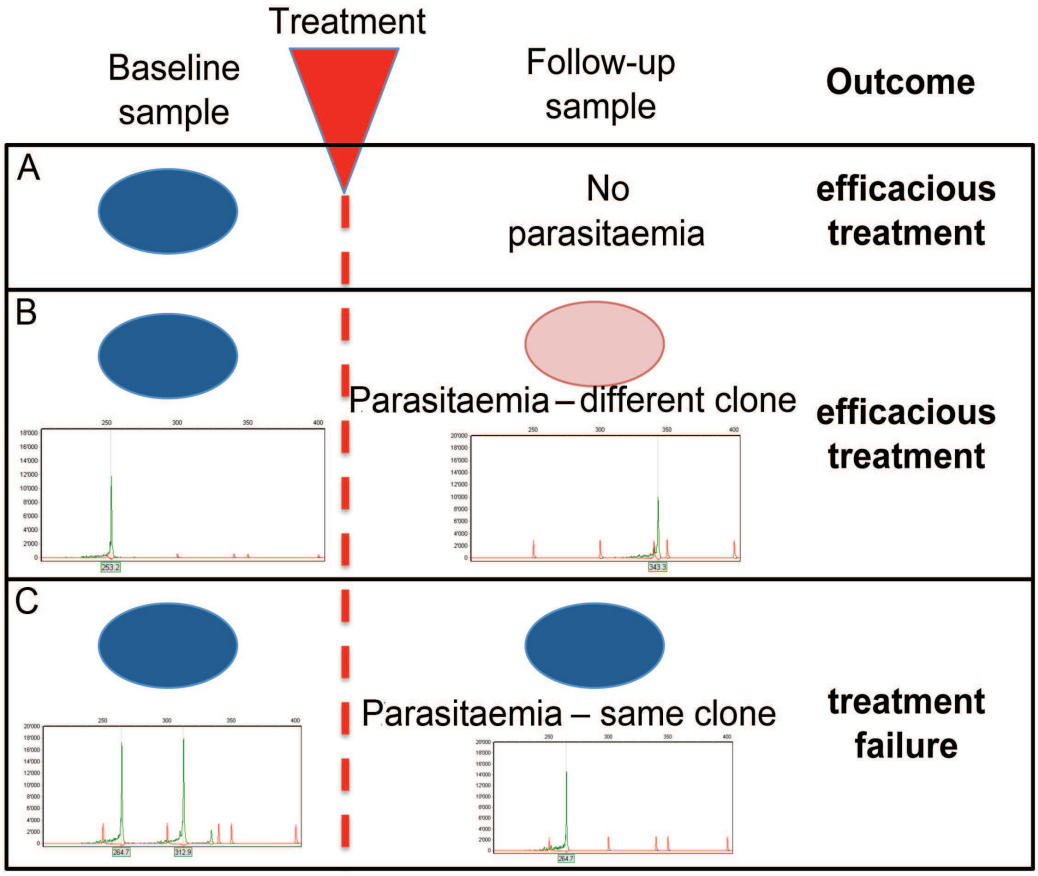

Figure 1. Molecular genotyping to distinguish between recrudescence and new infection in drug efficacy trials.

Two examples showing genotyping results of the *P. vivax* marker msp1F3 in a drug efficacy trial. A size-polymorphic DNA fragment was amplified by polymerase chain reaction (PCR) and the length of the fragments was measured by capillary electrophoresis.

X-axis: Length of the DNA fragment in base pairs

Y-axis: Relative Fluorescence Units, indicating the amount of amplified DNA

Patient A (top): No parasite detected during follow-up.

Patient B (middle): The parasite detected at baseline is of a different genotype from the parasite detected at day 28. Thus the drug was efficient in clearing the parasite present at day 0, and the parasite present at day 28 is a new infection.

Patient C (bottom): Two clones are detected at day 0 (multiplicity of infection = 2). At day 28, one of the clones (264 base pairs in length) is still present. This clone has survived drug treatment, thus this is a treatment failure.

Once transmission in PNG begins to decrease substantially as a result of the National Malaria Control Program, re-evaluation of the genotyping markers will be needed, as some alleles may reach high frequencies and thus make it difficult to distinguish between distinct clones. In addition, *P. vivax* relapse patterns might change, with relapses becoming less frequent and often more similar to the previous blood-stage infection. While these changes will be assessed in a research setting, recommendations on the choice of adequate genotyping markers will be provided to the National Malaria Control Program.

Molecular monitoring of drug resistance

Resistance of malaria parasites to commonly used antimalarial drugs such as CQ and SP has emerged and spread quickly after deployment in PNG. Both *P. falciparum* and *P. vivax* CQ resistance emerged in PNG in the mid-1970s (31) and late 1980s (32), respectively.

While in vivo efficacy trials remain the standard method to assess drug efficacy, genotyping parasites for mutations associated with drug resistance represents a valuable tool to monitor changes in parasite populations following the implementation of new antimalarial treatment strategies. For both *P. falciparum* and *P. vivax*, exposure to SP can result in the selection of mutant parasites in the dihydrofolate reductase (*dhfr*) and dihydropteroate synthase (*dhps*) genes. While *P. falciparum* 4-aminoquinoline (CQ and amodiaquine (AQ)) resistance has been associated with the presence of mutations in the chloroquine resistance transporter gene (*crt*) and the multidrug resistance 1 gene (*mdr1*), only the latter has been used as a marker of *P. vivax* resistance. The association between drug pressure and selection of mutant parasites has been effectively demonstrated in the PNG context by analysing drug usage at the health centre level and the prevalence of mutations in *P. falciparum* parasites over a 12-year period (33). The prevalence of *pfdhfr* double mutant parasites increased by a factor of 2.5 in the 3 years following adoption of SP as the first-line treatment of malaria.

A high-throughput molecular method (16,17) was recently used by PNGIMR to assess the prevalence of *P. falciparum* and *P. vivax* molecular markers of resistance in East Sepik and Madang Provinces in 2006 (CB, unpublished data). Data showed that the prevalence of mutations involved in SP and 4-aminoquinoline resistance was significantly higher in Madang than in East Sepik Province. For example, the quintuple mutant *P. vivax* parasite (quadruple mutant *pvdhfr,* single mutant *pvmdr1*), previously associated with AQ + SP treatment failure in PNG (34), was 4 times more frequent in Madang Province than in East Sepik (43.2% vs 10.5%, χ^2 test $p < 0.001$). This result correlates with efficacy trial results, which show variable rates of treatment failure in the same two provinces and illustrates the utility of molecular markers to detect early signs of drug resistance, as well as geographical heterogeneity among parasite populations (35).

PNGIMR is currently monitoring changes in drug resistance mutation frequencies and the appearance of new genotypes across PNG in the context of the National Malaria Control Program by genotyping *P. falciparum* and *P. vivax* isolates collected country-wide. A first survey was conducted in 2008-2009, which is 4 years after the introduction of intensified and sustained malaria control measures but before the implementation of a new treatment protocol (36,37). Interestingly, the removal of CQ and AQ may lead to the re-emergence of drug-sensitive parasites (38-40), because wild-type parasites remained in the community at low prevalence despite widespread drug use. The re-emergence of drug-sensitive parasites is thought to occur because drug-associated mutations may impact on parasite fitness (ie, the resistant parasite is less efficiently propagated and transmitted in the absence of drug pressure) (38). In order to maintain effective antimalarial regimens, it is therefore important to monitor drug efficacy of the classical antimalarials CQ, AQ and SP (which remain widely available (37) and are still the drugs of choice in PNG for prevention of malaria in pregnancy) in addition to new first- and second-line antimalarial treatments.

AL is the current first-line treatment in PNG. To date, no molecular markers are known for resistance to artemisinin derivatives, which has been characterized as a delayed parasite clearance following treatment (41). Given that artemisinin should always be administered together with a partner drug (lumefantrine in PNG) for the treatment of uncomplicated malaria cases it is difficult to detect resistance in vivo, in particular if the partner drug remains highly effective. Numerous research groups

around the world are at present trying to identify artemisinin resistance markers by comparing whole genome sequencing results of drug-sensitive and drug-resistant isolates (42,43). Once identified and validated, these markers would represent a major improvement for surveillance of the emergence and spread of artemisinin resistance (44,45). Genotyping data could then be used by policy makers to request new efficacy trials to confirm early detection of resistance, or reversal to classical antimalarial sensitivity.

Mapping parasite population structure and tracking outbreaks

Genotyping malaria parasites using panels of microsatellites (length polymorphic markers) (46-48) or point mutations (single nucleotide polymorphisms, SNPs) (49,50) can define relationships amongst populations from different geographic areas. If parasite populations are closely related, this suggests that they are mixing due to movement of infected hosts (eg, via a major transport route). On the other hand, genetic differences between parasite populations from different areas suggest limited movement between populations and possibly that these populations are isolated from each other. Such isolated populations are important targets for malaria control since the risk of reintroduction would be reduced.

Studies carried out using microsatellites have shown that *P. falciparum* populations across the north coast of PNG are structured into fragmented subpopulations (51) while *P. vivax* populations may be mixing to a greater extent (52). Currently, SNP markers are being developed in a research project specifically to distinguish between parasite populations in PNG. They will be used to genotype and understand genetic relationships amongst samples collected as part of country-wide malaria surveys. In addition, genotyping of parasites from cases reporting to clinics in areas where malaria is normally absent is expected to reveal key insights into the geographic origin(s) of such infections and to determine whether malaria transmission is local or imported (53). Once a good understanding of parasite genotypes across PNG and neighbouring countries is available, comparing genotypes of imported infections to the database should identify their principal geographic origin. Depending on whether cases are imported or transmitted locally, different reactions are needed: stopping local transmission might require enhanced vector control or mass drug administration, while imported cases could lead to screening at local and/or national borders.

In countries where transmission has been reduced to very low levels, population structure and diversity show distinct patterns in space and time, tracking control activities and informing future elimination strategies. A study from Djibouti (Horn of Africa) showed declining parasite diversity over a period of 11 years, consistent with declining transmission, and in addition it suggested that most remaining cases were imported from neighbouring countries (54). In the Comoros Archipelago, genetic distances between parasites from several islands were small, while parasites from one island exhibited a distinct genetic composition (55). Likewise in Malaysia, *P. falciparum* populations showed a partly fragmented structure with some populations clearly separated from others, suggesting that elimination is feasible in these isolated foci of transmission (56). The recently completed surveys in PNG can serve as a baseline for future studies. Given its island setting and limited transport within the country, population structure analysis will be useful to select areas where intensified control will be most successful and to assess the risk of reintroduction once transmission decreases.

Conclusions and Outlook

PNGIMR has established the laboratory capacity to process large quantities of blood samples collected in the field. The amount of blood required to perform molecular analysis is minimal; DNA extraction can be performed from blood collected by finger prick that is then taken to the PNGIMR laboratories in Madang and Goroka. From the viewpoint of a malaria control program, knowledge of the prevalence of drug resistance or of the number of infections acquired over time can be an important guide to adjust antimalarial interventions.

Ongoing studies will shed further light on the epidemiology of malaria in PNG. The number of infections per year is currently being measured in different cohorts before and after implementation of intensified malaria control, allowing a better understanding of the effects of malaria control on transmission.

The efficacy of the current first-line drug AL is being monitored in in vivo efficacy trials, including correction by genotyping. Further molecular tools are being developed to map parasite populations and track the origins of outbreaks and imported infections.

More recently, a focus was placed on transmission from the human host to the mosquito vector. Do all infected humans transmit the parasite to the mosquito? Is there a difference between the parasite density in the human host and transmission, or between different *Plasmodium* species? Assays to detect gametocytes (the sexual form of the malaria parasite that the mosquito takes up during a blood meal) have now been established in PNG and will help answer these questions.

Some of the assays discussed in this paper have been integrated into the evaluation of the National Malaria Control Program. Parasite detection by PCR is performed on blood samples collected during regular surveys and will provide a more accurate estimate of parasite prevalence in the country. Other parameters, such as assessing molFOB, require sophisticated sampling schemes that go far beyond sample collection through the National Malaria Control Program. Also, sample collection for RNA extraction and subsequent gametocyte detection requires special procedures. Furthermore, completion of all laboratory assays and data analysis can take several years. It is therefore critical to define together with the National Malaria Control Program the molecular indicators that should be provided to the Program, and to ensure that results are reported back to policy makers in a timely and relevant manner.

Clearly, reports of clinical cases from clinics and microscopy readings in the field will remain the first source to assess the epidemiological situation in the country. In the long run, and especially once transmission in PNG drops to a low level, molecular tools can play a crucial role in understanding where transmission occurs and in designing new control strategies.

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REFERENCES

1 **Hetzel MW, Pulford J, Maraga S, Barnadas C, Reimer LJ, Tavul L, Jamea-Maiasa S, Tandrapah T, Maalsen A, Makita L, Siba PM, Mueller I.** Evaluation of the Global Fund-supported National Malaria Control Program in Papua New Guinea, 2009-2014. *PNG Med J* 2014;57:7-29.

2 **Mueller I, Genton B, Betuela I, Alpers MP.** Vaccines against malaria: perspectives from Papua New Guinea. *Hum Vaccin* 2010;6:17-20.

3 **Cortés A, Felger I, Beck HP.** Molecular parasitology of malaria in Papua New Guinea. *Trends Parasitol* 2003;19:246-249.

4 **Betuela I, Rosanas-Urgell A, Kiniboro B, Stanisic DI, Samol L, de Lazzari E, Del Portillo HA, Siba P, Alonso PL, Bassat Q, Mueller I.** Relapses contribute significantly to the risk of *Plasmodium vivax* infection and disease in Papua New Guinean children 1-5 years of age. *J Infect Dis* 2012;206:1771-1780.

5 **Lin E, Kiniboro B, Gray L, Dobbie S, Robinson L, Laumaea A, Schöpflin S, Stanisic D, Betuela I, Blood-Zikursh M, Siba P, Felger I, Schofield L, Zimmerman P, Mueller I.** Differential patterns of infection and disease with *P. falciparum* and *P. vivax* in young Papua New Guinean children. *PLoS One* 2010;5:e9047.

6 **Michon P, Cole-Tobian JL, Dabod E, Schoepflin S, Iga J, Susapu M, Tarongka N, Zimmerman PA, Reeder JC, Beeson JG, Schofield L, King CL, Mueller I.** The risk of malarial infections and disease in Papua New Guinean children. *Am J Trop Med Hyg* 2007;76:997-1008.

7 **Senn N, Rarau P, Stanisic DI, Robinson L, Barnadas C, Manong D, Salib M, Iga J, Tarongka N, Ley S, Rosanas-Urgell A, Aponte JJ, Zimmerman PA, Beeson JG, Schofield L, Siba P, Rogerson SJ, Reeder JC, Mueller I.** Intermittent preventive treatment for malaria in Papua New Guinean infants exposed to *Plasmodium falciparum* and *P. vivax*: a randomized controlled trial. *PLoS Med* 2012;9:e1001195.

8 **Karunajeewa HA, Mueller I, Senn M, Lin E, Law I, Gomorrai PS, Oa O, Griffin S, Kotab K, Suano P, Tarongka N, Ura A, Lautu D, Page-Sharp M, Wong R, Salman S, Siba P, Ilett KF, Davis TME.** A trial of combination antimalarial therapies in children from Papua New Guinea. *N Engl J Med* 2008;359:2545-2557.

9 **McNamara DT, Kasehagen LJ, Grimberg BT, Cole-Tobian J, Collins WE, Zimmerman PA.** Diagnosing infection levels of four human malaria parasite species by a polymerase chain reaction/ligase detection reaction fluorescent microsphere-based assay. *Am J Trop Med Hyg* 2006;74:413-421.

10 **McNamara DT, Thomson JM, Kasehagen LJ, Zimmerman PA.** Development of a multiplex PCR-ligase detection reaction assay for diagnosis of infection by the four parasite species causing malaria in humans. *J Clin Microbiol* 2004;42:2403-2410.

11 **Rosanas-Urgell A, Mueller D, Betuela I, Barnadas C, Iga J, Zimmerman PA, del Portillo HA, Siba**

P, Mueller I, Felger I. Comparison of diagnostic methods for the detection and quantification of the four sympatric *Plasmodium* species in field samples from Papua New Guinea. *Malar J* 2010;9:361.

12 **Falk N, Maire N, Sama W, Owusu-Agyei S, Smith T, Beck HP, Felger I.** Comparison of PCR-RFLP and Genescan-based genotyping for analyzing infection dynamics of *Plasmodium falciparum*. *Am J Trop Med Hyg* 2006;74:944-950.

13 **Koepfli C, Ross A, Kiniboro B, Smith TA, Zimmerman PA, Siba P, Mueller I, Felger I.** Multiplicity and diversity of *Plasmodium vivax* infections in a highly endemic region in Papua New Guinea. *PLoS Negl Trop Dis* 2011;5:e1424.

14 **Schoepflin S, Valsangiacomo F, Lin E, Kiniboro B, Mueller I, Felger I.** Comparison of *Plasmodium falciparum* allelic frequency distribution in different endemic settings by high-resolution genotyping. *Malar J* 2009;8:250.

15 **Bruce MC, Donnelly CA, Packer M, Lagog M, Gibson N, Narara A, Walliker D, Alpers MP, Day KP.** Age- and species-specific duration of infection in asymptomatic malaria infections in Papua New Guinea. *Parasitology* 2000;121(Pt 3):247-256.

16 **Barnadas C, Kent D, Timinao L, Iga J, Gray LR, Siba P, Mueller I, Thomas PJ, Zimmerman PA.** A new high-throughput method for simultaneous detection of drug resistance associated mutations in *Plasmodium vivax dhfr*, *dhps* and *mdr1* genes. *Malar J* 2011;10:282.

17 **Carnevale EP, Kouri D, DaRe JT, McNamara DT, Mueller I, Zimmerman PA.** A multiplex ligase detection reaction-fluorescent microsphere assay for simultaneous detection of single nucleotide polymorphisms associated with *Plasmodium falciparum* drug resistance. *J Clin Microbiol* 2007;45:752-761.

18 **Senn N, Rarau P, Manong D, Salib M, Siba P, Robinson LJ, Reeder J, Rogerson S, Mueller I, Genton B.** Rapid diagnostic test-based management of malaria: an effectiveness study in Papua New Guinean infants with *Plasmodium falciparum* and *Plasmodium vivax* malaria. *Clin Infect Dis* 2012;54:644-651.

19 **malERA Consultative Group on Monitoring, Evaluation and Surveillance.** A research agenda for malaria eradication: monitoring, evaluation, and surveillance. *PLoS Med* 2011;8:e1000400.

20 **Harris I, Sharrock WW, Bain LM, Gray KA, Bobogare A, Boaz L, Lilley K, Krause D, Vallely A, Johnson ML, Gatton ML, Shanks GD, Cheng Q.** A large proportion of asymptomatic *Plasmodium* infections with low and sub-microscopic parasite densities in the low transmission setting of Temotu Province, Solomon Islands: challenges for malaria diagnostics in an elimination setting. *Malar J* 2010;9:254.

21 **Okell LC, Bousema T, Griffin JT, Ouédraogo AL, Ghani AC, Drakeley CJ.** Factors determining the occurrence of submicroscopic malaria infections and their relevance for control. *Nat Commun* 2012;3:1237.

22 **Kasehagen LJ, Mueller I, McNamara DT, Bockarie MJ, Kiniboro B, Rare L, Lorry K, Kastens W, Reeder JC, Kazura JW, Zimmerman PA.** Changing patterns of *Plasmodium* blood-stage infections in the Wosera region of Papua New Guinea monitored by light microscopy and high throughput PCR diagnosis. *Am J Trop Med Hyg* 2006;75:588-596.

23 **Mueller I, Widmer S, Michel D, Maraga S, McNamara DT, Kiniboro B, Sie A, Smith TA, Zimmerman PA.** High sensitivity detection of *Plasmodium* species reveals positive correlations between infections of different species, shifts in age distribution and reduced local variation in Papua New Guinea. *Malar J* 2009;8:41.

24 **Koepfli C, Mueller I, Marfurt J, Goroti M, Sie A, Oa O, Genton B, Beck HP, Felger I.** Evaluation of *Plasmodium vivax* genotyping markers for molecular monitoring in clinical trials. *J Infect Dis* 2009;199:1074-1080.

25 **Koepfli C, Schoepflin S, Bretscher M, Lin E, Kiniboro B, Zimmerman PA, Siba P, Smith TA, Mueller I, Felger I.** How much remains undetected? Probability of molecular detection of human plasmodia in the field. *PLoS One* 2011;6:e19010.

26 **Koepfli C, Colborn KL, Kiniboro B, Lin E, Speed TP, Siba PM, Felger I, Mueller I.** A high force of *Plasmodium vivax* blood-stage infection drives the rapid acquisition of immunity in Papua New Guinean children. *PLoS Negl Trop Dis* 2013;7:e2403.

27 **Mueller I, Schoepflin S, Smith TA, Benton KL, Bretscher MT, Lin E, Kiniboro B, Zimmerman PA, Speed TP, Siba P, Felger I.** Force of infection is key to understanding the epidemiology of *Plasmodium falciparum* malaria in Papua New Guinean children. *Proc Natl Acad Sci USA* 2012;109:10030-10035.

28 **World Health Organization.** Methods for Surveillance of Antimalarial Drug Efficacy. Geneva: World Health Organization, 2009.

29 **Imwong M, Snounou G, Pukrittayakamee S, Tanomsing N, Kim JR, Nandy A, Guthmann JP, Nosten F, Carlton J, Looareesuwan S, Nair S, Sudimack D, Day NP, Anderson TJ, White NJ.** Relapses of *Plasmodium vivax* infection usually result from activation of heterologous hypnozoites. *J Infect Dis* 2007;195:927-933.

30 **Barnadas C, Koepfli C, Karunajeewa HA, Siba PM, Davis TME, Mueller I.** Characterization of treatment failure in efficacy trials of drugs against *Plasmodium vivax* by genotyping neutral and drug resistance-associated markers. *Antimicrob Agents Chemother* 2011;55:4479-4481.

31 **Yung AP, Bennett NM.** Chloroquine-resistant falciparum malaria in Papua New Guinea. *Med J Aust* 1976;2:320-321.

32 **Rieckmann KH, Davis DR, Hutton DC.** *Plasmodium vivax* resistant to chloroquine? *Lancet* 1989;2:1183-1184.

33 **Nsanzabana C, Hastings IM, Marfurt J, Müller I, Baea K, Rare L, Schapira A, Felger I, Betschart B, Smith TA, Beck HP, Genton B.** Quantifying the evolution and impact of antimalarial drug resistance: drug use, spread of resistance, and drug failure over a 12-year period in Papua New Guinea. *J Infect Dis* 2010;201:435-443.

34 **Marfurt J, de Monbrison F, Brega S, Barbollat L, Müller I, Sie A, Goroti M, Reeder JC, Beck HP, Picot S, Genton B.** Molecular markers of in vivo *Plasmodium vivax* resistance to amodiaquinine plus sulphadoxine-pyrimethamine: mutations in *pvdhfr* and *pvmdr1*. *J Infect Dis* 2008;198:409-417.

35 **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;77:947-954.

36 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.

37 **Pulford J, Mueller I, Siba PM, Hetzel MW.** Malaria case management in Papua New Guinea prior to the introduction of a revised treatment protocol. *Malar J* 2012;11:157.

38 **Kublin JG, Cortese JF, Njunju EM, Mukadam RA, Wirima JJ, Kazembe PN, Djimdé AA, Kouriba B, Taylor TE, Plowe CV.** Reemergence of chloroquine-sensitive *Plasmodium falciparum* malaria after cessation of chloroquine use in Malawi. *J Infect Dis* 2003;187:1870-1875.

39 **Mita T, Kaneko A, Lum JK, Bwijo B, Takechi M, Zungu IL, Tsukahara T, Tanabe K, Kobayakawa T, Björkman A.** Recovery of chloroquine sensitivity and low prevalence of the *Plasmodium falciparum* chloroquine resistance transporter gene mutation K76T following the discontinuance of chloroquine use in Malawi. *Am J Trop Med Hyg* 2003;68:413-415.

40 **Takechi M, Matsuo M, Ziba C, MacHeso A, Butao D, Zungu IL, Chakanika I, Bustos MD.** Therapeutic efficacy of sulphadoxine/pyrimethamine and susceptibility in vitro of *P. falciparum* isolates to sulphadoxine-pyrimethamine and other antimalarial drugs in Malawian children. *Trop Med Int Health* 2001;6:429-434.

41 **Dondorp AM, Nosten F, Yi P, Das D, Phyo AP, Tarning J, Lwin KM, Ariey F, Hanpithakpong W, Lee SJ, Ringwald P, Silamut K, Imwong M, Chotivanich K, Lim P, Herdman T, An SS, Yeung S, Singhasivanon P, Day NP, Lindegardh N, Socheat D, White NJ.** Artemisinin resistance in *Plasmodium falciparum* malaria. *N Engl J Med* 2009;361:455-467.

42 **Cheeseman IH, Miller BA, Nair S, Nkhoma S, Tan A, Tan JC, Al Saai S, Phyo AP, Moo CL, Lwin KM, McGready R, Ashley E, Imwong M, Stepniewska K, Yi P, Dondorp AM, Mayxay M, Newton PN, White NJ, Nosten F, Ferdig MT, Anderson TJ.** A major genome region underlying artemisinin resistance in malaria. *Science* 2012;336:79-82.

43 **Takala-Harrison S, Clark TG, Jacob CG, Cummings MP, Miotto O, Dondorp AM, Fukuda MM, Nosten F, Noedl H, Imwong M, Bethell D, Se Y, Lon C, Tyner SD, Saunders DL, Socheat D, Ariey F, Phyo AP, Starzengruber P, Fuehrer HP, Swoboda P, Stepniewska K, Flegg J, Arze C, Cerqueira GC, Silva JC, Ricklefs SM, Porcella SF, Stephens RM, Adams M, Kenefic LJ, Campino S, Auburn S, MacInnis B, Kwiatkowski DP, Su XZ, White NJ, Ringwald P, Plowe CV.** Genetic loci associated with delayed clearance of *Plasmodium falciparum* following artemisinin treatment in Southeast Asia. *Proc Natl Acad Sci USA* 2013;110:240-245.

44 **Wongsrichanalai C, Sibley CH.** Fighting drug-resistant *Plasmodium falciparum*: the challenge of artemisinin resistance. *Clin Microbiol Infect* 2013;19:908-916.

45 **World Health Organization.** Global Plan for Artemisinin Resistance Containment (GPARC). Geneva: World Health Organization, 2011.

46 **Anderson TJ, Haubold B, Williams JT, Estrada-Franco JG, Richardson L, Mollinedo R, Bockarie M, Mokili J, Mharakurwa S, French N, Whitworth J, Velez ID, Brockman AH, Nosten F, Ferreira MU, Day KP.** Microsatellite markers reveal a spectrum of population structures in the malaria parasite *Plasmodium falciparum*. *Mol Biol Evol* 2000;17:1467-1482.

47 **Imwong M, Sudimack D, Pukrittayakamee S, Osorio L, Carlton JM, Day NP, White NJ, Anderson TJ.** Microsatellite variation, repeat array length, and population history of *Plasmodium vivax*. *Mol Biol Evol* 2006;23:1016-1018.

48 **Karunaweera ND, Ferreira MU, Munasinghe A, Barnwell JW, Collins WE, King CL, Kawamoto F, Hartl DL, Wirth DF.** Extensive microsatellite diversity in the human malaria parasite *Plasmodium vivax*. *Gene* 2008;410:105-112.

49 **Mu J, Awadalla P, Duan J, McGee KM, Joy DA, McVean GA, Su XZ.** Recombination hotspots and population structure in *Plasmodium falciparum*. *PLoS Biol* 2005;3:e335.

50 **Neafsey DE, Schaffner SF, Volkman SK, Park D, Montgomery P, Milner DA Jr, Lukens A, Rosen D, Daniels R, Houde N, Cortese JF, Tyndall E, Gates C, Stange-Thomann N, Sarr O, Ndiaye D, Ndir O, Mboup S, Ferreira MU, Moraes Sdo L, Dash AP, Chitnis CE, Wiegand RC, Hartl DL, Birren BW, Lander ES, Sabeti PC, Wirth DF.** Genome-wide SNP genotyping highlights the role of natural selection in *Plasmodium falciparum* population divergence. *Genome Biol* 2008;9:R171.

51 **Schultz L, Wapling J, Mueller I, Ntsuke PO, Senn N, Nale J, Kiniboro B, Buckee CO, Tavul L, Siba PM, Reeder JC, Barry AE.** Multilocus haplotypes reveal variable levels of diversity and population structure of *Plasmodium falciparum* in Papua New Guinea, a region of intense perennial transmission. *Malar J* 2010;9:336.

52 **Koepfli C, Timinao L, Antao T, Barry AE, Siba P, Mueller I, Felger I.** A large *Plasmodium vivax* reservoir and little population structure in the South Pacific. *PLoS One* 2013;8:e66041.

53 **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.

54 **Khaireh BA, Assefa A, Guessod HH, Basco LK, Khaireh MA, Pascual A, Briolant S, Bouh SM, Farah IH, Ali HM, Abdi AI, Aden MO, Abdillahi Z, Ayeh SN, Darar HY, Koeck JL, Rogier C, Pradines B, Bogreau H.** Population genetics analysis during the elimination process of *Plasmodium falciparum* in Djibouti. *Malar J* 2013;12:201.

55 **Rebaudet S, Bogreau H, Silaï R, Lepere JF, Bertaux L, Pradines B, Delmont J, Gautret P, Parola P, Rogier C.** Genetic structure of *Plasmodium falciparum* and elimination of malaria, Comoros Archipelago. *Emerg Infect Dis* 2010;16:1686-1694.

56 **Anthony TG, Conway DJ, Cox-Singh J, Matusop A, Ratnam S, Shamsul S, Singh B.** Fragmented population structure of *Plasmodium falciparum* in a region of declining endemicity. *J Infect Dis* 2005;191:1558-1564.

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Long-lasting insecticidal nets remain efficacious after five years of use in Papua New Guinea

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SUMMARY

Long-lasting insecticide-treated nets (LLINs) have been distributed throughout Papua New Guinea since 2004 as part of the country's malaria control program. This study aimed to evaluate the efficacy of these used bednets over time and with respect to the various household factors related to their use in order to enable the National Department of Health to maximize on the benefits of LLINs. In 2008 and early 2009, used LLINs (0-9 years old) were collected in various villages in Papua New Guinea as part of the Global Fund to Fight AIDS, Tuberculosis and Malaria (GFATM)-supported National Malaria Control Program and data were collected on net usage. A subset of the nets were tested for residual insecticide content. Net efficacy was measured by the rate of knockdown of *Anopheles farauti* s.s. following exposure to LLINs using the World Health Organization cone bioassay. Optimal effectiveness (>95% knockdown 1 hour post exposure) was observed in 92% of the LLINs. A slight reduction in efficacy was observed after two years of household use and there was a significant relationship between the number of years in use and percent knockdown ($p < 0.001$) as well as deltamethrin concentration ($p < 0.001$). Washing of nets was not associated with a reduction in deltamethrin concentration, but drying them in the sun was ($p = 0.008$). The physical conditions of these nets also degraded over time with a significant increase in the number of large holes after 5 years ($p = 0.02$). These findings are in support of the current recommendation to replace LLINs after five years of use, and demonstrate that proper net care can extend the length of efficacy.

Introduction

Malaria is one of the leading diseases in the world that accounts for the majority of morbidity and mortality seen in over 90 countries (1,2). It is transmitted by *Anopheles* mosquitoes (3) and many malaria control campaigns have included the use of vector control to curb transmission of the disease (4).

Vector control programs have primarily focused on insecticide-based interventions: indoor residual spraying (IRS), insecticide-treated nets (ITNs) and, more recently, long-lasting insecticide-treated bednets (LLINs). ITNs offer personal and community protection against mosquito bites (5,6) and are widely accepted as an important malaria control tool (7). ITNs must be manually treated every 6 months with pyrethroid insecticides, the only insecticide class recommended by the World Health Organization (WHO) Pesticide Evaluation Scheme (WHOPES) for bednet impregnation due to their low mammalian

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toxicity and fast knockdown effect against mosquitoes (8,9). With LLINs, technological advances have enabled insecticides to be incorporated into the net fibres (3), allowing for slow and continuous movement of insecticides (10) from the fibre to the surface of the nets. Therefore, the introduction of LLINs has eliminated the need for retreatment, creating a practical and sustainable vector control tool.

Although LLINs remain effective for a longer period of time, they will require replacement when the mosquito knockdown effect is compromised. The current recommendation is to replace after 5 years of use, and many studies have been carried out to examine insecticide reduction and loss of LLIN efficacy over time (4,11-14). Many studies have found LLINs to be effective in the range of 2-5 years. Lindblade and others (11) found LLIN PermaNet 1.0 (Vestergaard Frandsen) to be effective for 2 years (82.2% mortality). PermaNet 2.0 in Uganda performed effectively after 3 years with 90% of the nets under observation resulting in a mortality rate of >80% (12). In Tanzania, LLINs (PermaNet 2.0) used for 5 years induced less mortality than did new nets sealed for the same duration (4). Efficacy was found to be significantly reduced after 5 years (4,9,13).

Under laboratory conditions, PermaNet 2.0 LLINs retained efficacy after 20 or more washes (6). In Colombia, high efficacy was also observed for PermaNet LLINs after 23 washes, having a lower limit for residual deltamethrin at 3 mg/m^2 and achieving at least 80% mortality in *Anopheles* mosquitoes (14). Insecticide loss and reduced LLIN efficacy have been attributed to differences in the use and wear of the nets in households, primarily the washing practices of users (3). The loss of insecticide could be attributed to the strong detergents used for washing and direct sunlight for drying (4,5,9,15-17) or other factors such as firewood smoke and dust particles (4,9,15) that interfere with exposure.

The presence of holes in LLINs also reduces their effectiveness (4,13,18). Large holes can enable a mosquito to easily enter a net, minimizing or eliminating insecticide contact thus compromising the protective efficacy of the bednets (3). There was a rapid increase in the proportion of nets with holes observed in Uganda after only a year of use (12). Kilian and others attributed the physical degradation of nets to the number of washes and socioeconomic status (12).

LLINs are employed throughout many areas of the world as part of malaria control programs. However, most LLIN efficacy studies have been performed in Africa (4,5,9,15,18) or Asia (6,19). In Papua New Guinea (PNG), the National Department of Health has been distributing LLINs throughout the country as part of the malaria control program since 2004 (20). The main malaria vectors are members of the *Anopheles punctulatus* group including *Anopheles farauti* s.s*, Anopheles hinesorum, Anopheles farauti* 4*, Anopheles koliensis* and *Anopheles punctulatus* (21). These vectors have been shown to be highly susceptible to pyrethroids, the insecticides used in LLINs (22); however, there have been no studies on LLIN efficacy in PNG after use in the home. Due to different geographical, environmental and socioeconomic factors present in PNG, previous studies may not be able to provide applicable information that can guide LLIN redistribution schedules. This study aimed to address these gaps in our knowledge of local LLIN efficacy and help inform the National Malaria Control Program.

Methods

Anopheles farauti s.s. colony

A colony of *An. farauti* s.s. was maintained in an insectary at the Papua New Guinea Institute of Medical Research in Madang, PNG. The colony was established from mosquitoes collected in Rabaul, East New Britain Province. Preliminary studies showed the colony to be equally susceptible to insecticides as local wild *An. farauti* s.s. The insectary was maintained at ambient temperature (25-28°C) and adult cages were covered with damp towels to attain 80% relative humidity (RH) throughout the day.

LLIN collection

Between October 2008 and August 2009, used PermaNet 2.0 LLINs (n = 83) were collected from 21 villages in 9 provinces – Western (Fly), Simbu, Eastern Highlands, Western Highlands, Morobe, Madang, East Sepik, West Sepik (Sandaun) and Central Province – in Papua New Guinea, in the frame of a country-wide household survey (20). Collected nets were replaced with new nets of the same brand. At the time of the collection, owners were asked to recall the age of the

nets, number of washings, whether they were washed in soap or water alone and whether they were dried in sun or shade. The nets were then sealed individually in resealable storage bags and stored in the dark until the bioassays were conducted.

Residual deltamethrin testing

The used bednets (n = 83) were cut in half and sent to PermaNet® manufacturer Vestergaard Frandsen Laboratories in Hanoi, Vietnam for residual insecticide analysis. Chemical analysis was done using X-ray fluorescence spectroscopy (XRF) to quantify deltamethrin concentration. Those nets with deltamethrin content under the quantification limit of XRF were tested again with high-performance liquid chromatography (HPLC).

WHO cone bioassay

Adult female mosquitoes aged 2 to 5 days were exposed to a subset of used LLINs (n = 51) that were quantified for deltamethrin concentration, using standard WHO cone bioassays (23). They were exposed at 25^{o}C (± 2^{o}C) and 70-80% relative humidity for three minutes and held in the insectary for sixty minutes with access to 10% sucrose-soaked cotton pads. 5 female mosquitoes were placed in each cone, with each net subjected to 10 cones distributed across the side and top of the net (50 mosquitoes per net). During each bioassay, 1 to 5 cones (5 mosquitoes each) were placed on an untreated net as a control. Knockdown was recorded at three minutes, ten minutes, thirty minutes and sixty minutes post exposure and functional mortality was recorded 24 hours post exposure. Mortality observed on untreated nets under 20% was then adjusted for using the Abbott's formula (23). If greater than 20% mortality was observed in the control mosquitoes, the exposure was repeated.

Physical condition observation

Physical condition of the nets was evaluated by estimating the average surface area lost to wear and tear. The number of holes on each net half was counted, and each hole placed in a size category determined by diameter: very small (<1 cm), small (1-5 cm), medium (5-15 cm), large (15-30 cm) and very large holes (>30 cm). The hole index was calculated by multiplying the number of holes in each category by the minimum surface area in cm^2 per hole for each category. The presence of smoke odour was qualitatively recorded as either no odour, faint, moderate or strong smoke odour. Two researchers independently scored each net and a third observer was included if the two observations were discordant.

Statistics

Since very high rates of knockdown were observed 1 hour post exposure, we also calculated time to 50% knockdown and used this to measure the impact of usage variables on net efficacy. This was done by fitting the knockdown data to the equation [y = a/ (1.0 + exp (-(x-b)/c))]. The relationships between net efficacy and usage variables were analysed by 2-sample t-test or single factor analysis of variance when there were 3 or more groups. GraphPad Prism 6.0b was used for statistical analyses and construction of graphs.

Results

Number of years in use

The ages of LLINs included in this study were 9 years (n = 1), 7 years (n = 3), 4 years (n = 7), 3 years (n = 9), 2 years (n = 5), 1 year (n = 23) and less than 1 year (n = 3). Of the 51 LLINs tested, 47 (92%) exhibited optimal effectiveness (>95% knockdown 1 hour post exposure) and all nets resulted in over 85% knockdown of exposed mosquitoes. However, the nets started showing reduced efficacy after 2 years of home use (Figure 1) and percent knockdown was significantly associated with the number of years in use ($p < 0.001$). Residual deltamethrin (DM) concentration ranged between 70 mg/m^2 and undetectable levels. There was a significant relationship between DM concentration and number of years in use ($p < 0.001$) (Figure 2).

Washing practices

The number of washes were very low (range: 0-6 washes), with a median of 2, 0, 1, 2, 1, 1, 0 washes for initial years of net use in 2009, 2008, 2007, 2006, 2005, 2002 and 2000 respectively, with over 90% of these nets washed three or fewer times. Median DM concentration between the washes was 49.2 mg/m^2, 18.6 mg/m^2, 11.55 mg/m^2, 19.55 mg/m^2, 20.6 mg/m^2 and 31.00 mg/m^2 for 0, 1, 2, 3, 4, 6 washes respectively. There was not a significant relationship between number of

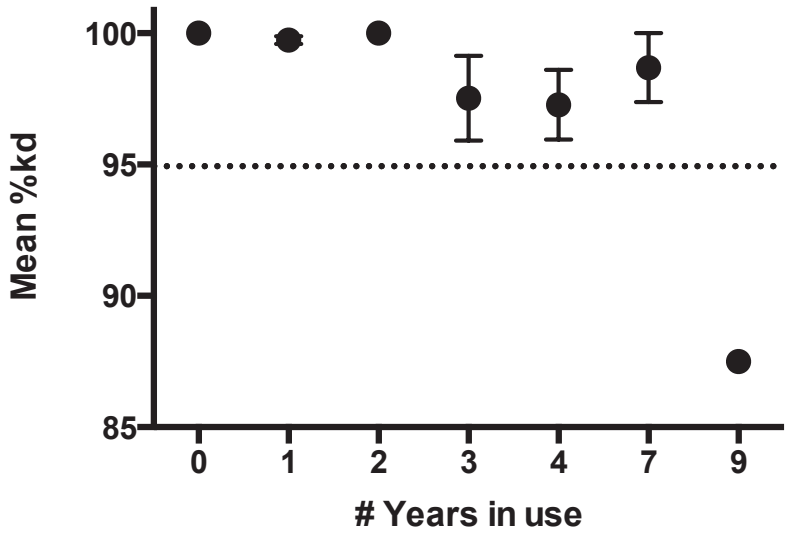

Figure 1. Mean percent knockdown in mosquitoes 60 minutes post exposure to LLINs that had been removed from household use after the given number of years. The dotted line represents the World Health Organization criterion for net efficacy. Error bars represent standard error of the mean.

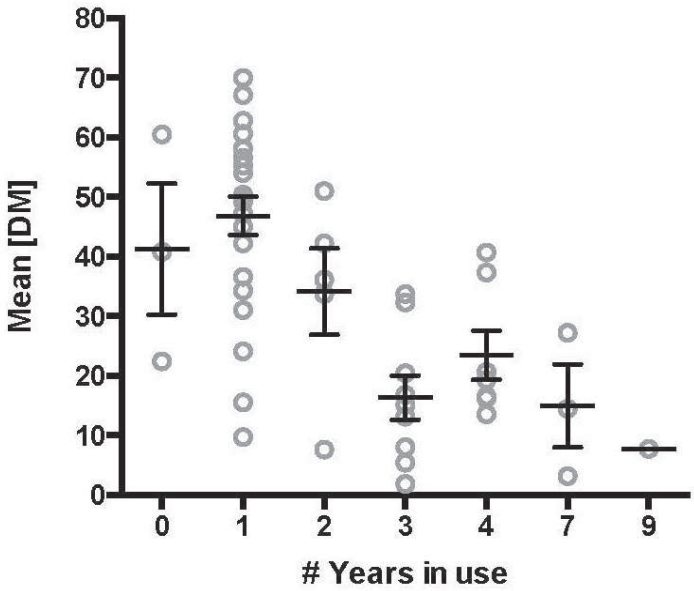

Figure 2. Mean deltamethrin concentration (mg/m^2) of long-lasting insecticidal nets (LLINs) after the given number of years of household use. Error bars represent standard error of the mean.

washings and DM concentration or percent knockdown. The majority of washed nets (84% of 25 nets) had been washed using soap. There was no significant relationship between whether soap was used in the washing and DM concentration or time to 50% knockdown.

Drying practices

The majority of washed nets (80% of 24 nets) were hung to dry in the sun and this practice was significantly correlated with low DM concentration (Figure 3, p = 0.008). The mean concentration in nets dried in the sun was 20.2 ±3.0 mg/m^2 compared to 36.3 ±7.9 mg/m^2 in the shade. There was not a significant relationship between drying in the sun and time to 50% knockdown.

Physical conditions and odour

The wear and tear of the nets (measured by hole surface area, Figure 4) was significantly correlated with number of years in use (p <0.001). In addition nets over 4 years old were more likely to have holes that were categorized as large or very large than nets under 5 years old (Table 1; p = 0.02), a condition that could greatly reduce net effectiveness. A total of 85% of nets up to 4 years old were classified as being in good condition with less than 50 cm^2 total hole surface area on half of the net (24). The presence of some smoke odour, an indicator of proximity to a cooking fire, was detected in 56% of the nets. However, there was no relationship between smoke odour and time to 50% knockdown or DM concentration.

Discussion

The WHO cone bioassay was used in this study to look at the survival rate of the mosquitoes after exposure to the LLINs. This study presents the proportion knockdown observed 1 hour post exposure, and no recovery was observed in knocked-down mosquitoes 24 hours post exposure. Since the majority of nets remained highly efficacious according to WHO standards, we also measured decreases in deltamethrin concentration as well as increases in time to 50% knockdown to determine if the usage variables had any relationship with efficacy. Results are presented as proportion or time to knockdown rather than mortality; however, we did not observe any recovery of knocked-down mosquitoes after 24 hours. One limitation of the study is that the age of the nets was self-reported and may be subject to recall bias. Additionally the age of nets may not directly correspond with the number of years of in-home use and may also include time that the

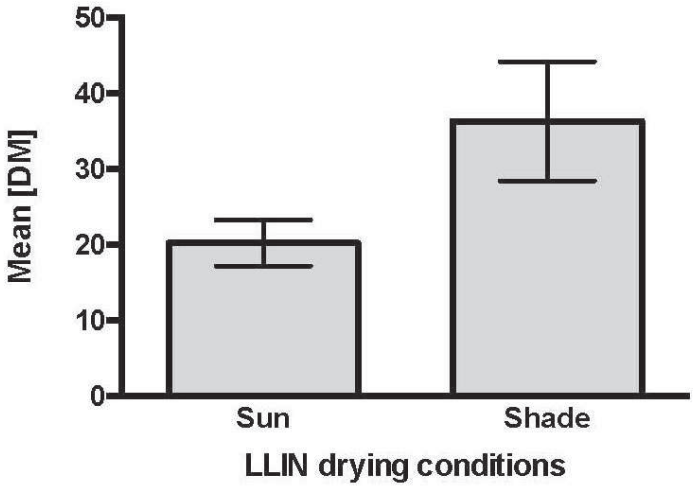

Figure 3. Mean deltamethrin concentration (mg/m^2) of washed long-lasting insecticidal nets (LLINs) that had been dried in the sun or shade (p = 0.008). Error bars represent standard error of the mean.

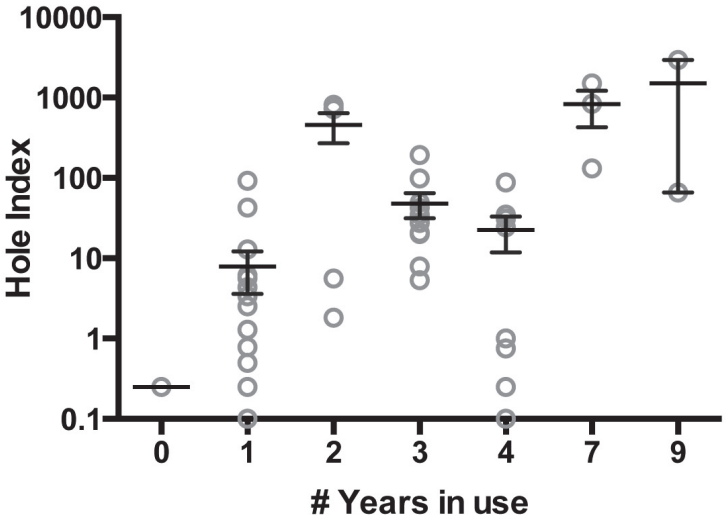

Figure 4. Mean hole index for long-lasting insecticidal nets (LLINs) (estimated cm^2 of compromised net surface on one half of the net) after the given number of years of household use. Error bars represent standard error of the mean.

TABLE 1

MEAN NUMBER AND SIZE OF HOLES IN LLINS COLLECTED AFTER THE GIVEN NUMBER OF YEARS OF HOUSEHOLD USE

Number of years in use	Very small (<1 cm)	Small (1-5 cm)	Medium (5-15 cm)	Large (15-30 cm)	Very large (>30 cm)
0	1.0	0.0	0.0	0.0	0.0
1	7.4	2.2	0.2	0.0	0.0
2	19.6	5.0	0.8	0.2	0.6
3	54.5	9.6	1.2	0.2	0.0
4	29.4	6.8	0.4	0.1	0.0
7	51.3	18.7	3.3	1.3	1.0
9	33.0	10.0	3.5	0.5	2.0

LLINs = long-lasting insecticidal nets

net remained in the package.

Mean percent knockdown was above the WHO criterion for net efficacy (>95%) through 7 years of home use; the 4 individual nets that exhibited slightly lower knockdown (85%<KD<95%) were over 3 years old. In order to preserve maximum deltamethrin residues, users may wash nets as needed but should always dry nets in a shaded area. Although high efficacy was seen in nets over 5 years old, a significant increase in wear and tear was observed in older nets, supporting the recommendation to replace LLINs every five years. This observation may not be a true representation of all nets in use and may have been influenced by owners preferring to donate nets in poor condition. Further studies on the durability of LLINs beyond five years and the relationship between wear and tear and risk of exposure are needed in order to provide comprehensive guidance to the National Malaria Control Program.

These encouraging results are dependent on the susceptibility of local vectors to pyrethroids. In this study the *An. farauti* colony was used in the bioassays in order to remove any individual variations in mosquito populations, allowing us to examine the relative impacts of usage on knockdown effect. Previous studies have shown members of the *An. punctulatus* group to be highly susceptible to pyrethroids (22) and our study observed no difference in knockdown rates between wild and colony *An. farauti*. However, future studies should continue to monitor insecticide resistance, especially as pyrethroid-based control efforts are scaling up, so that it can be mitigated before malaria control is compromised.

The protective effect of an LLIN distribution, and the optimum time for re-intervention, will be governed by entomological factors as well as insecticide degradation (25). LLINs have proven to be a very powerful tool in controlling malaria by reducing mosquito densities and by shortening the lifespan of mosquitoes (7) so that very few live through the extrinsic incubation period for *Plasmodium* spp. The utility of this control measure relies on the vector population entering human dwellings in search of a bloodmeal at the time the user is asleep, two conditions which are not always true in the South-West Pacific region (26,27). Continuous monitoring of mosquito-biting behaviours is also recommended to ensure that LLINs are effectively targeting and reducing the vector population. Otherwise it may be necessary to use an integrated approach to target early outdoor biters, such as larval site reduction, larvicides, odour-baited traps or spatial repellents (28).

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REFERENCES

1 **Hay SI, Guerra CA, Tatem AJ, Noor AM, Snow RW.** The global distribution and population at risk of malaria: past, present, and future. *Lancet Infect Dis* 2004;4:327-336.
2 **Ahmed SM, Zerihun A.** Possession and usage of insecticide bed nets among the people of Uganda: is BRAC Uganda Health Programme pursuing a pro-poor path? *PLoS One* 2010;5:e12660.
3 **Norris LC, Norris DE.** Efficacy of long-lasting insecticidal nets in use in Macha, Zambia, against the local *Anopheles arabiensis* population. *Malar J* 2011;10:254.
4 **Kweka EJ, Himeidan YE, Mahande AM, Mwang'onde BJ, Msangi S, Mahande MJ, Mazigo HD, Nyindo M.** Durability associated efficacy of long-lasting insecticidal nets after five years of household use. *Parasit Vectors* 2011;4:156.
5 **Guillet P, Alnwick D, Cham MK, Neira M, Zaim M, Heymann D, Mukelabai K.** Long-lasting treated mosquito nets: a breakthrough in malaria prevention. *Bull World Health Organ* 2001;79:998.
6 **Sreehari U, Mittal PK, Razdan RK, Ansari MA, Rizvi MM, Dash AP.** Efficacy of PermaNet 2.0 against *Anopheles culicifacies* and *Anopheles stephensi*, malaria vectors in India. *J Am Mosq Control Assoc* 2007;23:220-223.
7 **Takken W.** Do insecticide-treated bednets have an effect on malaria vectors? *Trop Med Int Health* 2002;7:1022-1030.
8 **World Health Organization.** Technical consultation on specifications and quality control of netting materials and mosquito nets. Geneva: World Health Organization, 2005.
9 **Atieli FK, Munga SO, Ofulla AV, Vulule JM.** The effect of repeated washing of long-lasting insecticide-treated net (LLINs) on the feeding success and survival rates of *Anopheles gambiae*. *Malar J* 2010;9:304.
10 **Vestergaard Frandsen PermaNet® 2.0 Brochure.** Long-lasting insecticidal net. Lausanne, Switzerland: Vestergaard Frandsen. www.vestergaard-frandsen.com/component/docman/cat_view51-product-brochures/61-permanetr/52-permanetr-

20?orderby=dmdate_publish&ascdesc=DESC
11 **Lindblade KA, Dotson E, Hawley WA, Bayoh N, Williamson J, Mount D, Olang G, Vulule J, Slutsker L, Gimnig J.** Evaluation of long-lasting insecticidal nets after 2 years of household use. *Trop Med Int Health* 2005;10:1141-1150.
12 **Kilian A, Byamukama W, Pigeon O, Atieli F, Duchon S, Phan C.** Long-term field performance of a polyester-based long-lasting insecticidal mosquito net in rural Uganda. *Malar J* 2008;7:49.
13 **Malima RC, Magesa SM, Tungu PK, Mwingira V, Magogo FS, Sudi W, Mosha FW, Curtis CF, Maxwell C, Rowland M.** An experimental hut evaluation of Olyset nets against anopheline mosquitoes after seven years use in Tanzanian villages. *Malar J* 2008;7:38.
14 **Kroeger A, Skovmand O, Phan QC, Boewono DT.** Combined field and laboratory evaluation of a long-term impregnated bednet, PermaNet. *Trans R Soc Trop Med Hyg* 2004;98:152-155.
15 **Atieli FK, Munga SO, Ofulla AV, Vulule JM.** Wash durability and optimal drying regimen of four brands of long-lasting insecticide-treated nets after repeated washing under tropical conditions. *Malar J* 2010;9:248.
16 **Green MD, Atieli F, Akogbeto M.** Rapid colorimetric test to determine levels of deltamethrin on PermaNet surfaces: association with mosquito bioactivity. *Trop Med Int Health* 2009;14:381-388.
17 **Skovmand O, Bosselmann R.** Strength of bed nets as a function of denier, knitting pattern, texturizing and polymer. *Malar J* 2011;10:87.
18 **Chandre F, Dabire RK, Hougard JM, Djobenou LS, Irish SR, Rowland M, N'guessan R.** Field efficacy of pyrethroid treated plastic sheeting (durable lining) in combination with long lasting insecticidal nets against malaria vectors. *Parasit Vectors* 2010;3:65.
19 **Tsuzuki A, Khamlome B, Kawada H, Eto H, Phompida S, Takagi M.** The efficacy and physical condition of Olyset insecticide-treated nets after 5 years use in rural Lao PDR. *Southeast Asian J Trop Med Public Health* 2011;42:268-273.
20 **Hetzel MW, Gideon G, Lote N, Makita L, Siba PM, Mueller I.** Ownership and usage of mosquito nets after four years of large-scale free distribution in Papua New Guinea. *Malar J* 2012;11:192.
21 **Cooper RD, Waterson DGE, Frances SP, Beebe NW, Pluess B, Sweeney AW.** Malaria vectors of Papua New Guinea. *Int J Parasitol* 2009;39:1495-1501.
22 **Keven JB, Henry-Halldin CN, Thomsen EK, Mueller I, Siba PM, Zimmerman PA, Reimer LJ.** Pyrethroid susceptibility in natural populations of the *Anopheles punctulatus* group (Diptera: Culicidae) in Papua New Guinea. *Am J Trop Med Hyg* 2010;83:1259-1261.
23 **World Health Organization.** Test procedures for insecticide resistance monitoring in malaria vectors, bio-efficacy and persistence of insecticides on treated surfaces. Geneva: World Health Organization, 1998.
24 **Kilian A, Byamukama W, Pigeon O, Gimnig J, Atieli F, Koekemoer L, Protopopoff N.** Evidence of a useful life of more than three years for a polyester-based long-lasting insecticidal mosquito net in Western Uganda. *Malar J* 2011;10:299.
25 **Briët OJ, Hardy D, Smith TA.** Importance of factors determining the effective lifetime of a mass, long-lasting, insecticidal net distribution: a sensitivity analysis. *Malar J* 2012;11:20.
26 **Taylor B.** Changes in feeding behavior of a malaria vector, *Anopheles farauti* Lav., following the use of DDT as a residual spray in houses in the British Solomon Islands Protectorate. *Trans R Entomol Soc Lond* 1975;127:277-292.
27 **Bugoro H, Cooper RD, Butafa C, Iro'ofa C, Mackenzie DO, Chen CC, Russell TL.** Bionomics of the malaria vector *Anopheles farauti* in Temotu Province, Solomon Islands: issues for malaria elimination. *Malar J* 2011;10:133.
28 **Govella NJ, Ferguson H.** Why use of interventions targeting outdoor biting mosquitoes will be necessary to achieve malaria elimination. *Front Physiol* 2012;3:199.

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TRIBUTE

Missing in the line of duty

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Papua New Guinea Institute of Medical Research, Goroka, Swiss Tropical and Public Health Institute and University of Basel, Basel, Switzerland and School of Population Health, University of Queensland, Brisbane, Australia

Introduction

The 1st of August 2011 will be remembered as a black day in the history of medical research in Papua New Guinea (PNG). On that day, five young staff members of the Papua New Guinea Institute of Medical Research (PNGIMR) disappeared while conducting a malaria survey in West New Britain (WNB) Province. Gibson Gideon, Tania Oakiva, Leonard Vavana, Lydia Petrus and George Dogoya, together with their boat crew, became the victims of criminals in a remote and difficult-to-access part of PNG. A coronial inquest concluded in April 2014 that the PNGIMR team had most likely perished in foul play, but that official investigations into the matter had been poor and insufficient (1). Until today, no traces of the PNGIMR team members' remains have been found. Families, friends and colleagues have to find consolation without really understanding their loved ones' disappearance or knowing that those responsible have been brought to justice.

This article summarizes the events leading up to and following the team's disappearance, recounted to the best of our knowledge. Despite being deeply affected by this tragic incident, we have made all attempts to report objectively.

Malaria survey in West New Britain

WNB was one of the last provinces to be covered in a national household survey carried out in 2011 to evaluate the PNG National Malaria Control Program (NMCP) (2). PNGIMR field research teams visited randomly selected villages across the country, interviewed household members about mosquito net ownership and use, and collected small samples of finger-prick blood to determine infection with malaria parasites back in the laboratory in Goroka (Figure 1). The team assigned to visit the five villages in WNB was experienced but also a little tired from long weeks in the field all across the country, completing thousands of questionnaires and preparing thousands of malaria blood slides.

The team was led by Gibson Gideon, a graduate scientific officer, who had joined the PNGIMR on 9th of June 2008 and had since gained extensive experience with malaria surveys. Gibson was the first staff member employed for the country-wide evaluation of the Global Fund-supported NMCP. While with PNGIMR, he obtained a Graduate Diploma in Science from the University of PNG in the frame of the Global Infectious Disease Research Training Program supported by the Fogarty International Centre. Leonard

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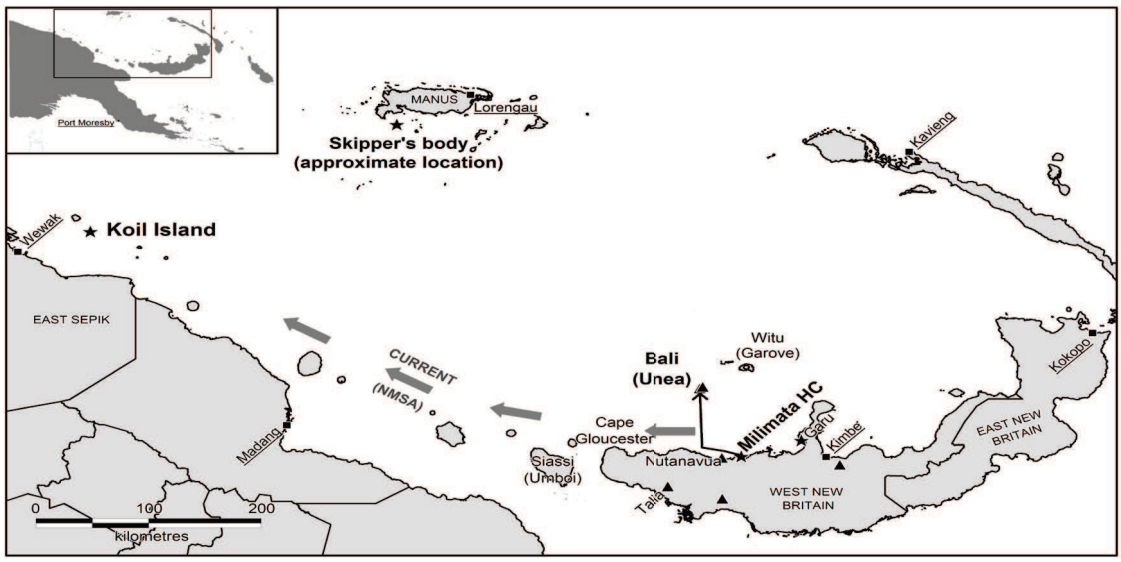

Figure 1. Map of West New Britain Province and randomly sampled survey villages.
HC = health centre
← = represents the planned travel route of the team's dinghy
▲ = indicates the randomly selected survey villages
★ = important locations mentioned in this paper
The likely direction of prevailing currents was provided by the National Maritime Safety Authority.

Vavana had worked with PNGIMR as a field research nurse from October 2008 to October 2009 and again from 10th of May 2010, being responsible for collecting blood samples for the diagnosis of malaria. Lydia Petrus previously worked as a casual field research nurse for a sexual health project and transferred to the malaria team on 21st of June 2010 to work alongside Leonard. George Dogoya joined the Institute on 10th of May 2010 as a research assistant conducting interviews with members of selected households. Tania Oakiva was employed on the 26th of October 2010 as a graduate scientific officer for an operational research study investigating reasons for non-use of mosquito nets, linked to the national household survey (3). WNB was scheduled to be the last trip for this team before the conclusion of the country-wide survey as the remaining two provinces would be covered by others.

The preparation of the survey in WNB had followed standard procedures: the team leader had informed the WNB Director Rural Health Services by fax about the details of the planned survey work and asked for local support from the provincial health office in organizing the team's logistics and informing the selected communities of the researchers' coming. As in other provinces, the team leader had also requested a provincial health officer to accompany them on the survey, and the health extension officer responsible for the area had been assigned to this task. As usual, the last fine-tuning of the schedule was done in the province once the team had arrived. In this case, travel by boat to reach one of the villages (Talia) was considered unsafe due to rough seas around Cape Gloucester. The team leader therefore agreed with us in the PNGIMR office in Goroka to survey Manopa, a randomly sampled second village on Bali (also Unea or Uneapa) Island instead. Such changes were relatively common (as conditions on the ground frequently and rapidly change), and the teams were used to not taking any risk if visiting a place was considered unsafe.

In Kimbe, the capital of WNB, Gibson's team met up with a group of PNGIMR colleagues who had travelled to the province a few weeks earlier to conduct a health facility survey under the umbrella of the same malaria control program evaluation. Both teams made use of a 23-foot dinghy named 'Bineve 2' that belonged to a member of the provincial

health office and that was skippered by an experienced crew originating themselves from Bali Island. The household survey team first visited the village of Nutanavua, which is, like most villages in the area, located on a small island only a few metres off-shore from the main island of New Britain. After completing this village, the group stayed overnight at nearby Milimata health centre, from where they left on Monday morning, the 1st of August of 2011, heading for Bali Island, less than 50 nautical miles off the coast to the North (Figure 1). The next couple of days were to be spent surveying the two sampled villages on the small island.

Accompanied by the skipper Peter Divu, his crew Bambon Pakela and Jerry Wani, a relative of Gibson's, they were travelling in the 'Bineve 2', the dinghy that carried the slogan 'Yamaha, Powering the Nation' on its sides and which was powered by a 60-horse-power outboard motor. It was equipped with standard safety gear such as paddles, bailing devices and an anchor, sufficient fuel was available in several containers, and the PNGIMR team was carrying the life jackets that the Institute had provided. The sea was said to be choppy that day, but nothing the local skippers wouldn't be used to on the two-hour trip to Bali.

Lost contact

On Friday afternoon, 5th of August 2011, a phone call from Kimbe reached the PNGIMR office in Goroka. The provincial health office reported that they had lost contact with the PNGIMR team since the day the team had left from Milimata. Without informing the PNGIMR or their superior, health officers had tried searching for the team by sending another boat along the same route, but no trace of the 'Bineve 2' was found. Attempts to contact the team members had failed and communities on Bali Island, once reached, could not confirm their arrival. The boat with the PNGIMR team had evidently never reached its intended destination.

With four days since the last contact, immediate action was required. In Goroka, the PNGIMR Director released funds for a search and rescue operation including aerial searches and two senior staff from Goroka travelled to Kimbe to support the operations. In Kimbe, Dr Joseph Nale, the Director Rural Health Services and Leo Mapmani, the Director of the Provincial Disaster Office (PDO) initiated the first response. The National Maritime Safety Authority (NMSA) and the National Disaster Centre (NDC) were notified the same day and the people in WNB were alerted through the local radio channel. NMSA issued an alert to vessels in the coastal waters of PNG to look out for the missing dinghy. The assumption at that time was that the boat had gone off course due to bad weather or engine problems and was either drifting with prevailing winds and currents, or had stranded somewhere on the shore.

On Saturday, a local helicopter was chartered to search for the missing dinghy around the Bali/Witu group of islands and along the coastline of the main island of West New Britain. At the same time, provincial health and police officers verified information at the boat's departure point. By Monday, 8th of August 2011, the gathered evidence pointed to two likely scenarios: either the dinghy had engine problems and was now drifting or had stranded somewhere, or the team had been ambushed and kidnapped. The possibility that the boat had capsized was considered unlikely: the sea had not been very rough, five other dinghies had arrived on Bali Island without difficulty on the same day, the boat was not carrying a heavy load, and no debris originating from the boat had been sighted. The conclusion of the search and rescue team on the ground was to intensify the search efforts and at the same time mobilize the police to investigate potential foul play.

Guided by advice from NMSA on prevailing currents, by local reports of suspicious sightings and taking into account details of previous boat mishaps, four additional aerial searches were carried out by fixed-wing aircraft and helicopter. The coastline and small islands up to Cape Gloucester and Siassi Island were searched by dinghy and engaging the provincial government's trawler MV Artemis. The National Department of Health (NDoH) informed health facilities along the coasts of neighbouring provinces to look out for the missing dinghy or any debris. Yet, unfortunately, all search efforts by land, sea and air remained unsuccessful (Table 1). No trace of the missing boat or its passengers could be found.

Involving a PNG Defence Force boat and aircraft through the NMSA was being considered, with plans to widen the search

TABLE 1

MAIN AERIAL AND SEA SEARCH OPERATIONS CONDUCTED IMMEDIATELY AFTER THE REPORTED DISAPPEARANCE OF THE BOAT

Date	Operation	Operator
6 Aug 2011	Bali/Witu Islands, Cape Gloucester/Bariai, coastline to Garu	Niugini Helicopters, Kimbe
7 Aug 2011	Around Bali Island	Private dinghies
8 Aug 2011	Hoskins-Lolobau-Point Takis (East New Britain)-Hoskins	New Tribes Mission Aviation, Hoskins
8 Aug 2011	Coastline Karaiai-Garu	Private dinghy
9 Aug 2011	Around Sakar and Siassi Islands – WNB coastline	North Coast Aviation, Lae
10 Aug 2011	Madang – Rai coast – Long Island – Bagabag – Karkar – North-East of Karkar approximately 60 nautical miles over open waters	Islands Airways, Madang
11-14 Aug 2011	Bali/Witu Islands – Cape Gloucester - Sakar/Siassi – Kimbe	MV Artemis
12 Aug 2011	Coastal rivers near Garu	Niugini Helicopters, Kimbe
12 Aug 2011	Coastal rivers near Garu	Dinghy with police, PDO

WNB = West New Britain
PDO = Provincial Disaster Office

to waters outside the New Britain areas, yet these plans never eventuated. In the meantime, officers from provincial police and PDO continued their investigations on land. Reports by local villagers were sketchy and sometimes contradictory but some warranted follow-up. Several teams were dispatched to follow leads about suspicious activity in coastal streams. Other information arrived from outside WNB, some of which was considered less plausible (such as a sighting of the dinghy by a foreign spy-plane including details of the health status of the boat occupants). About ten days after the reported disappearance of the dinghy, first reports of a pirate attack emerged. An informant knew of a hijacked vehicle that was allegedly used in preparation of a hold-up and another reported observing a suspicious dinghy with passengers allegedly in agony the evening of the 1st of August. In the early morning of Wednesday 3rd of August, two men reportedly sought treatment in Kimbe town for cuts originating from a knife fight. Informants from a village near Milimata reported sighting the missing boat on two occasions with two ladies on board. In order to facilitate the investigations, village elders from Kaliai-Kove were invited to Kimbe for confidential discussion with the PDO. They raised concerns over weather conditions on the day the team left but also confirmed knowledge of reports suggesting foul play. They assured their support of the investigations once back in their respective villages.

In the meantime, PNGIMR offered a reward of K5,000.00 through the PDO to anyone providing information that could lead to the safe return of the missing staff.

Then, on 24th of August 2011, a report was received of an almost completely submerged boat being recovered off Koil Island in the East Sepik Province. East Sepik Provincial Administration and PNGIMR staff confirmed the boat to be the missing 'Bineve 2' (Figure 2). The boat itself showed no damage and the

Figure 2. 23-foot dinghy 'Bineve 2' and its motor recovered near Koil Island, East Sepik Province. (Photographs provided by Kenny Rupa, Papua New Guinea Institute of Medical Research, Wewak)

position of the motor at the time of recovery indicated that it had been stopped before it sank. No additional evidence of the boat occupants or their equipment was found.

Subsequent reports of the PNGIMR staff members' equipment hidden at Nukakau village (near the team's departure point) led to the engagement of a police mobile squad from East New Britain. The mobile police team together with officers from Kimbe searched several villages and hideouts but no physical evidence of the missing people or their equipment was discovered. In consideration of the unsuccessful search, the Provincial Disaster Committee on the 8th of September 2011 suspended the search and declared the PNGIMR staff and their companions 'missing persons', in accordance with NMSA guidelines. This process was said to allow the police to take over and investigate the cause of the team's disappearance. It was also the time for PNGIMR to end its operational support on the ground in Kimbe and leave the investigations in the hands of police detectives. The search operations to that

date had relied almost exclusively on funds provided by PNGIMR. The Institute paid for ground, sea and aerial searches conducted by disaster and police officers. The PDO lacked adequate funds and equipment to conduct these search operations (4) and requests from the provincial police commander for funding support from the national government were allegedly still pending.

Police investigations

When the mandate was handed over to the police more than one month after the team's disappearance, the possibility of the boat having capsized at sea was considered unlikely and the possibility of the passengers drifting alive in open waters without their boat was unreasonable to maintain. Circumstantial evidence gathered during the search had much rather substantiated the suspicion of foul play. Whether the passengers had been killed or whether some of them might be held captive remained uncertain. Based on reports from informants and potential witnesses, several individuals needed to be further interrogated. This included people who had been in direct contact with the team before their departure to Bali Island. Conflicting statements and reported suspicious behaviour required detailed investigation.

While police now investigated independently, PNGIMR continued to face the pressure of family members of the missing staff who were demanding not only answers but also financial compensation. The Institute continued to provide support within its means but did not give up hope that staff and boat crew could still be found.

In the week of 19th September 2011, police exhumed a dead body that had been washed ashore on a small Manus island and was buried by villagers in mid-August. The partly decomposed corpse was identified by relatives as the skipper of the missing boat, but how could the skipper's body and the dinghy end up in so distant places? On 30th of September 2011, PNGIMR was informed by WNB police that a suspect was arrested and interrogations revealed the whereabouts of accomplices allegedly involved in hijacking the PNGIMR boat. The suspects were linked to William Kapris who had been arrested for a series of bank robberies. The arrested suspect also reported that he had recently seen one or two females belonging to the PNGIMR team in the company of his brother. With financial support from PNGIMR and New Britain Palm Oil Ltd, a police mobile squad raided a forest hideout in which firearms, live ammunition and other evidence of recent occupation were found (5). A 5-day ultimatum given to the local community to provide information of the whereabouts of the suspects and potentially any PNGIMR team members expired without new information emerging. Due to a lack of funding, police were unable to continue their investigations, even though at the time a breakthrough was considered to be just around the corner.

A story with no ending

Throughout the search operations, PNGIMR provided as much operational and financial support as possible to the search teams and the police. The incident was shocking for the families of the missing but it also deeply affected their supervisors and colleagues at the PNGIMR. Much of the research work came to a standstill while all efforts were made to support the search on the ground in WNB. Sadly, some local media provided a platform for certain individuals to air their anger and raise wild allegations against PNGIMR, which particularly affected those of us who had done all we could to keep staff safe and were now investing everything to find them (6,7). It required a lot of sensibility from the PNGIMR management to calm the families and assure them of the Institute's support. In February 2012, the Institute increased the award for any credible information about the missing staff to 30,000 Kina, hoping for a new impetus in the slowed-down investigations (Figure 3).

At the same time, the PNGIMR Director pleaded in a letter written to the then Minister for Health, Hon. Jamie Maxton-Graham, for the national government to finally provide the necessary funds that would enable a thorough police investigation. It had to be made clear that the Institute was no longer in a position to continue funding other state agencies to carry out their duties. The use of research donor funds to finance police operations would have had detrimental effects on the Institute's credibility. But it seemed that there was little movement despite promises from the government, and the provincial police commander of WNB remained without funding to continue the investigation (8).

It was in August 2013, two years after

Figure 3. Announcement of K30,000 reward published in local newspapers (Tok Pisin version).

the disappearance of the boat, that Prime Minister Peter O'Neill announced during a visit to PNGIMR in Goroka that a coronial inquiry would be funded by the government and be concluded before the end of the year (9). Yet before the inquiry started, new evidence suddenly emerged in November 2013 with the surrender of wanted criminal Don Aka, who was suspected to be behind the team's disappearance and was previously reported to be seen in company of the missing female PNGIMR staff. The surrender made headlines across PNG and beyond, including suggestions that the female PNGIMR staff might still be alive (10,11). The Institute renewed its commitment to provide the necessary funding for rescuing their staff members or retrieving their bodies should their whereabouts eventually be determined. The planned inquiry was subsequently postponed to allow further police investigations (12). However, the captured Don Aka did not provide the promised leads.

The ultimate breakthrough appeared to be reached in December 2013, when police investigators visited PNGIMR in Goroka with photographs of hard evidence seized during a dawn raid of a house in the Kimbe area. The pictures showed the laptop and voice recorder as well as paper survey forms carried by the missing PNGIMR team. According to the officers, the occupant of the house fled before police arrived and hence escaped custody. Despite this important lead, the media reported on 31st of December 2013 that the police would conclude their investigations due to insufficient progress and lack of cooperation from local communities and rather compile all documentation to assist the coronial inquest (13).

The inquiry to establish the facts surrounding the cause and circumstances of the missing PNGIMR staff and three others started in January 2014 and was led by coroner Lawrence Kangwia. After three months of fact-finding in WNB, East Sepik, Manus and Port Moresby, hearing the testimony of witnesses and scrutinizing a range of statements, affidavits and photographs, the coroner's report was presented to the public on 17th April 2014. Coroner Kangwia concluded, based on the gathered evidence, that Gibson, Leonard, George and the skipper can be presumed dead, while Tania and Lydia may be dead or still alive. He further confirmed that the boat had not met its fate in bad weather, pointing to foul play as the reason for its disappearance. The coroner also stressed that the police investigations carried out so far had been insufficient and recommended for police to

immediately set up a fresh investigation team from outside of WNB to follow up on leads and suspects (1). Of particular interest was Jerry Wani, the relative of Gibson, who had accompanied the team and was reported by some to have been seen alive in Kimbe after the boat had disappeared (14).

The findings from the coroner's report allowed some relatives to find closure and relieved PNGIMR from the accusation of negligence raised by some family members. On the other hand, it clearly identified that more needed to be done by the State to do justice to its missing citizens. It remained highly likely that (at least some of) those responsible for this terrible incident were still at large. Yet what followed after the presentation of the coroner's findings unfortunately reflected the situation of two years earlier: urgent police operations once again lacked the required funding, despite the promises and commitments made by the government (15). And so one could read once more in the media in November 2014 that police were waiting for funds to conduct further investigations (16).

Honouring our colleagues

Whilst it is possible to describe the events surrounding the disappearance of our five PNGIMR colleagues, as we have attempted to do above, it is not possible to adequately describe the anguish and sense of loss that all of us who worked with Gibson, Tania, Lydia, Leonard and George felt at the time of their disappearance and continue to feel to this day. We realize that our pain must pale in comparison to that of their family members, but our pain is real nonetheless. PNGIMR made a brave and difficult decision to continue with the evaluation of the National Malaria Control Program after this tragic event. The basis for this decision was our collective belief that through successfully completing this project we would be honouring the memory and spirit of our five missing colleagues.

Gibson, Tania, Lydia, Leonard and George were intelligent, well-educated and young Papua New Guineans. They committed their working lives to improving the health and well-being of their beloved nation through their fledgling medical research careers. This commitment not only lives on at PNGIMR, but is strengthened by our determination to honour the memories of our missing colleagues (Figure 4). May God bless Gibson, Tania, Lydia, Leonard and George, their families and loved ones and all who work towards

Figure 4. Brass plaque erected at the headquarters of the Papua New Guinea Institute of Medical Research in Goroka in remembrance of our missing colleagues.

improving the lives of Papua New Guineans.

The Medical Society of Papua New Guinea adopted the following resolution during its Annual Symposium in 2011 in Kimbe, WNB: "The Medical Society of Papua New Guinea is concerned about the safety and wellbeing of the country's health workforce. The Society shall therefore urge the Government of Papua New Guinea to proactively implement appropriate measures to ensure the safety and wellbeing of all health workers, allied health workers and health researchers in Papua New Guinea."

ACKNOWLEDGEMENTS

The Papua New Guinea Institute of Medical Research expresses heartfelt gratitude to all individuals and organizations who assisted in the search effort for our missing colleagues, assisted in or carried out the various police investigations, contributed towards the coronial inquiry, and supported in whatever way they could the Institute and its staff members during this difficult time. We thank all those colleagues at PNGIMR and friends from PNG and around the world who contributed in cash and kind towards the expensive search and investigations carried out to date. Our deepest sympathies are once again extended to the family members and loved ones of our missing colleagues. Finally, we plead once again to the Government of Papua New Guinea to release funds to allow a full and thorough investigation of our missing colleagues to be completed.

REFERENCES

1 **Radio New Zealand International.** Inquest releases findings into missing scientists in PNG. *Radio New Zealand International*, 18 Apr 2014. www.radionz.co.nz/international/pacific-news/242001/inquest-releases-findings-into-missing-scientists-in-png

2 **Hetzel MW, Pulford J, Maraga S, Barnadas C, Reimer LJ, Tavul L, Jamea-Maiasa S, Tandrapah T, Maalsen A, Makita L, Siba PM, Mueller I.** Evaluation of the Global Fund-supported National Malaria Control Program in Papua New Guinea, 2009-2014. *PNG Med J* 2014;57;7-29.

3 **Pulford J, Oakiva T, Angwin A, Bryant M, Mueller I, Hetzel MW.** Indifferent to disease: a qualitative investigation of the reasons why some Papua New Guineans who own mosquito nets choose not to use them. *Soc Sci Med* 2012;75:2283-2290.

4 **The National.** Rescuers ill-equipped. *The National*, 31 Jan 2014. www.thenational.com.pg/?q=node/63456

5 **Blackwell E.** Fears missing scientists taken by pirates in Papua New Guinea. *The Australian*, 11 Oct 2011. www.theaustralian.com.au/news/latest-news/fears-missing-scientists-taken-by-pirates-in-papua-new-guinea/story-fn3dxity-1226166103998

6 **Levo P.** Missing girl's kin protests meet. *Post-Courier*, 23 Aug 2011:5. www.postcourier.com.pg/20110823/news09.htm

7 **Levo P.** Gulf leader calls for action against IMR. *Post-Courier*, 24 Aug 2011:18. www.postcourier.com.pg/20110824/southernpost01.htm

8 **The National.** Lack of funding hinders probe into missing eight. *The National*, 31 May 2012. www.thenational.com.pg/?q=node/33680

9 **Per Z.** O'Neill wants probe into missing scientists. *The National*, 27 Aug 2013:4. www.thenational.com.pg/?q=node/55119

10 **Kelola T.** Police have lead in missing scientists mystery. *Post-Courier*, 4 Nov 2013:1-2. www.postcourier.com.pg/20131104/news01.htm

11 **Radio New Zealand International.** PNG police hope recaptured prisoner can shed light on missing scientists. *Radio New Zealand International*, 5 Nov 2013. www.radionz.co.nz/international/pacific-news/226818/png-police-hope-recaptured-prisoner-can-shed-light-on-missing-scientists

12 **The National.** State defers inquest. *The National*, 12 Nov 2013. www.thenational.com.pg/?q=node/59525

13 **Kelola T.** Coronial inquest in January. *Post-Courier*, 31 Dec 2013:2. www.dev.postcourier.com.pg/Stories/coronial-inquest-in-january/

14 **Patjole C.** Missing scientists mystery: sixth man surfaces as possible suspect. *PNG Loop*, 18 Apr 2014. www.pngloop.com/2014/04/18/missing-scientists-mystery-sixth-man-surfaces-as-possible-suspect/

15 **Patjole C.** Investigation into missing scientists still on hold. *PNG Loop*, 22 Aug 2014. www.pngloop.com/2014/08/22/ivestigation-missing-scientists-still-hold/

16 **Kisselpar J.** Scientists mystery: police await funds to press on. *PNG Loop*, 10 Nov 2014. www.pngloop.com/2014/11/10/scientists-mystery-police-await-funds-press/

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OBITUARY

Dr Adolf Saweri MBE, MD (Hon, UPNG), Dip Med & Surg (PMC), DTM&H (Liverpool)

Isi H. Kevau[1]

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Dr Adolf Saweri passed away in the early hours of Sunday 11 November 2012, Armistice Day or the Day of Peace. In 1960 he came from what was then Dutch New Guinea to Port Moresby to be trained at the Papuan Medical College (PMC) as a medical doctor. In the same group were several other young men seeking tertiary education in medicine and other fields, including Dr Ambrose Suebu, the late Dr Chris Marjen, the late Dr Peter Pangkatana, the late Dr Hein Danomira and Rev. Dr Joshua Daimoi.

Adolf trained as an Assistant Medical Practitioner (AMP) from 1962 to 1965, graduating with the Diploma in Medicine & Surgery. Between 1966 and 1969 he was posted to different parts of this country as a resident medical officer (RMO) and as a service medical officer working at Nonga Base Hospital (Rabaul), Boram Hospital (Wewak), Saiho Rural Health Centre in Northern (Oro) Province, Kerowagi Hospital in Chimbu (Simbu) Province and Okapa Rural Health Centre in the Eastern Highlands.

The young Dr Saweri then decided that he would follow the pathway of his mentor, Professor Ian Maddocks, to become a specialist physician and an academic in Internal Medicine.

In 1971 he was accepted as a Senior Resident Medical Officer at the Royal Prince Alfred (RPA) Hospital in Sydney, becoming the first doctor from Papua New Guinea (PNG) to work in the world-renowned teaching hospital. He succeeded in this highly competitive environment, paving the way for those of us who followed. He worked under eminent specialists and was remembered for, among other things, his skill in lumbar punctures and in performing a lymph node excision biopsy under local anaesthesia when the expectation was to take the patient to the operating theatre.

In 1972 he undertook postgraduate studies at the Liverpool School of Tropical Medicine in the United Kingdom. He graduated with the Diploma in Tropical Medicine & Hygiene (DTM&H). This training prepared him to be an expert in infectious diseases, including malaria.

When he returned from the UK in January 1973, he became the first native medical doctor and the first native physician to take up an academic position. For Dr Adolf Saweri, a home-grown product, to become a physician and a Lecturer in Medicine and join expatriate physicians in the pre-Independence era was indeed a major achievement.

Adolf's PNG, RPA and Liverpool School of Tropical Medicine experience combined to make him a top clinician, a comprehensive teacher, an admirable mentor and, above all, an Ambassador of Clinical Medicine. In the resource-limited environment of PNG he realized the fundamental importance of a thorough history, a thorough and skilled examination and the use of simple and available tests. He gave an enormous amount of time and patience to listen to a patient's presenting medical complaints. He excelled in three subspecialties of Internal Medicine. He developed Respiratory Medicine in PNG and established the Asthma Clinic at Port

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Moresby General Hospital (PMGH). His special interest in Neurology dated from the days when he worked in Okapa and saw patients with kuru, which is principally a cerebellar neurodegenerative disease. This, together with his long-time involvement with the PNG Institute of Medical Research, was respected by the scientific community when he and Professor Michael Alpers and a number of other scientists attended the landmark celebration marking the end of kuru that was held in London at the Royal Society in October 2007. In his third area of expertise Adolf functioned as the Infectious Diseases encyclopaedia for his colleagues.

Adolf served the Faculty and School and the PMGH for almost 40 years, making an enormous contribution to the country's first university, and imparting his knowledge and wisdom to doctors and specialist physicians now practising in PNG, the Pacific, Australia, New Zealand and elsewhere. He was chairman of the Department (Division) of Clinical Sciences for 24 years from 1988. He was a founding member of the Association of Physicians of Papua New Guinea, and served for 38 years on the Council of the PNG Institute of Medical Research – for 33 years as the Council Chairman. In 2002 Adolf was made an Inaugural Life Member of the Medical Society of Papua New Guinea and in 2008 he was awarded the MBE for his contribution to Medicine, Medical Education and Health Services. In the same year he was awarded the Honorary Degree of Doctor of Medicine by the University of Papua New Guinea in recognition of his contribution to Medical Education and Clinical Sciences.

Adolf's 'Medical Treasure Box' is no longer with us – but he will be remembered as a pioneer in the development of medicine in PNG, an outstanding physician and teacher, a humble man imbued with a very high intellect, a man of wisdom and, for many of us, a true friend and colleague.

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OBITUARY

Adolf Saweri (28 Jul 1941 – 11 Nov 2012)

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It has been my privilege to have known and to have worked with Dr Saweri for most of my professional life. Adolf was gifted with a very high intellect and made a point of keeping up to date not only with progress in the medical sciences but also in other areas of science and with world events. He was probably one of the wisest people I have known. He was transparently honest and his views were his own – formulated by his experience, his considerable knowledge of the workings and machinations of human beings, and his generosity of spirit. He was not afraid to take on the powers-that-be over important issues when he felt that things were not right. As the first local graduate to be appointed as an academic staff member of the University of Papua New Guinea (UPNG), he championed the development of other local graduates in academic roles and he championed the development not only of undergraduate programs but also of UPNG-based postgraduate programs. He was highly regarded as a physician, with a particular interest and expertise in the areas of Respiratory Medicine and Neurology. Whilst he no doubt encountered discrimination during his earlier years, he never demonstrated resentment to his expatriate colleagues. I valued Adolf's advice on a number of issues relating to the School and, at a particularly difficult time of my own life, on a personal issue. His advice was not what I expected – but was genuine, sound and helpful.

Dr Saweri was always likely to fight on the side of the underdog. He championed students' rights and worked towards improving their conditions. He was impatient with inefficient bureaucracy. His presence in School and University meetings was marked by incisive comments – often appearing to come from 'left field' but on consideration always relevant and contributory. His comments were also usually made with his wry sense of humour.

Dr Saweri was a remarkable person who made a remarkable contribution to Medical Education in Papua New Guinea and the Pacific region. His legacy is in the doctors and other health professionals who have graduated and those who will graduate in the future from the School of which he was an integral part.

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OBITUARY

In celebration of Adolf Saweri

Ian Maddocks[1]

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As a member of staff at the newly established Papuan Medical College in the early 1960s, I was very grateful for the small group of students which had arrived from West Papua. They had been better prepared for tertiary study than any local students, and they brought a familiarity with aspects of European culture such as classical music and poetry and an interest in Indonesian cooking. In Adolf's own words, they "contributed 'spice' literally and figuratively to the life at the Papuan Medical College and perhaps beyond".

Adolf's second-year student report on a 1962 village survey in Marshall Lagoon is remarkable for its fluent English (it was his fourth language), its sensible accurate reporting, its careful sketches and graphs, but particularly for its philosophy and its mild barbed comment – characteristics which accompanied him throughout his life.

> "Most of us will keep a good memory from this journey. And for myself, I prefer to stay at an outpost rather than hang around in big towns. I want to ensure that patrol work and practical survey work is done – the best method by which a medical student (particularly in our country) can be made familiar with the things he is called upon to do. … The biggest mistake is using holiday for this kind of work. We should get a special time of the year to do this work so that students will do the work more seriously."

Adolf always had a serious intent. His unique style of speech was not always easy to follow but it was always worth the effort. He spiced his wisdom with witty and sometimes outrageous comment; he made us sit up, and think again while we laughed.

Adolf was a repository of Papuan Medical College history. He recalled the tricks for obtaining alcohol in the days when it was prohibited to 'natives'; he received the weekly ration of two sticks of black tobacco, tea leaves and two bars of laundry soap. When the Papuan Medical Students Society formed in 1964, predictably, he was elected Secretary. He divided New Guinea's expatriates into three 'Ms' (Missionaries, Mercenaries and Misfits) and he might have included himself as a 'misfit'. He was fearless in confronting trouble with wit and forthright comment; he could expose himself, literally, in a student skit on 'the bare facts' (Figure 1).

After graduation (Figure 2) he completed his residency years and two years in the field, and then returned to Port Moresby as one of the earliest national academic appointments. Adolf proved himself an effective and reliable independent clinician, but also an excellent team person. He saw expatriate staff come and go, curricula change, institutions be reorganized. Never averse to offering comment and criticism, he consistently demonstrated outstanding loyalty to the many institutions he served, which included the Papuan Medical College, the Port Moresby General Hospital, the Faculty of Medicine/ School of Medicine and Health Sciences and, after the Independence of Papua New Guinea (PNG), the PNG Institute of Medical Research and the PNG Medical Journal.

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Figure 1. Adolf Saweri as lead ballerina in 'Bare Facts', a ballet he choreographed for Island Night, Papuan Medical College, 1965. Behind his left hand are fellow student Solo Tonga and Dr Walter Wood, Lecturer in Anatomy.

Figure 2. Drs Saweri and Pangatana at their graduation in 1965, when Adolf was awarded the prize in Medicine.

Adolf's approach was honest and perceptive. In 1979 he wrote: "Last week the Faculty decided to recommend the reorganization of the Department of Clinical Sciences into 4 separate departments – medicine, surgery, paediatrics and O&G, each headed by a Professor. The right or wrong of it is not the point; what disturbed me was that the proponents of the idea have, at the most, 12 months to serve before they return to the safe confines of their own archaic systems. Then the next bunch of experts will start their own recommendation which will be implemented after *their* departure." His

scepticism saw examinations as "arbitrary hurdles to separate 'goats from sheep'", but he ran them conscientiously.

So much respected for his integrity, Adolf's rejection of self-serving practice was an important model for his students and his colleagues in a world much assailed by corruption. PNG medicine would have been well-served had he been called to chair the Medical Board.

Adolf's 1962 student report includes this comment: "A good community depends largely upon its basic unit – the family. Good families will produce good children and will grow into good citizens." The story of Adolf and Wila, finding each other in PNG 17 years after childhood pen-friend exchanges, is one of this country's great romances. The marriage produced three strong effective children, and the family continues impressive commitment and service to PNG.

The fate of their homeland, West Papua, was a great concern for Adolf and his colleagues. They were active in their support of the Provisional Government of the Republic of West Papua, based in The Hague, and in their condemnation of the so-called 'Bunker Plan' which would bring the country under Indonesian control while seeking a sham plebiscite on future options. Not long after the take-over of authority by Indonesia, I received a call from the student quarters: "Doctor, will you please come, we have heard that the Indonesians have shot Adrians' brother." As late as 1975, when exiled West Papua New Guinea leaders met in Port Moresby and drafted an ultimatum to be directed to Indonesia, it included among its 9 signatories Drs Peter Pangatana and Adolf Saweri. In the long term, however, there was little left for them to do but to get on with what a life of exile offered in PNG.

Adolf kept a sceptical eye on movements in medical education: "Philosophically the school of nursing that had acted as the surrogate mother to the school of medicine in the late 1950s and early 1960s had become a wrinkled, toothless middle-aged woman needing major plastic surgery." He also wondered about progress in his adopted country, writing in 1979: "Last year the medical superintendent was nearly lynched by mourning relatives of a DOA (dead on arrival). Maybe this is what they mean by freedom from colonial oppression. Under the colonial rule, anyone who took the law into their own hands was punished; now they are promoted into parliament or even knighted."

An issue of great concern in the early 1970s was the relative status of the Dip. Med. awarded to Medical College graduates up to 1972 and the MBBS from the Faculty of Medicine, University of Papua New Guinea (UPNG) which succeeded it. There were suggestions of a refresher course of up to a year to allow Diplomates to be awarded the MBBS. Faculty students claimed that that would undermine the status of their MBBS. The Health Department would consider no more than a 3 months' refresher. The discussion went round and round without resolution, leading to frustration and depression. Luke Rovin wrote that if graduates were any good they would not need the MBBS, and if they were not up to it, it would not help them, so why not just give it to them. Adolf, in disgust, said if they offered it to him he would refuse. The final outcome was to press on with the preparation of local postgraduate M.Med. qualifications, open to all graduates, and this has been in many ways the saving of the whole medical system in PNG, giving the nation a specialist workforce of its own.

Adolf showed both determination and persistence and a strong academic interest in medicine. He passed the DTM&H in Liverpool at a time when he had to fight for any recognition of his Diploma in the United Kingdom.

He was not a candidate for the M.Med. degree, being already recognized for his physician skills, and he was a key teacher in the M.Med. course (Figure 3).

In an obituary for the Royal Australasian College of Physicians, under the heading 'Adolf Saweri, Pioneer Physician', I wrote: "Through over nearly 40 years of service to PNG medicine and to the UPNG as physician and teacher he provided a remarkable continuity and consistency of leadership through many staff changes. He received and mentored scores of Australian physicians who came to provide consultations and teaching in Port Moresby, and medical scientists who undertook research there or through the Institute of Medical Research. In spite of his recognized expertise and seniority, he was humble and generous, always a willing work

Figure 3. Adolf in teaching mode.

Figure 4. Professor Sir Isi Kevau (Dean), Emeritus Professor Ian Maddocks (UPNG Council Member) and Dr Adolf Saweri, on the occasion of the award of an honorary MD to Dr Saweri.

horse."

Adolf's long service in the teaching of internal medicine was finally recognized with the award of Doctor of Medicine by UPNG (Figure 4).

Together with a huge number of medical colleagues both inside and outside Papua New Guinea I have been enriched by the opportunity to know and work with Adolf Saweri.

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OBITUARY

Adolf Saweri

Miila Gena[1,2]

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Dr Adolf Saweri had an inspirational and role model influence on my practice as a medical doctor. I learnt from Dr Saweri that my dream 'is possible' and 'to pay attention to detail in my clinical practice'.

I first knew about Dr Adolf Saweri when he was given residence at my high school during the time he worked at Kerowagi Hospital. The information among students was that he was the doctor working at the hospital so we would be looking through the hedges to see him. I was particularly curious to see a black doctor for the first time and I tried hard to catch a glimpse of him through the hedges. Although I was not successful, because of that opportunity to see a black doctor who lived on the school grounds it dawned on me that my dream to become a doctor was possible.

I was born and raised by my parents in Kundiawa hospital compound as they were health workers there, so I was acutely aware of the expatriate doctors and nurses who worked at the hospital. My father worked on the children's ward and also worked with the nutritional support program for sick children. I also witnessed cooking demonstrations extended into the community health program.

My encounter with Adolf as a medical student was listening to his Saturday morning side-room tutorials on the ward. He stressed paying attention to what we were reading by first trying to understand the basics. We first had to learn the basics well so we could later build in the details to increase our understanding. At the same time we must try to remove the stress by relaxing with a beer in the hand when trying to pack the brain with new information, most of all when learning the basics. Not all of us drank beer but he was making a point that learning must be enjoyable and conducted with understanding and clarity in order to provide the foundation on which we could build our knowledge of pathological conditions and add new information.

As a medical registrar, one day I informed him that the asthmatic patients had too many medications to take daily. Would he consider reducing some so the patients could become more compliant with less medications? He looked at me and stated that the different tablets worked on different sites so there was no way of reducing the number of medications.

Dealing with asthmatic patients, I tried my hardest to explain to patients the correct way to do a spirometry. It took a lot of effort on my part until one day I had the opportunity to see Dr Adolf Saweri doing a spirometry with one of his patients, where he connected the mouthpiece and gave it to the patient, looked away and said, "Blow", and then just waited. The patient did blow and I walked out telling myself not to try so hard, since patients are intelligent and will do what's right. Learning to respect patients' intelligence also helped me with making diagnoses in patients for whom previous consultations with others had not been conclusive.

When I was working as his registrar, we cared for a young woman with gross splenomegaly who was repeatedly admitted to the obstetric ward with 'anaemia in pregnancy' – though she was not pregnant but had abdominal distension from the gross enlargement of her spleen − and each time we transferred her out to the medical ward. The patient developed congestive cardiac

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failure each time and she had massive intravascular haemolysis. I watched him take time to evaluate the patient and think and eventually make the decision that she should have a splenectomy. Following this procedure we started her on pneumococcal vaccine, which we had been unable to give beforehand. The follow-up of this patient was at Gordons market where she sold her garden produce. I would remind her of the upcoming pneumococcal vaccine injection and report back to Dr Adolf that our patient had survived.

Adolf Saweri, with others, fostered deep learning for me and put speed and clarity into my clinical assessment and management.

Adolf Saweri was a man of few words and would often stammer. One needed to listen with care in order to hear and appreciate the wisdom in his few words.

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OBITUARY

Dr Adolf Saweri – an inspirational academic, researcher and mentor

Peter Max Siba[1]

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I was introduced to Dr Adolf Saweri in 1985 during a Council meeting in Goroka, and this was one year after I joined the Papua New Guinea Institute of Medical Research (PNGIMR) as a Scientific Officer in Virology. He was a very confident and self-assured person, and was surely a living encyclopaedia in science and medicine. I had admiration for this great man due to his wisdom and knowledge in medicine and science. He was always up to date with the latest innovations and practices in medicine and science.

He was a very disciplined person and he conducted his responsibilities meticulously. As the Chairman of the Papua New Guinea Institute of Medical Research Council, he presided over meetings in a very relaxed but professional manner. He was very knowledgeable about all the rules and regulations governing the PNGIMR and other research and academic institutions. He was a person who was open minded about issues, was honest and committed and deliberated on matters immediately instead of deferring them.

He will go down in the history of Papua New Guinea as the first 'native physician' and the longest-serving 'native chairman' of the PNGIMR Council. He had a passion to see national clinicians and scientists become researchers, and over the years had testified that he was happy to see that many nationals had been groomed to become researchers. It was under his leadership of the PNGIMR Council that another 'native' like me was appointed Deputy Director and eventually became the first national Director of the PNGIMR. I have learned a lot from this great and wise clinician, researcher and mentor.

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OBITUARY

A brief personal reflection on Adolf Saweri

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I found Adolf to be a simple down-to-earth person. Perhaps his greatest talent was his intuitive sense for interesting academic and/or research problems, combined with a remarkable ability to explain the most complex data sets as a simple story with a clear punch line. I came to know Adolf in 1977 as a student and worked closely with him when I joined as a Teaching Fellow in pharmacology in the Department of Human Biology (now Division of Basic Medical Sciences) in the Faculty of Medicine (now School of Medicine and Health Sciences) in 1984. He approached me to see if I was interested in taking up internal medicine as a specialization but I jokingly told him that "it is not my calling". He just laughed but did not press on with the discussion. We remained in close contact and he became my mentor in research activities throughout his life and my life at the School of Medicine and Health Sciences.

Adolf was a profound thinker but also folksy and unaffected and always ready to laugh and joke. He was able to exude charm and empathy, but nothing I did seemed to be quite good enough, particularly in the early years of my writing research papers. For this I greatly acknowledge him for my success in later years. Just two years before he passed on, I wrote an email asking for his advice on establishing a medical school at Divine Word University and his reply was: "Francis, whatever you do, never build a second class medical school, but I am willing to provide guidance." This was to be his last email to me. What I think he meant was belief in yourself in what you want to do, and for me this is possibly his best prescription for eliciting the very best efforts from many of us, including his students and postgraduates. I am privileged to have worked closely with Adolf, a simple, humble and honest person.

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OBITUARY

Adolf Saweri: a tribute

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I first came to Papua New Guinea (PNG) in 1961 and worked on kuru in the Okapa area of the Eastern Highlands. After that I carried out experimental studies and epidemiological analyses on kuru in the United States. When I moved to Perth in Australia I returned to field studies in the PNG highlands as soon as I possibly could. I had to pass through Okapa station to undertake my community-based research work and when I stopped there I usually stayed with the doctor; on my first return visit in 1969 the doctor in Okapa was Adolf Saweri. We became good friends and remained so until his death. During my time living in Perth I made these field visits of about 6 weeks to Papua New Guinea each year until I moved to Goroka at the beginning of 1977 to become Director of the Papua New Guinea Institute of Medical Research (PNGIMR). On one of these annual visits I saw Adolf in Port Moresby and he was on the Council of the Institute that appointed me as Director.

About three years afterwards he became the Chairman of the Institute Council and remained so throughout the rest of my tenure. For 20 years we conspired together to run and develop the PNGIMR. We succeeded and it became a flourishing national institution: it grew from 5 expatriate and 5 national staff when I came to 250 national staff when I left. We always operated according to the book, with regular Council meetings, minutes, resolutions and appointments by correct protocol, but the important decisions were initially made in private. We worked things out together for the benefit and advancement of the PNGIMR, its research programs and its staff. We were friends who engaged with the world on the same terms, despite our different backgrounds. We shared the same vision for the Institute and worked together to create its reality. The PNGIMR not only became a national institution but also had an international impact that continues today. A significant part of that achievement was due to the strength of the relationship between the Institute and its Council. Adolf and I effectively created this strong bond and then maintained it. We did so by discussing issues and ideas over the phone or in each other's offices and homes. Since we were always on the same wavelength we quickly reached harmonious decisions and, although these interactions were occasional, the sense of harmony was continuous. Moreover, at times of difficulty and crisis he was always there to help: when one morning the Institute, together with other statutory bodies, was abolished by act of parliament, it was to Adolf that I first turned – and he got immediately into action. Adolf has now left us and I remain the only witness to our long-lasting collaboration. There is no paper trail that others can follow except the Council proceedings and the publications and reports of the Institute: these tell what was achieved but not the story of how it was done.

Adolf was also a much cherished and effective Chairman for my two successors, John Reeder and Peter Siba. As Chairman of Council he has been formally acknowledged for his role in the achievements of the Institute. However, few will be aware of the full extent of his influence. Indeed, no-one could gauge just how vital he was to the Institute in its growing years, since the critical activities took place in a space that only Adolf and I shared. My grief for his death is thus profound, enduring and hard to articulate. I owe it to Adolf to try

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and explain why he was so important to the PNGIMR and to me, and I have made my best effort to do so here. I offer my tribute to him and his part in a firm and deep friendship that flowed on without ever rippling the surface. It is interesting to speculate just how many powerful relationships such as this have remained hidden from history because of their private nature. Adolf's wife Wila had the closest relationship with him, and she will have her own view of Adolf's engagement with the PNGIMR. A strong spousal relationship is another insufficiently acknowledged historical force, and in honouring Adolf we also honour Wila. We should do so explicitly, not taking her role for granted, and I have pleasure in paying tribute to her here.

When Adolf retired from the PNGIMR Council in May 2012 he was given a warm and moving farewell in the Adolf Saweri Lecture Theatre at the Institute's headquarters in Goroka. When I saw him at the Medical Symposium in Port Moresby in the following September he was not well; nevertheless, he attended, at least for part of the proceedings. It was lovely to see him, even if only for a few brief moments. The following November I was in Atlanta at the meeting of the American Society of Tropical Medicine and Hygiene. Soon after I arrived Deborah called me with the sad news that Adolf had died; because of her spousal insight she knew what effect this news would have. I began my lecture the next day with a tribute to Adolf Saweri.

In 2007 the End of Kuru meeting was held at the Royal Society in London. Adolf attended (Figure 1), as did Wila and their daughter Wisa. He was there in his capacity as Chairman of the PNGIMR Council but also because he had been a doctor in Okapa. He contributed to the meeting in several different ways, with his customary insightful comments, and talked about his time as a doctor in the heart of the kuru-affected region (1).

Adolf was particularly interested in asthma and other respiratory diseases. Ann Woolcock and I had a collaborative study on adult asthma in the Okapa area, where this disease was new and in remarkably high prevalence. Adolf joined one of our field research visits to affected communities. His participation was appreciated by everyone involved. His rigorous attitude to data collection, cheerful disposition and sense of proportion made him an excellent colleague to work with in the

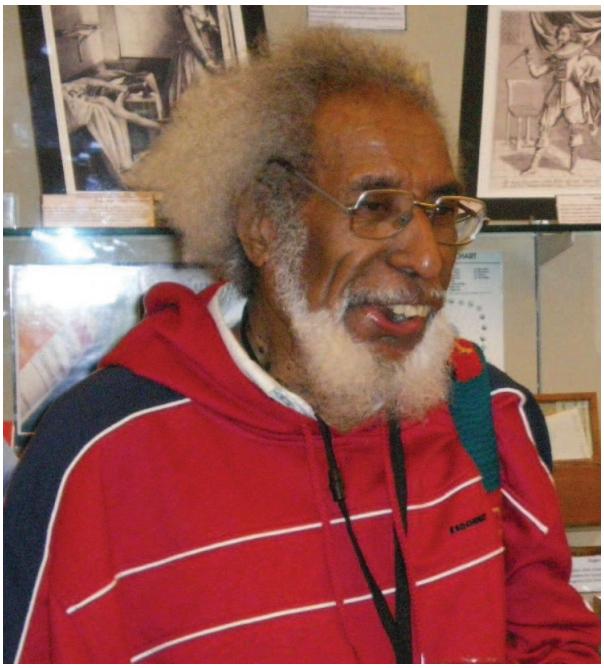

Figure 1. Adolf Saweri at the End of Kuru meeting in London, October 2007. (Photograph courtesy of Dr Deborah Lehmann)

community.

Adolf achieved many firsts in the Faculty of Medicine of the University of Papua New Guinea (2) and became the first physician to specialize in respiratory medicine. He took his data on asthma in Port Moresby to write up at the Royal Prince Alfred Hospital in Sydney. He worked there with Ann Woolcock at the Institute of Respiratory Medicine that now bears her name after her untimely death from cancer. He learned a lot in Sydney and contributed significantly to the Institute's work but, despite the prodding that he got from different directions, we were not able to persuade him to concentrate on himself and complete a PhD or MD from the findings of his research work. He was not interested in striving simply to become a professor. He was content to play his part in leading the Medical School in his own way, in providing a clinical service of the highest achievable standard and in teaching, which he was passionate about. He was awarded an honorary MD by his university in 2008, and that was for him adequate reward for his years of service and many achievements.

Adolf and I used to meet often in his office at the Medical School but as the years went on this became more and more difficult. The room was piled high with papers struggling to fill every available space and the working surface of his desk competed with further piles of books and papers, which eventually won the war and drove him out.

When living in Goroka I had – too often – to travel to Port Moresby on business. On occasions during these visits I would stay with Adolf and Wila and had the chance to meet their children. It has been a pleasure for me later in life to get to know Wisa, Moyai and Nelly better.

Adolf shared my dislike of filling in forms and had no love for the demands of bureaucracy. On one of my visits to Port Moresby he was complaining to me over a predinner drink about how difficult it was to renew his passport and how long he had been waiting for a new one. Wila intervened by asking him if he had applied for it. He replied, "Well, no, not yet." We both smiled at each other. Wila was probably somewhat less amused.

Everyone who has worked with or been taught by Adolf Saweri has an Adolf story. He had a quick wit, an agile mind and a lively imagination. He kept in touch with world events and with advances in science, not just in medicine and medical science. Furthermore, he listened. His interventions during the annual medical symposia were legendary. He was likely to enliven a discussion with an analogy drawn from black holes or global warming or to draw attention to pertinent results presented during a session on the previous day. He was enthusiastic about new scientific findings but he had to understand their relevance to human health and well-being before he was satisfied. Moreover, you had to earn Adolf's good impression and he had a well-developed detector for the 'bullshit factor'. He thought laterally and made interesting tangential comments, which were not always understood, on a wide range of topics. However, when he concentrated on an issue of central importance he identified the heart of the matter and pursued it vigorously. As well as keeping up his professional skills in medicine and science he never lost touch with people and he felt deeply about issues of justice and equity. Adolf was a remarkable person of his own special kind, and for all of us who knew him, to whatever degree, it is hard to measure the extent and depth of our loss.

REFERENCES

1 **Saweri A.** Reminiscences about kuru and the people of Okapa. *Philos Trans R Soc B Biol Sci* 2008;363:3660. doi:10.1098/rstb.2008.4027

2 **Maddocks I.** Adolf Saweri MBE, MD(Hon), DipMed, DTM&H. *Med J Aust* 2013;198:56. doi:10.5694/mja12.11714

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OBITUARY

Celebration of Adolf's life

Saweri Family[1,2,3,4]

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The Saweri family is grateful to the Editors of the PNG Medical Journal for dedicating this special collective tribute to the memory of Adolf. We miss him dearly and every day we are reminded of his mannerisms and his unique blend of humour, wit and wisdom. He was a remarkable man, who had many interests, which spanned beyond the peripheries of medicine. He wrote extensively, including numerous letters-to-the-editor, lecture notes, babalau primer ('witchdoctor' primer, a medical textbook for Papua New Guinean students), medical school history and of course his children's favourite past-time, stories. He also loved learning and was excited about modern technology and all sorts of trendy gadgets.

In what ultimately would be his last year, he encountered many new challenges: his health, in particular his failing eyesight, the problems facing the medical school and the passing of one of his closest friends and brother, Dr Peter Pangkatana. The loss of one of his oldest and dearest friends remains etched in my (Wila's) mind. During the funeral service, the few remaining brothers preceded the coffin, as Hai Tanhaku Papua (the national anthem of West Papua) was played. To understand what this meant to Adolf and his brothers is a lesson in history. As school children they were told to work hard and their duty was to build and serve their country. The tragedy of this reality is that they had to watch from afar and were helpless to prevent the atrocities which were on-going in their home land. They grew old in a foreign land, which, nevertheless, they served with distinction.

Adolf never forgot his home. Many refugees would find their way to his office. Many were poor, and seemingly forgotten by aid agencies, jobs were hard to come by and their lives tough. Quietly, he counselled them as he treated them, outlining the holistic approach to medicine he advocated, because each person is unique.

He left Sarmi as a small boy to go to school. He left a free country in 1962 and refused to return to an occupied territory; however, he maintained a deep bond with the Sobei culture, language and customs and his village Sarmi. As a lover of nature, he was heartbroken when he returned to Sarmi in 2008 to visit his sister, who was very sick at the time. In 46 years the beaches where he used to play as a kid had disappeared, the result of environmental destruction, particularly logging. He always insisted that trees were allowed to grow unhindered. When we first moved into Valkyrie Place 14, the place was bare. That first Independence Day was commemorated by the planting of laulau (bush apple) saplings. Wisa helped her father. From then on the garden flourished and blossomed. The dry leaves were left to rot, so as to fertilize the soil. The gardener was not allowed to burn them. After a few rainy days the laulau bush would be full of fruits, little red bells irresistible to all the neighbourhood kids. It was their favourite playground, climbing

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all the trees and running around the house. When we left 28 years later the garden was full of huge trees and the laulau had grown into an enormous bush.

The family was always asked to take part in his many endeavours, and in turn he spent hours helping the children with their school projects and fixing things around the house. Adolf was a good carpenter and the kids were asked to assist and were taught many valuable skills at the same time. He believed that it was not only important to know how to read and write, but also to use tools and build things. He built a cot when Wisa was born. The cot was of course painted blue (his favourite colour). When Nelly was born, Wisa and Moyai were engaged to help with the repainting of the cot. As a natural progression, the cot was transformed into a desk, which the three children used. Later on his son re-engineered it back into a cot for his first grandson, under his watchful supervision.

In 2006 we were all involved in the preparations for his keynote speech to the Medical Society, "Doing it by half". We got into the famous brown car and drove around Port Moresby to take pictures to highlight the paper. Lively discussions followed about what would be the best approach to take a picture and what props would be required for this undertaking. Two examples that tied in with the beliefs of Adolf: Moyai showing off all the latest gadgets as a poster boy for the modern doctor; Adolf and Nelly's feet bound together with the chain from our gate, a sign for young and upcoming doctors to remind them to give back to the country that has given so much to them.

Adolf did not believe in fences anyway. When it was time to fence in the yard he asked the University to put up a low fence. During the day the gate stood open.

Adolf was always open to other people and ready to help. When a serious question was posed he would give a mini lecture. Grudgingly the children learnt that there are no 'yes or no' answers in the Saweri household: indeed an answer was often "Yes and No" at the same time. He enjoyed teaching, and always encouraged everybody to study. We always jokingly said to and of Adolf that he was a man who taught others to become professors and yet he did not have the papers to do so himself. In 2008 he received two awards, an MBE and his Honorary MD title.

The last weeks of his life are vivid in my (Wila's) memory. I had just received the results of the Household and Income Survey 2009-2010 (HIES) and mentioned my concern over the high incidence of wasting in young children. High rates of stunting were to be expected, but wasting was never so high. We discussed the new figures and immediately Adolf remarked that a decline in breastfeeding could be one of the reasons for these high rates. The following Monday the Minister for Health would visit the School of Medicine and Health Sciences and Adolf asked me to accompany him and brief the Minister on the results of the HIES. During the meeting Adolf expressed his concern that the School was unable to attract academics from within the medical fraternity. He mentioned that unequal pay between staff at the Health Department and the School could be a compelling reason and urged the Minister to address this issue.

He championed equality and never ceased the battle against injustice. He taught us to believe that we were no less important than anyone else, but that respect and kindness could go a long way in helping others.

He was a very caring, loving and an exceptionally funny gentleman; he had numerous interests at heart, but his dedication to others was second to none. He would happily give his last coin to somebody and not worry about his own well-being.

On the Thursday of that same week he suffered a stroke and a week later a heart attack, from which he never recovered.

We have been saddened by his death, but we as a family also believe that we are truly blessed to have the opportunity of honouring him and, fittingly, we would like to give him the last words. Among his papers left on the computer was this chapter – "The new millennium" – that speaks volumes about this extraordinary man we knew and loved.

The new millennium

The 21st century for me should largely be a time for reflection and winding down of activities. There is, however, some unfinished business to attend to. So instead of accepting the role of the 'PNG standard old man', I still want to actively contribute

to the development of medical education. This is based mainly on my own dictum that, "Whoever brings in a new curriculum or establishes a new school must stay on for a minimum of 10 years to nurture it and fine tune the knots and kinks that are bound to occur with the new programme."

Eric J. Wright started the Papuan Medical College in 1957 or thereabout to train nurses and left about 1963-1964. As the Assistant Medical Practitioners' (AMPs') training took in the first batch of students in 1960, he left the School before the decade was out. I think he attended the first graduation, and am unsure if he attended mine in 1965. Certainly, he was not at the graduation dinner of 1965. He left others to interpret the blueprints that he had drawn up. I don't think those in whose hands he entrusted the blueprints had interpreted them correctly. They were making their own templates according to their own visions. They saw PMC School as a stepping stone to a proper medical school, like what they themselves had known from their own experience. In that sense it was useful; it made the transition easier. But the basic tenets of the school, the soul of the first school, was lost. A number of the first intake into the AMP School had done a year of Nursing as 'Preliminary Year'. Of the second intake only my friends and I had completed 5 months of probationary nursing training. These 5 months had been very valuable in my career. It taught me how to make hospital beds properly and how to shoulder-lift a patient without breaking my back. It taught me how to give an intramuscular injection without causing damage to other tissue and how to care for surgical instruments.

This is not a criticism of the Foundation Dean and Professor of Clinical Sciences, Professor Ian Maddocks, nor his cohort (Foundation Professors and the staff of the second Medical School). They had a charter to write for a new School: in other words, new functions, aims and objectives, a new motto and a new 'soul'. The blueprint of one Department of Human Biology/ Basic Medical Sciences, Pathology, Public Health and Clinical Sciences was way ahead of its time: it was superior to any of the programmes of Medical Schools in Australia, UK or wherever. Professor David Maddison had made appreciative remarks on this when he reviewed the curriculum in 1979.

But like the saga of the Sydney Opera House, the main architect of the second medical school had also left before the decade was out, leaving others to interpret the blueprints. For two decades or more every newcomer brought his/her own brand of 'tin fish' for training in the 'PNG School'. In doing so the school lost sight of the coordinated teaching that was envisioned by the founding fathers. The School lost its soul. This meant that the aims and objectives fluctuated with every new appointment. The 'Correlation Clinic' was abandoned, the Clinical Sciences Department withdrew from the neuroanatomy, neurophysiology programme, the Pathology Department withdrew from the Infectious Diseases Block, and so on.

So why did not I do something about it? I was a junior academic (Senior Tutor) and one voice among many 'iconoclasts'. The professors and the deans were interested only in their own little 'ponds full of tadpoles'. I pulled the plug on Goroka Block in 1996, when the cost threatened to drown the School. Waigani's Gunther Building and Health Department's Aopi Haus demanded for its re-instatement. My reply to them was simple: "Put the money where your mouth is." I heard nothing from them since. I maintained the Infectious Diseases Block until its natural demise with the introduction of the PBL [problem-based learning] hybrid.

The School asked me to review the curriculum three times but most of the suggestions made were never implemented. The recommendation for one-year extension of the course was rejected on financial grounds.

The Dean asked me to review the PBL hybrid a year before the Official Visit by the World Federation of Medical Schools. Reading from the staff responses, I gathered that I probably trod on many toes. I'd do it again if the opportunity presented itself, because I see it as my duty to the School to put in writing what I think was wrong or right.

When people asked me, "When are you going to retire?", my answer has always

been, "when the School and the University no longer need my services". The real answer is I wanted to see PBL take root and contribute a minimum of 10 years to the running of the programme. I was part of the group of staff members who had decided to take on the PBL hybrid. Even though I was not actively involved in the write-up of the curriculum, I felt I could not run away from it. That arbitrary ten years is now nearing its end. For the School it means a major review is in order in the near future. It must happen if the School is going to maintain its competitive edge. The School is now reaching 10 years of teaching the PBL hybrid and she must take stock for the direction of the next 10. Equally important is the notion that PBL is transferable to other programmes being offered by the School of Medicine and Health Sciences and other University Programmes. The notion of teachers imparting knowledge is passé, for most of what is to be learnt through the students' cognitive skills has been written up in books, and students are well capable of reading and acquainting themselves. They do not need and can do without the 'pre-masticated cud'.

I have no axe to grind with anybody. I am content to have been able to contribute to the training of doctors in and for Papua New Guinea and other selected Pacific Island nations. I know that this is not a big deal, but there it is. My mother wanted me to be a school teacher and I did not want to, but in the end I ended up teaching. Some friends asked me, "You have been with the School almost from the beginning and you have not progressed up the academic ladder. You have not been promoted to a Chair in Medicine. Does that not worry you?" The students have most up-to-date 'navigation charts', to borrow a seafarer's metaphor, but to sail the vast murky waters of 'clinical practice' they need a helping hand. So the answer to the question is, "I put up my hand to teach clinical medicine, and as long as I am allowed to do that without interference I am quite happy."

The concluding remarks

So it brought to an end a journey that I started in 1953 when I left Sarmi. I went to boarding school to learn something. Then I learned something more, and every time I finished something there were still other things to learn. After a while I did not know how to stop learning. Sarmi failed to attract me back and Papua Province even less so. But so did Port Moresby and PNG. It was not the place I came to know as a nineteen-year-old. Though Ela Beach was out of bounds for the black skin, there were other beaches near Motuan villages. You just asked the owners for permission to use and leave it as you found it. No garbage, no tire marks. That was what my friends and I did in Sarmi a long time ago. We play, wrestle and fight and come dusk would leave the beach. When you returned the next day the tide had re-ordered the sand and washed away the foot prints. The sand is as good as new, ready for another onslaught of little feet.

List of Medical Research Projects in Papua New Guinea

Approved or Noted

By the Medical Research Advisory Committee in 2013

Investigations into the gut microbiome of Papua New Guinean people and the potential impacts on health

Dr Paul Horwood, Dr Masahiro Umezaki, Dr Andrew Greenhill, Dr Kevin Soli, Dr Ayako Morita, Dr Tadayuki Iwase, Dr Aaron Jex and Prof. Peter Siba (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Neglected zoonoses of Papua New Guinea

Dr Mohammad Yazid Abdad (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Observational study of lymphatic filariasis transmission and elimination in Papua New Guinea

Dr Daniel Tisch and Prof. Peter Siba (The Centre for Global Health & Diseases, Case Western Reserve University School of Medicine, Wolstein Research Building, Room 4-125 2103, 2103 Cornell Road, Cleveland, Ohio 44106-7286, United States of America)

Evaluate triple drug therapy with diethylcarbamazine (DEC), albendazole (ALB) and ivermectin (IVM) that could accelerate LF elimination

Dr Christopher L. King and Prof. Peter Siba (The Centre for Global Health & Diseases, Case Western Reserve University School of Medicine, Wolstein Research Building, Room 4-125 2103, 2103 Cornell Road, Cleveland, Ohio 44106-7286, United States of America)

Effects of bubble-CPAP on outcome of severe pneumonia and neonatal respiratory distress

Dr Trevor Duke (Adjunct Professor for Child Health, School of Medicine and Health Sciences, University of Papua New Guinea and Director, Centre for International Health, Department of Paediatrics, University of Melbourne, Australia)

Exploring the involvement of fathers and elderly women in the perinatal health care of mothers and their families in Papua New Guinea

Ms Esther Pelly Batia (School of Health Sciences, Pacific Adventist University, Private Mail Bag, Boroko, National Capital District, Papua New Guinea)

Evaluation of medical supplies distribution and usage in Papua New Guinea

Dr Christopher Morgan (Burnet Institute for International Health (CIH), 85 Commercial Road, Melbourne, Victoria 3004, Australia)

Malaria parasite populations and outbreaks in PNG (Stage II)

Dr Inoni Betuela, Prof. Peter Siba, Dr Alyssa Barry and Prof. Ivo Mueller (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

Validation of a novel syphilis testing protocol in PNG

Ms Nola N'Drewei, Dr Claire Ryan, Dr Andrew Vallely, Dr Johanna Wapling, A/Prof. David Anderson and Ms Joy Liu (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Investigations into the epidemiology of infectious disease outbreaks in Papua New Guinea

Dr Paul Horwood (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Pilot intervention study to investigate the acceptability, operational feasibility and public health impact of point of care HPV-DNA testing as a screening tool for early detection and treatment of cervical cancer in Papua New Guinea

Dr Andrew Vallely (Papua New Guinea

Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Investigating the burden of *Plasmodium ovale curtisi* and *Plasmodium ovale wallikeri* infection in Papua New Guinea
Dr Celine Barnadas and Dr Eline Kattenberg (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Does immunological hyporesponsiveness exist following 23-valent pneumococcal polysaccharide vaccine in PNG children primed with pneumococcal conjugate vaccine?
Dr William Pomat, Dr Deborah Lehmann and Dr Peter Richmond (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

The risk factors involved with acute lower respiratory infections and meningitis in Papua New Guinea
Dr Rebecca Ford, Dr William Pomat and Dr Paul Horwood (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Investigations into the gut microbiome of infants in Papua New Guinea
Dr Kevin Soli (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Antibodies to capsular polysaccharides and protein antigens of *Streptococcus pneumoniae* and *Haemophilus influenzae* in a malaria endemic region of Papua New Guinea and factors affecting transplacental transfer of these antibodies to their offspring
Dr William Pomat and Dr Inoni Betuela (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Prevalence of azithromycin resistance markers in malaria parasites infecting women in late pregnancy in Papua New Guinea
Dr Eline Kattenberg (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

Natural development of antibodies specific to *Streptococcus pneumoniae* choline binding protein A, and non-typeable *Haemophilus influenzae* protein D and outer membrane proteins in PNG highland infants
Ms Jacinta Francis (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Assessment of in vitro sensitivity of local *Plasmodium falciparum* strains to sulfadoxine-pyrimethamine
Ms Tamarah Koleala, Dr Brioni Moore and Dr Eline Kattenberg (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

Adaptation of *Plasmodium* spp. drug resistance genotyping protocols to the requirements of MagPix instrumentation
Ms Sarah Javati (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Validating SNP-based molecular barcoding of *Plasmodium vivax* in Papua New Guinea
Mr Lincoln Timinao (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Investigation into the prevalence of human enterovirus 71 and other enterovirus infections among children in Papua New Guinea
Mr Matthew Omena (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Exploring the role of midwives for improving maternal health in PNG
Ms Susan Crabtree (University of Auckland, Centre for Development Studies, Human Sciences Building, Level 8, 10 Symonds Street, Auckland, New Zealand)

To assess nurses' perceptions of their knowledge, skills and preparedness for disaster management
Sr Renagi Moliyola (Laloki Psychiatric Hospital, PO Box 1239, Boroko, National Capital District, Papua New Guinea)
Evaluation of a dual-point of care rapid test simultaneously detecting non-treponemal and treponemal antibodies in patients with yaws

Dr Oriol Mitja (Lihir Medical Centre, PO Box 34, Lihir, New Ireland Province, Papua New Guinea)

A cross-sectional study evaluating the side effects of the new malaria treatment protocol, artemether-lumefantrine and primaquine, in Papua New Guinea
Dr Evelyn K. Lavu (Manager, Central Public Health Laboratory (CPHL), Port Moresby General Hospital, Private Mail Bag 1, Boroko, National Capital District, Papua New Guinea)

Conduits and barriers to health worker implementation of the new national malaria treatment protocol in Papua New Guinea
Ms Rosheila Dagina (School of Medicine and Health Sciences, University of Papua New Guinea, PO Box 5623, Boroko, National Capital District, Papua New Guinea)

To identify challenges faced within the organization structure during the implementation of Provincial Health Authority Act 2007 in Milne Bay Province
Ms Victoria Aitsi (Divine Word University, PO Box 483, Madang, Madang Province, Papua New Guinea)

To determine the safety and efficacy of adding thioridazine to DOTS therapy for six months in patients with pulmonary tuberculosis starting DOTS and MDR-TB treatment
Dr Martin Daimen (SMO, Internal Medicine, Modilon Teaching Hospital, PO Box 2030, Madang, Madang Province, Papua New Guinea)

Assessing the water, sanitation and hygiene practices and needs during childbirth in the Western Highlands, PNG
Ms Alison Macintyre (Western Highlands Province, C/- Water Aid PNG, PO Box 250, Boroko, National Capital District, Papua New Guinea)

Formative evaluation PNG: rural primary health services delivery project
Mr Luke Elich (JTA International, PO Box 87, Waigani, National Capital District, Papua New Guinea)

Anal intercourse and STI risk in Papua New Guinea: a pilot study to ascertain the acceptability and feasibility of anorectal swab collection and STI testing
Dr Angela Kelly and Dr Johanna Wapling (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Sexuality, health and human rights of marginalised men in Papua New Guinea
Dr Angela Kelly and Ms Geraldine Maibani (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Vaginal carriage of Group B *Streptococcus* in pregnant women in Madang Province, PNG
Ms Bernadine Kasian, Dr Regina Wangnapi, Dr William Pomat, Dr Andrew Greenhill, Prof. Peter Siba, Dr Maria Ome, Prof. Ivo Mueller, Dr Johanna Wapling, Prof. Stephen Rogerson, Dr Holger Unger and Dr Eline Kattenberg (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

Prevalence and disease burden of enteric parasites amongst children in Goroka, Eastern Highlands Province, Papua New Guinea
Ms Grace Bande (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

p^2: a rapid assessment of provider initiated counselling and testing and post-exposure prophylaxis for HIV prevention in the highlands of PNG
Mr Matthew David (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

Metabolic markers of *Plasmodium vivax* liver stage infection and genetic determinants of relapses
Dr Inoni Betuela, Dr Leanne Robinson, Dr Celine Barnadas and Prof. Ivo Mueller (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

A rapid assessment of criminal law, HIV, sex work and male to male sex in Papua New Guinea: HIV and other public health impact
Dr Angela Kelly (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

CMCA health: Middle and South Fly baseline study
Mr Geoff Scahill (JTA International, Gateway

Hotel, PO Box 1215, Boroko, National Capital District, Papua New Guinea)

Experiences of girls and women around first menstruation and body changes in Papua New Guinea
Ms Elizabeth Gumbaketi (Faculty of Medicine and Health Science, School of Medicine and Dentistry, Cairns Clinical – Cairns Base Hospital, PO Box 902, Cairns, Queensland 4870, Australia)

Preliminary investigation of neglected zoonoses in humans, animals and invertebrate pests of Papua New Guinea
Dr Mohammad Yazid Abdad (Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

What hinders nurses and midwives addressing sexuality issues with pregnant women during antenatal care in Papua New Guinea
Ms Esther Koski (Pacific Adventist University, Private Mail Bag, Boroko, National Capital District, Papua New Guinea)

The role of antenatal parental education (APE) linking expectant fathers to inclusive antenatal care (ANC)
Mr McKenzie Maviso (Pacific Adventist University, Private Mail Bag, Boroko, National Capital District, Papua New Guinea)

A safety, tolerability and preliminary efficacy study of azithromycin plus piperaquine as preventative treatment in pregnant Papua New Guinean women
Dr Brioni Moore (Papua New Guinea Institute of Medical Research, PO Box 378, Madang, Madang Province, Papua New Guinea)

Mobility and HIV risk across the Papua New Guinea/Indonesia border
Associate Prof. Heather Worth (C/- Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

What makes for good leadership in Papua New Guinea's response to HIV?
Associate Prof. Heather Worth (C/- Papua New Guinea Institute of Medical Research, PO Box 60, Goroka, Eastern Highlands Province, Papua New Guinea)

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 2013.

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, National Capital District, Papua New Guinea).

MEDLARS BIBLIOGRAPHY

PUBLICATIONS OF RELEVANCE TO PAPUA NEW GUINEA AND MELANESIA

Bibliographic Citation List generated from MEDLARS

1 **Ang MJ, Yong GH, Poulsen A, Then SW, Li Z, Joy J, Hill J, Chia CS.**
Substrate-based peptidomimetic inhibitors of the Murray Valley encephalitis virus NS2B/NS3 serine protease: a P1-P4 SAR study.
Eur J Med Chem 2013 Oct;68:72-80. doi: 10.1016/j.ejmech.2013.07.028. Epub 2013 Aug 9.

Murray Valley encephalitis is an infectious disease spread by a mosquito-borne virus endemic in Papua New Guinea and northern Australia. In the past decade, it has spread to various regions of Australia and there is currently no therapeutic treatment against this disease. An attractive drug target is the viral serine protease NS2B/NS3, a critical enzyme involved in viral replication. Herein, we report the inhibitory activities of 37 C-terminal agmatine peptidomimetic inhibitors which led to the design of a novel structurally-constrained competitive inhibitor 38 possessing a Ki of 2.5±0.5 μM. We believe our data provides crucial insights into the viral protease active site specificity which could be used to facilitate drug design against Murray Valley encephalitis viral infections.

2 **Arnott A, Mueller I, Ramsland PA, Siba PM, Reeder JC, Barry AE.**
Global population structure of the genes encoding the malaria vaccine candidate, *Plasmodium vivax* apical membrane antigen 1 (PvAMA1).
PLoS Negl Trop Dis 2013 Oct 31;7(10):e2506. doi: 10.1371/journal.pntd.0002506. eCollection 2013.

BACKGROUND: The *Plasmodium vivax* apical membrane antigen 1 (PvAMA1) is a promising malaria vaccine candidate; however, it remains unclear which regions are naturally targeted by host immunity and whether its high genetic diversity will preclude coverage by a monovalent vaccine. To assess its feasibility as a vaccine candidate, we investigated the global population structure of PvAMA1. METHODOLOGY AND PRINCIPAL FINDINGS: New sequences from Papua New Guinea (PNG, n = 102) were analysed together with published sequences from Thailand (n = 158), India (n= 8), Sri Lanka (n = 23), Venezuela (n = 74) and a collection of isolates from disparate geographic locations (n = 8). A total of 92 single nucleotide polymorphisms (SNPs) were identified including 22 synonymous SNPs and 70 non-synonymous (NS) SNPs. Polymorphisms and signatures of balancing (positive Tajima's D and low FST values) selection were predominantly clustered in domain I, suggesting it is a dominant target of protective immune responses. To estimate global antigenic diversity, haplotypes comprised of (i) non-singleton (n = 40) and (ii) common (≥10% minor allele frequency, n = 23) polymorphic amino acid sites were then analysed revealing a total of 219 and 210 distinct haplotypes, respectively. Although highly diverse, the 210 haplotypes comprised of only common polymorphisms were grouped into eleven clusters; however, substantial geographic differentiation was observed, and this may have implications for the efficacy of PvAMA1 vaccines in different malaria-endemic areas. The PNG haplotypes form a distinct group of clusters not found in any other geographic region. Vaccine haplotypes were rare and geographically restricted, suggesting potentially poor efficacy of candidate PvAMA1 vaccines. CONCLUSIONS: It may be possible to cover the existing global PvAMA1 diversity by selection of diverse alleles based on these analyses; however, it will be important to first define the relationships between the genetic and antigenic diversity of this molecule.

3 **Céspedes N, Arévalo-Herrera M, Felger I, Reed S, Kajava AV, Corradin G, Herrera S.**
Antigenicity and immunogenicity of a novel chimeric peptide antigen based on the *P. vivax* circumsporozoite protein.
Vaccine 2013 Oct 1;31(42):4923-4930. doi: 10.1016/j.vaccine.2013.05.082. Epub 2013 Aug 15.

BACKGROUND: *Plasmodium vivax* circumsporozoite (PvCS) protein is a major sporozoite surface antigen involved in parasite invasion of hepatocytes and is currently being considered as vaccine candidate. PvCS contains a dimorphic central repetitive fragment flanked by conserved regions that contain functional domains. METHODS: We have developed a chimeric 137-mer synthetic polypeptide (PvCS-NRC) that includes the conserved region I and region II-plus and the two natural repeat variants known as VK210 and VK247. The antigenicity of PvCS-NRC was tested using human sera from PNG and Colombia endemic areas and its immunogenicity was confirmed in mice with different genetic backgrounds; the polypeptide formulated either in Alum or GLA-SE adjuvants was assessed in inbred C3H, CB6F1 and outbred ICR mice, whereas a formulation in Montanide ISA51 was tested in C3H mice. RESULTS: Antigenicity studies indicated that the chimeric peptide is recognized by a high proportion (60-70%) of residents of malaria-endemic areas. Peptides formulated with either GLA-SE or Montanide ISA51 adjuvants induced stronger antibody responses as compared with the Alum formulation. Sera from immunized mice as well as antigen-specific affinity-purified human IgG antibodies reacted with sporozoite preparations in immunofluorescence and Western blot assays, and displayed strong in vitro inhibition of sporozoite invasion (ISI) into hepatoma cells. CONCLUSIONS: The polypeptide was recognized at high prevalence when tested against naturally induced human antibodies and was able to induce significant immunogenicity in mice. Additionally, specific antibodies were able to recognize sporozoites and were able to block sporozoite invasion in vitro. Further evaluation of this chimeric protein construct in preclinical phase, eg, in *Aotus* monkeys, in order to assess the humoral and cellular immune responses as well as protective efficacy against parasite challenge

of the vaccine candidate must be conducted.

4 **Cooper A, Stringer CB.**
Paleontology. Did the Denisovans cross Wallace's Line?
Science 2013 Oct 18;342(6156):321-323. doi: 10.1126/science.1244869.

5 **Cooper EL, Hirabayashi K.**
Origin of innate immune responses: revelation of food and medicinal applications.
J Tradit Complement Med 2013 Oct;3(4):204-212. doi: 10.4103/2225-4110.119708.

Much is known about the strong ecological impact that earthworms (Qiū Yǐn; Pheretima) have on soil in terms of fertility, nutrient production, and tilling. Even more interesting though is the impact they have had on our understanding of innate immunity, and from this discovery, there has been a simultaneous recognition of their potential through their historical use as food and their use in treatment of certain chronic health problems that often afflict humans. This bifurcating growing knowledge base has stemmed from centuries of honing and practicing traditional and complementary forms of medicine such as Ayurveda (India), Traditional Chinese Medicine (China), Kampo (Japan), and Traditional Korean Medicine (Korea). Earthworms (Dilong) have also been credited as a model for research concerning the nervous and endocrine systems. One of the reasons behind the earthworm's tremendous impact on research into these biomedical endeavors is partly due to its lack of ethical restrictions, like those imposed on vertebrate models. Using invertebrate models as opposed to mice or other mammalian models bypasses ethical concerns. Moreover, financial constraints consistently hover over biological research that requires living subjects, preferably mammals. Earthworms are a rich source of several vital biological macromolecules and other nutrients. They have long been used as food in several cultures such as the Ye'Kuana in Venezuela, the Maori in New Zealand, and the nomadic populations in Papua New Guinea. Earthworms and their nutritious products have been shown to exert significant effects in treating humans for disorders of inflammation and blood coagulation. One area that continues to be examined is the earthworm's ability to regenerate lost appendages, and these effects have been extended to mammals. Evidence reveals that earthworm extracts may actually promote the regeneration of damaged nerves. This presentation will explore how earthworms may reveal significant advances and conclusions that decipher innate immunity. This is intimately associated with them as sources of their various nutritional and medicinal benefits.

6 **Dimitrov BD, Valev D, Werner R, Atanassova PA.**
Cyclic patterns of cerebral malaria admissions in Papua New Guinea for the years 1987-1996.
Epidemiol Infect 2013 Nov;141(11):2317-2327. doi: 10.1017/S0950268812003111. Epub 2013 Jan 23.

Data on the dynamics of malaria incidence, admissions and mortality and their best possible description are very important to better forecast and assess the implementation of programmes to register, monitor (eg, by remote sensing) and control the disease, especially in endemic zones. Semi-annual and seasonal cycles in malaria rates have been observed in various countries and close similarity with cycles in the natural environment (temperature, heliogeophysical activity, etc.), host immunity and/or virulence of the parasite suggested. This study aimed at confirming previous results on malaria cyclicity by exploring whether trans-year and/or multiannual cycles might exist. The exploration of underlying chronomes (time structures) was done with raw data (without smoothing) by linear and nonlinear parametric regression models, autocorrelation, spectral (Fourier) and periodogram regression analysis. The strongest cyclical patterns of detrended malaria admissions were (i) annual period of 1·0 year (12 months or seasonality); (ii) quasi-biennial cycle of about 2·25 years; and (iii) infrannual, circadecennial cycle of about 10·3 years. The seasonal maximum occurred in May with the minimum in September. Notably, these cycles corresponded to similar cyclic components of heliogeophysical activity such as sunspot seasonality and solar activity cyclicities and well-known climate/weather oscillations. Further analyses are thus warranted to investigate such similarities. In conclusion, multicomponent cyclical dynamics of cerebral malaria admissions in Papua New Guinea were observed thus allowing more specific analyses and modelling as well as correlations with environmental factors of similar cyclicity to be explored. Such further results might also contribute to and provide more precise estimates for the forecasting and prevention, as well as the better understanding, of the dynamics and aetiology of this vector-borne disease.

7 **Douglas NM, Lampah DA, Kenangalem E, Simpson JA, Poespoprodjo JR, Sugiarto P, Anstey NM, Price RN.**
Major burden of severe anemia from non-falciparum malaria species in Southern Papua: a hospital-based surveillance study.
PLoS Med 2013 Dec;10(12):e1001575; discussion e1001575.
doi: 10.1371/journal.pmed.1001575. Epub 2013 Dec 17.

BACKGROUND: The burden of anemia attributable to non-falciparum malarias in regions with *Plasmodium* co-endemicity is poorly documented. We compared the hematological profile of patients with and without malaria in southern Papua, Indonesia. METHODS AND FINDINGS: Clinical and laboratory data were linked for all patients presenting to a referral hospital between April 2004 and December 2012. Data were available on patient demographics, malaria diagnosis, hemoglobin concentration, and clinical outcome, but other potential causes of anemia could not be identified reliably. Of 922,120 patient episodes (837,989 as outpatients and 84,131 as inpatients), a total of 219,845 (23.8%) were associated with a hemoglobin measurement, of whom 67,696 (30.8%) had malaria. Patients with *P. malariae* infection had the lowest hemoglobin concentration (n = 1,608, mean = 8.93 [95% CI 8.81-9.06]), followed by those with mixed species infections (n = 8,645, mean = 9.22 [95% CI 9.16-9.28]), *P. falciparum* (n = 37,554, mean = 9.47 [95% CI 9.44-9.50]), and *P. vivax* (n = 19,858, mean = 9.53 [95% CI 9.49-9.57]); p-value for all comparisons <0.001. Severe anemia (hemoglobin <5 g/dl) was present in 8,151 (3.7%) patients. Compared to patients without malaria, those with mixed *Plasmodium* infection were at greatest risk of severe anemia (adjusted odds ratio [AOR] 3.25 [95% CI 2.99-3.54]); AORs for severe anaemia associated with *P. falciparum*, *P. vivax*, and *P. malariae* were 2.11 (95% CI 2.00-2.23), 1.87 (95% CI 1.74-2.01), and 2.18

(95% CI 1.76-2.67), respectively, p <0.001. Overall, 12.2% (95% CI 11.2%-13.3%) of severe anemia was attributable to non-falciparum infections compared with 15.1% (95% CI 13.9%-16.3%) for *P. falciparum* monoinfections. Patients with severe anemia had an increased risk of death (AOR = 5.80 [95% CI 5.17-6.50]; p <0.001). Not all patients had a hemoglobin measurement, thus limitations of the study include the potential for selection bias, and possible residual confounding in multivariable analyses. CONCLUSIONS: In Papua *P. vivax* is the dominant cause of severe anemia in early infancy, mixed *P. vivax*/*P. falciparum* infections are associated with a greater hematological impairment than either species alone, and in adulthood *P. malariae*, although rare, is associated with the lowest hemoglobin concentration. These findings highlight the public health importance of integrated genus-wide malaria control strategies in areas of *Plasmodium* co-endemicity.

8 **Fletcher SM, Thiessen J, Gero A, Rumsey M, Kuruppu N, Willetts J.**
Traditional coping strategies and disaster response: examples from the South Pacific region.
J Environ Public Health 2013;2013:264503. doi: 10.1155/2013/264503. Epub 2013 Dec 23.

The Pacific Islands are vulnerable to climate change and increased risk of disasters not only because of their isolated and often low lying geographical setting but because of their economic status which renders them reliant on donor support. In a qualitative study exploring the adaptive capacity of Pacific Island Countries (PICs) across four countries, Cook Islands, Fiji, Samoa, and Vanuatu, it was clear that traditional coping strategies are consistently being applied as part of response to disasters and climate changes. This paper describes five common strategies employed in PICs as understood through this research: recognition of traditional methods; faith and religious beliefs; traditional governance and leadership; family and community involvement; and agriculture and food security. While this study does not trial the efficacy of these methods, it provides an indication of what methods are being used and therefore a starting point for further research into which of these traditional strategies are beneficial. These findings also provide important impetus for Pacific Island governments to recognise traditional approaches in their disaster preparedness and response processes.

9 **Fulu E, Jewkes R, Roselli T, Garcia-Moreno C; UN Multi-country Cross-sectional Study on Men and Violence Research Team.**
Prevalence of and factors associated with male perpetration of intimate partner violence: findings from the UN Multi-country Cross-sectional Study on Men and Violence in Asia and the Pacific.
Lancet Glob Health 2013 Oct;1(4):e187-207. doi: 10.1016/S2214-109X(13)70074-3. Epub 2013 Sep 10.

BACKGROUND: Male perpetration of intimate partner violence (IPV) is under-researched. In this article, we present data for the prevalence of, and factors associated with, male perpetration of IPV from the UN Multi-country Cross-sectional Study on Men and Violence in Asia and the Pacific. We aimed to estimate the prevalence of perpetration of partner violence, identify factors associated with perpetration of different forms of violence, and inform prevention strategies. METHODS: We undertook standardised population-based household surveys with a multistage representative sample of men aged 18-49 years in nine sites in Bangladesh, China, Cambodia, Indonesia, Sri Lanka, and Papua New Guinea between January, 2011, and December, 2012. We built multinomial regression models of factors associated with lifetime violence perpetration: physical IPV, sexual IPV, both physical and sexual IPV, multiple emotional or economic IPV versus none, and calculated population-attributable fractions. In the analysis, we considered factors related to social characteristics, gender attitudes and relationship practices, victimisation history, psychological factors, substance misuse, and participation in violence outside the home. FINDINGS: 10,178 men completed interviews in our study (between 815 and 1812 per site). The response rate was higher than 82·5% in all sites except for urban Bangladesh (73·2%) and Sri Lanka (58·7%). The prevalence of physical or sexual IPV perpetration, or both, varied by site, between 25·4% (190/746; rural Indonesia) and 80·0% (572/714; Bougainville, Papua New Guinea). When multiple emotional or economic abuse was included, the prevalence of IPV perpetration ranged from 39·3% (409/1040; Sri Lanka) to 87·3% (623/714; Bougainville, Papua New Guinea). Factors associated with IPV perpetration varied by country and type of violence. On the basis of population-attributable fractions, we show factors related to gender and relationship practices to be most important, followed by experiences of childhood trauma, alcohol misuse and depression, low education, poverty, and involvement in gangs and fights with weapons. INTERPRETATION: Perpetration of IPV by men is highly prevalent in the general population in the sites studied. Prevention of IPV is crucial, and interventions should address gender socialisation and power relations, abuse in childhood, mental health issues, and poverty. Interventions should be tailored to respond to the specific patterns of violence in various contexts. Physical and sexual partner violence might need to be addressed in different ways. FUNDING: Partners for Prevention a UN Development Programme, UN Population Fund, UN Women, and UN Volunteers regional joint programme for gender-based violence prevention in Asia and the Pacific; UN Population Fund Bangladesh and China; UN Women Cambodia and Indonesia; UN Development Programme in Papua New Guinea and Pacific Centre; and the Governments of Australia, the UK, Norway, and Sweden.

10 **Gray KA, Dowd S, Bain L, Bobogare A, Wini L, Shanks GD, Cheng Q.**
Population genetics of *Plasmodium falciparum* and *Plasmodium vivax* and asymptomatic malaria in Temotu Province, Solomon Islands.
Malar J 2013 Nov 22;12:429. doi: 10.1186/1475-2875-12-429.

BACKGROUND: Temotu Province, Solomon Islands is progressing toward malaria elimination. A baseline survey conducted in 2008 showed that most *Plasmodium* infections in the province were of low parasite density and asymptomatic infections. To better understand mechanisms underlying these malaria transmission characteristics genetic diversity and relationships among *Plasmodium falciparum* and *Plasmodium vivax* populations in the province were examined. METHODS: Forty-five *P. falciparum* and 67 *P. vivax* samples collected in the 2008 baseline survey were successfully genotyped using eight *P.*

falciparum and seven *P. vivax* microsatellite markers. Genetic diversity, relationships and distribution of both *P. falciparum* and *P. vivax* populations were analysed. RESULTS: *Plasmodium falciparum* population exhibited low diversity with 19 haplotypes identified and had closely related clusters indicating clonal expansion. Interestingly, a dominant haplotype was significantly associated with fever and high parasite density. In contrast, the *P. vivax* population was highly diverse with 58 haplotypes identified that were not closely related. Parasite populations between different islands in the province showed low genetic differentiation. CONCLUSION: The low diversity and clonal population of *P. falciparum* population may partially account for clinical immunity developed against illness. However, it is possible that importation of a new *P. falciparum* strain was the major cause of illness. High diversity in *P. vivax* population and low relatedness between strains suggested clinical immunity to *P. vivax* may be maintained by different mechanisms. The genetic diversity, population structure and distribution of strains indicate that transmission of *P. falciparum* was low, but that of *P. vivax* was still high in 2008. These data will be useful for assessing changes in malaria transmission resulting from interventions.

11 **Hall JJ, Gillespie JA, Rosewell A, Mapira P.**
The Papua New Guinea cholera outbreak: implications for PNG, Australia and the Torres Strait.
Med J Aust 2013 Nov 4;199(9):576-577.

12 **Jewkes R, Fulu E, Roselli T, Garcia-Moreno C; UN Multi-country Cross-sectional Study on Men and Violence Research Team.**
Prevalence of and factors associated with non-partner rape perpetration: findings from the UN Multi-country Cross-sectional Study on Men and Violence in Asia and the Pacific.
Lancet Glob Health 2013 Oct;1(4):e208-e218. doi: 10.1016/S2214-109X(13)70069-X. Epub 2013 Sep 10.

BACKGROUND: Rape perpetration is under-researched. In this study, we aimed to describe the prevalence of, and factors associated with, male perpetration of rape of non-partner women and of men, and the reasons for rape, from nine sites in Asia and the Pacific across six countries: Bangladesh, China, Cambodia, Indonesia, Papua New Guinea, and Sri Lanka. METHODS: In this cross-sectional study, undertaken in January 2011-December 2012, for each site we chose a multistage representative sample of households and interviewed one man aged 18-49 years from each. Men self-completed questions about rape perpetration. We present multinomial regression models of factors associated with single and multiple perpetrator rape and multivariable logistic regression models of factors associated with perpetration of male rape with population-attributable fractions. FINDINGS: We interviewed 10,178 men in our study (815-1812 per site). The prevalence of non-partner single perpetrator rape varied between 2·5% (28/1131; rural Bangladesh) and 26·6% (225/846; Bougainville, Papua New Guinea), multiple perpetrator rape between 1·4% (18/1246; urban Bangladesh) and 14·1% (119/846; Bougainville, Papua New Guinea), and male rape between 1·5% (13/880; Jayapura, Indonesia) and 7·7% (65/850; Bougainville, Papua New Guinea). 57·5% (587/1022) of men who raped a non-partner committed their first rape as teenagers. Frequent reasons for rape were sexual entitlement (666/909; 73·3%, 95% CI 70·3-76·0), seeking of entertainment (541/921; 58·7%, 55·0-62·4), and as a punishment (343/905; 37·9%, 34·5-41·4). Alcohol was a factor in 249 of 921 cases (27·0%, 95% CI 24·2-30·1). Associated factors included poverty, personal history of victimisation (especially in childhood), low empathy, alcohol misuse, masculinities emphasising heterosexual performance, dominance over women, and participation in gangs and related activities. Only 443 of 1933 men (22·9%, 95% CI 20·7-25·3) who had committed rape had ever been sent to prison for any period. INTERPRETATION: Rape perpetration committed by men is quite frequent in the general population in the countries studied, as it is in other countries where similar research has been undertaken, such as South Africa. Prevention of rape is essential, and interventions must focus on childhood and adolescence, and address culturally rooted male gender socialisation and power relations, abuse in childhood, and poverty. FUNDING: Partners for Prevention – a UN Development Programme, UN Population Fund, UN Women, and UN Volunteers regional joint programme for gender-based violence prevention in Asia and the Pacific; UN Population Fund Bangladesh and China; UN Women Cambodia and Indonesia; United Nations Development Programme in Papua New Guinea and Pacific Centre; and the Governments of Australia, the UK, Norway, and Sweden.

13 **Jonduo M, Wong SS, Kapo N, Ominipi P, Abdad M, Siba P, McKenzie P, Webby R, Horwood P.**
Surveillance of avian influenza viruses in Papua New Guinean poultry, June 2011 to April 2012.
Western Pac Surveill Response J 2013 Dec 19;4(4):11-15. doi: 10.5365/WPSAR.2013.4.4.004.

We investigated the circulation of avian influenza viruses in poultry populations throughout Papua New Guinea to assess the risk to the poultry industry and human health. Oropharyngeal swabs, cloacal swabs and serum were collected from 537 poultry from 14 provinces of Papua New Guinea over an 11-month period (June 2011 through April 2012). Virological and serological investigations were undertaken to determine the prevalence of avian influenza viruses. Neither influenza A viruses nor antibodies were detected in any of the samples. This study demonstrated that avian influenza viruses were not circulating at detectable levels in poultry populations in Papua New Guinea during the sampling period. However, avian influenza remains a significant risk to Papua New Guinea due to the close proximity of countries having previously reported highly pathogenic avian influenza viruses and the low biosecurity precautions associated with the rearing of most poultry populations in the country.

14 **Kayyal MH, Russell JA.**
Americans and Palestinians judge spontaneous facial expressions of emotion.
Emotion 2013 Oct;13(5):891-904. doi: 10.1037/a0033244. Epub 2013 Jun 24.

The claim that certain emotions are universally recognized from facial expressions is based primarily on the study of expressions that were posed. The current study was of spontaneous facial expressions shown by aborigines in Papua New Guinea (Ekman, 1980); 17 faces claimed to convey one (or, in the case of blends, two) basic emotions and five faces claimed to show other universal feelings. For each face, participants rated the degree to which each of

the 12 predicted emotions or feelings was conveyed. The modal choice for English-speaking Americans (n = 60), English-speaking Palestinians (n = 60), and Arabic-speaking Palestinians (n = 44) was the predicted label for only 4, 5, and 4, respectively, of the 17 faces for basic emotions, and for only 2, 2, and 2, respectively, of the 5 faces for other feelings. Observers endorsed the predicted emotion or feeling moderately often (65%, 55%, and 44%), but also denied it moderately often (35%, 45%, and 56%). They also endorsed more than one (or, for blends, two) label(s) in each face – on average, 2.3, 2.3, and 1.5 of basic emotions and 2.6, 2.2, and 1.5 of other feelings. There were both similarities and differences across culture and language, but the emotional meaning of a facial expression is not well captured by the predicted label(s) or, indeed, by any single label.

15 **Kelly-Hanku A, Vallely A, Man WY, Wilson D, Law G, Gray R.**
A systematic review of heterosexual anal intercourse and its role in the transmission of HIV and other sexually transmitted infections in Papua New Guinea. *BMC Public Health* 2013 Dec 1;13:1108. doi: 10.1186/1471-2458-13-1108.

BACKGROUND: Papua New Guinea (PNG) has a high burden of sexually transmitted infections (STIs) and the highest adult HIV prevalence in the Pacific region. Despite this burden of disease, heterosexual anal intercourse (HAI) has rarely been considered. Given the increasing number of, and interest in, behavioural surveys in PNG and the changing nature of PNG's HIV epidemic, it is timely to conduct a systematic review of HAI in PNG in order to improve sexual health. METHODS: We performed a systematic review of HAI in PNG as reported in peer-reviewed and non-peer-reviewed publications for the period 1950-May 2012. The search strategy identified 475 publications. After screening by geographical location, topic and methodology, we identified 23 publications for full text review, following which 13 publications were included in the final review. Using data from the review, we performed a risk equation analysis to demonstrate the potential impact of HAI on HIV acquisition and incidence in PNG. RESULTS: There is a paucity of well-informed behavioural research on HAI in PNG. Inconsistency in key questions on HAI made it impossible to conduct a meta-analysis. The data available on HAI shows that it is practiced in all geographical areas and among all populations. Of those who reported HAI, rates varied from as low as 8% to as high as 77% depending on the recall period and partner type. Condom use during HAI was consistently low. Our risk equation analysis indicates that even if only 20% of females engage in HAI, and only 10% of sex acts involve HAI, the total number of new HIV infections among females would be 40% greater than if vaginal intercourse only occurred. CONCLUSIONS: Our findings indicate that HAI may be an important driver of the HIV epidemic in PNG. In order to improve the sexual health of Papua New Guineans, efforts are required to improve behavioural surveillance of HAI as well as develop national HIV/STI programing and policy to better address the risks associated with unprotected HAI.

16 **Kennedy EC, Bulu S, Harris J, Humphreys D, Malverus J, Gray NJ.**
"Be kind to young people so they feel at home": a qualitative study of adolescents' and service providers' perceptions of youth-friendly sexual and reproductive health services in Vanuatu.
BMC Health Serv Res 2013 Oct 31;13:455. doi: 10.1186/1472-6963-13-455.

BACKGROUND: Sexual activity during adolescence is common in Vanuatu; however, many adolescents lack access to sexual and reproductive health (SRH) services and subsequently suffer a disproportionate burden of poor SRH. There is limited peer-reviewed research describing adolescents' SRH service delivery preferences in Vanuatu to inform policy and programs. The aim of this qualitative study was to explore the barriers preventing adolescents from accessing SRH services in Vanuatu and the features of a youth-friendly health service as defined by adolescents. METHODS: Sixty-six focus group discussions were conducted with 341 male and female adolescents aged 15-19 years in rural and urban communities. Additionally, 12 semi-structured interviews were undertaken with policymakers and service providers. Data were analysed using thematic analysis. RESULTS: Socio-cultural norms and taboos regarding adolescent sexual behavior were the most significant factors preventing adolescents from accessing services. These contributed to adolescents' own fear and shame, judgmental attitudes of service providers, and disapproval from parents and community gate-keepers. Lack of confidentiality and privacy, costs, and adolescents' lack of SRH knowledge were also important barriers. Adolescents and service providers identified opportunities to make existing services more youth-friendly. The most important feature of a youth-friendly health service described by adolescents was a friendly service provider. Free or affordable services, reliable commodity supply, confidentiality and privacy were also key features. The need to address socio-cultural norms and community knowledge and attitudes was also highlighted. CONCLUSIONS: There are significant demand and supply-side barriers contributing to low utilisation of SRH services by adolescents in Vanuatu. However, there are many opportunities to make existing SRH services more youth-friendly, such as improving service provider training. Investment is also required in strategies that aim to create a more supportive environment for adolescent SRH.

17 **Kurumop SF, Bullen C, Whittaker R, Betuela I, Hetzel MW, Pulford J.**
Improving health worker adherence to malaria treatment guidelines in Papua New Guinea: feasibility and acceptability of a text message reminder service. *PLoS One* 2013 Oct 7;8(10):e76578. doi: 10.1371/journal.pone.0076578. eCollection 2013.

The aim of this study is to assess whether a text message reminder service designed to support health worker adherence to a revised malaria treatment protocol is feasible and acceptable in Papua New Guinea (PNG). The study took place in six purposively selected health facilities located in the Eastern Highlands Province (EHP) of PNG. Ten text messages designed to remind participants of key elements of the new NMTP were transmitted to 42 health workers twice over a two-week period (two text messages per day, Monday to Friday) via the country's largest mobile network provider. The feasibility and acceptability of the text message reminder service was assessed by transmission reports, participant diaries and group discussions. Findings indicate that the vast majority of text messages were successfully

transmitted, participants had regular mobile phone access and that most text messages were read most of the time and were considered both acceptable and clinically useful. Nevertheless, the study found that PNG health workers may tire of the service if the same messages are repeated too many times and that health workers may be reluctant to utilize more comprehensive, yet complementary, resources. In conclusion, a text message reminder service to support health worker adherence to the new malaria treatment protocol is feasible and acceptable in PNG. A rigorous pragmatic, effectiveness trial would be justified on the basis of these findings.

18 **Kuzma J.**
Knowledge, attitude and practice related to infant feeding among women in rural Papua New Guinea: a descriptive, mixed method study.
Int Breastfeed J 2013 Nov 21;8(1):16. doi: 10.1186/1746-4358-8-16.

BACKGROUND: Despite the well-recognized effectiveness of exclusive breastfeeding for the first six months of an infant life for reducing infant mortality, adherence to this practice is not widespread in the developing world. Although several studies on infant nutrition practices have been conducted in urban settings of Papua New Guinea (PNG), there is only scant information on infant feeding practices in rural settings. Therefore, this study aimed to investigate knowledge, attitude and practice associated with exclusive breastfeeding in various locations in rural PNG. METHODS: A mixed method study using interviews based on a semi-structured questionnaire (n = 140) and Focus Group Discussions (FGDs) was conducted among mothers in rural PNG between August and September 2012. Participants were selected using convenience sampling. Included in the study were both primiparous and multiparous mothers with a child below the age of two years. Content analysis was used for qualitative data and descriptive statistics were used for quantitative data. RESULTS: Whereas most women indicated breastfeeding as a better way to feed babies, knowledge of the reasons for its superiority over infant formula was generally poor. Only 17% of mothers practiced exclusive breastfeeding for the first six months postpartum. Our study showed that the size of the gap between exclusive breastfeeding practice and global recommendations was striking. Taking into account the low educational profile of the participants, the disparity may be explained by the fact that most of the mothers in this study had no formal education on infant feeding. CONCLUSIONS: This study showed a lack of understanding of the importance of and poor adherence to exclusive breastfeeding for the first six months postpartum among rural mothers. As exclusive breastfeeding promotion has been proved to be one of most effective ways to improve infant survival, more attention should be given to it, especially targeting the large proportion of women who missed formal education on infant feeding in school. A proper community-based program including the tools for monitoring its implementation and effectiveness needs to be developed to transform policy recommendations into action in rural PNG.

19 **Laman M, Manning L, Siba PM, Davis TME.**
Prevalence and implications of cerebrospinal fluid leukocytosis in Papua New Guinean children hospitalized with severe malaria.
Am J Trop Med Hyg 2013 Nov;89(5):866-868. doi: 10.4269/ajtmh.13-0281. Epub 2013 Sep 9.

Cerebrospinal fluid (CSF) leukocytosis in severe malaria was assessed in 87 children in Papua New Guinea participating in a detailed longitudinal observational study who had undergone lumbar puncture for further investigation of altered consciousness and/or convulsions. After rigorous exclusion of non-malarial infection, 16 (20.5%) of 78 children with *Plasmodium falciparum* monoinfection but 0 of 9 with *P. vivax*/mixed-species malaria had a detectable CSF leukocytosis, which was unrelated to prior, including complex, seizures. There were eight children with a CSF leukocyte density >10 cells/μL (9.2% of the total sample), half of whom had cerebral malaria (4 of 22, 18.1%). Cerebrospinal fluid leukocytosis is infrequent in severe pediatric malaria, especially in children with *P. vivax* infections, and it is generally mild. Its presence in a blood slide-positive child should prompt consideration of alternative diagnoses and empiric antibiotic therapy.

20 **Limb RJ, Rosenfeld JV, McLean C.**
Giant orbital oncocytoma.
Asian J Neurosurg 2013 Oct;8(4):192-194. doi: 10.4103/1793-5482.125668.

BACKGROUND AND IMPORTANCE: Oncocytomas are rare benign tumours often arising from the lacrimal or salivary glands, usually small in size. CLINICAL PRESENTATION: We report a giant unilateral orbital oncocytoma in a 19-year-old male from Papua New Guinea, presenting with progressive proptosis-threatening vision. Due to retro-ocular extension of the lesion, surgical excision was performed via a fronto-orbitozygomatic craniotomy and orbitotomy. A sub-total excision of the lesion was achieved, with overall improvement in proptosis and cosmesis. CONCLUSION: This appears to be the first documented case of a giant intra-orbital oncocytoma being resected neurosurgically via craniotomy.

21 **Lu Z, Koch M, Harper MK, Matainaho TK, Barrows LR, Van Wagoner RM, Ireland CM.**
Plakinamine M, a steroidal alkaloid from the marine sponge *Corticium* sp.
J Nat Prod 2013 Nov 22;76(11):2150-2152. doi: 10.1021/np400649e. Epub 2013 Nov 6.

By means of bioassay-guided fractionation, a new steroidal alkaloid, plakinamine M (1), and the known compound, plakinamine L (2), with a unique acyclic side chain, were isolated from the marine sponge *Corticium* sp. collected from New Britain, Papua New Guinea. The structures were determined on the basis of extensive 1D and 2D NMR and HRESIMS. The two compounds showed inhibition of *Mycobacterium tuberculosis* with MIC values of 15.8 and 3.6 μg/mL, respectively.

22 **Martin E, de Leeuw E.**
Exploring the implementation of the Framework Convention on Tobacco Control in four small island developing states of the Pacific: a qualitative study.
BMJ Open 2013 Dec 9;3(12):e003982. doi: 10.1136/bmjopen-2013-003982.

OBJECTIVES: To determine what variables influence the implementation of the Framework Convention on Tobacco Control (FCTC) in small island developing states of the Pacific and how they affect its success or failure. To explore how barriers can be overcome and opportunities utilised to ensure an effective FCTC implementation in the Pacific Islands. DESIGN: A mixed methods, multiple case

study consisting of primarily qualitative data in the form of semistructured interviews, document analysis and opportunistic observation. SETTING: Field visits were undertaken to collect data in the Cook Islands, Vanuatu, Palau and Nauru. The key informants were interviewed in the major cities or islands of each respective country: Rarotonga, Port Vila, Koror and Nauru. PARTICIPANTS: Purposive sampling was used to select 39 informants, whose roles were associated with FCTC implementation. Most of the participants worked in health-oriented positions in the government and non-government organisations. RESULTS: Each country made a significant progress towards FCTC implementation. Overall, strong policy content, public support and limited pro-tobacco coalition activity were conducive to FCTC implementation, but the challenges were evident in the form of limited capacity, limited antitobacco coalition activity and limited political commitment outside the ministries of health in each country. CONCLUSIONS: Further efforts are needed for full FCTC implementation, through building capacity and using resources effectively, growing commitment to FCTC beyond the health sector, fostering growth in antitobacco coalition activity, exploiting the limited pro-tobacco activity that may be present and garnering public support for tobacco control. These lessons may be particularly important for other small island developing states in the Pacific and developing countries elsewhere.

23 **Mitjà O, Bassat Q.**
Developments in therapy and diagnosis of yaws and future prospects.
Expert Rev Anti Infect Ther 2013 Oct;11(10):1115-1121. doi: 10.1586/14787210.2013.833059. Epub 2013 Sep 27.

Yaws, a chronic and debilitating infectious disease caused by *Treponema pallidum* subsp. *pertenue*, and closely related to syphilis, although transmitted by skin-to-skin contact, remains an important public health challenge, causing a significant burden of morbidity in children in certain areas of the Pacific and Africa. Recent advances in its diagnosis and treatment have led to an enthusiastic upsurge of activities related to its control, and exciting perspectives of global eradication. Although possibly considered among the most neglected of all neglected diseases during decades, there seems to be now agreement that massive drug administration of the antibiotic azithromycin, coupled with adequate surveillance of foci of transmission could result in its eradication. In this review, we summarize current knowledge regarding the therapeutics of yaws and its diagnosis.

24 **Mitjà O, Šmajs D, Bassat Q.**
Advances in the diagnosis of endemic treponematoses: yaws, bejel, and pinta.
PLoS Negl Trop Dis 2013 Oct 24;7(10):e2283. doi: 10.1371/journal.pntd.0002283. eCollection 2013.

Improved understanding of the differential diagnosis of endemic treponematoses is needed to inform clinical practice and to ensure the best outcome for a new global initiative for the eradication of yaws, bejel, and pinta. Traditionally, the human treponematoses have been differentiated based upon their clinical manifestations and epidemiologic characteristics because the etiologic agents are indistinguishable in the laboratory. Serological tests are still considered standard laboratory methods for the diagnosis of endemic treponematoses and new rapid point-of-care treponemal tests have become available which are extremely useful in low-resource settings. In the past ten years, there has been an increasing effort to apply polymerase chain reaction to treponematoses and whole genome fingerprinting techniques have identified genetic signatures that can differentiate the existing treponemal strains; however, definitive diagnosis is also hampered by widespread unavailability of molecular diagnostics. We review the dilemmas in the diagnosis of endemic treponematoses, and advances in the discovery of new diagnostic tools.

25 **Mola G, Kirby B.**
Discrepancies between national maternal mortality data and international estimates: the experience of Papua New Guinea.
Reprod Health Matters 2013 Nov;21(42):191-202. doi: 10.1016/S0968-8080(13)42725-8.

Over the past 30 years maternal mortality estimates for Papua New Guinea have varied widely. There is no mandatory vital registration in PNG, and 85% of the population lives in rural areas with limited or no access to health services. Demographic Health Survey data for PNG estimates the maternal mortality ratio to be 370 deaths per 100,000 live births in 1996 and 733 in 2006, whereas estimates based upon mathematical models (as calculated by international bodies) gave figures of 930 for 1980 and 230 for 2010. This disparity has been a source of considerable confusion for health workers, policy makers and development partners. In this study, we compared 2009 facility-based survey data with figures from the National Health Information System records. The comparison revealed similar maternal mortality ratios: for provincial hospitals (245 and 295), government health centres (574 and 386), church agency health centres (624 and 624), and nationally (394 and 438). Synthesizing these estimates for supervised births in facilities and data on unsupervised births from a community-based survey in one province indicates a national MMR of about 500. Knowing the maternal mortality ratio is a necessary starting point for working out how to reduce it.

26 **Owen R, Bedford P, Leditschke J, Schlenker A, Hartman D.**
Post mortem sampling of the bladder for the identification of victims of fire related deaths.
Forensic Sci Int 2013 Dec 10;233(1-3):14-20. doi: 10.1016/j.forsciint.2013.07.018. Epub 2013 Aug 2.

In a coronial setting a deceased person must be formally identified. It is difficult to identify a deceased person when their physical features are disrupted and identification by visual means cannot occur. In the absence of visual identification, the confirmation of identity of a deceased person relies on the scientific comparison of information obtained post mortem with ante mortem information. The ante mortem information may include dental and medical records, fingerprints, and DNA profiling. For cases involving incinerated remains, this traditionally requires the collection of blood, muscle or bone samples from the deceased (depending on the severity of the burns) for DNA analysis and subsequent comparison to a reference sample for kinship determination. Following on from work conducted during the DVI response to a plane crash in Papua New Guinea in 2011, a study has been performed examining the viability of utilising material obtained from bladder swabs in deaths associated with fires. Twenty-eight cases were

analysed during 2012 with deaths occurring in motor vehicle and aviation accidents, as well as house fires, homicides and from self-immolation. Bladder and conventional (blood, muscle or bone) samples were subjected to DNA analysis and compared. Our findings demonstrate that the bladder samples all gave DNA of sufficient quality for DNA profiling. This easily obtained sample (when available) can be now recommended in the scientific identification process of fire affected deceased persons.

27 **Pulford J, Kurumop SF, Ura Y, Siba PM, Mueller I, Hetzel MW.**
Malaria case management in Papua New Guinea following the introduction of a revised treatment protocol.
Malar J 2013 Nov 27;12:433. doi: 10.1186/1475-2875-12-433.

BACKGROUND: This paper reports on the availability of diagnostic tools and recommended anti-malarials in the 12-month period immediately following the implementation of a new national malaria treatment protocol (NMTP) in Papua New Guinea (PNG). Health worker adherence to the new NMTP is also examined and comparisons made with previously reported pre-implementation findings. METHODS: A countrywide cross-sectional survey in randomly selected primary health care facilities (n = 88). Data were collected via passive observation of the clinical case management of fever or suspected malaria patients and via an interviewer administered questionnaire completed with the officer in charge of each participating health care facility. RESULTS: Malaria rapid diagnostic tests (RDTs) and the new first-line anti-malarial medication, artemether-lumefantrine (AL), were available in 53.4% and 51.1% of surveyed health facilities, respectively. However, they were more widely available in the larger health centres as compared to the smaller aid-posts (90.2% vs. 21.3% and 87.8% vs. 19.2%, respectively). Overall, 68.3% of observed fever cases (n = 445) were tested for malaria by RDT and 39% prescribed an anti-malarial, inclusive of 98.2% of RDT positive patients and 19.8% of RDT negative cases. The availability and use of malaria RDTs was greater in the current survey as compared to pre-implementation of the new NMTP (8.9% vs. 53.4% & 16.2% vs. 68.3%, respectively) as was the availability of AL (0% vs. 51.1%). The percentage of fever patients prescribed anti-malarials decreased substantially post implementation of the new NMTP (96.4% vs. 39.0%). CONCLUSIONS: PNG has achieved high coverage of malaria RDTs and AL at the health centre level, but these resources have yet to reach the majority of aid-posts. Malaria case management practice has substantially changed in the 12-month period immediately following the new NMTP, although full protocol adherence was rarely observed.

28 **Ralph AP, Kenangalem E, Waramori G, Pontororing GJ, Sandjaja, Tjitra E, Maguire GP, Kelly PM, Anstey NM.**
High morbidity during treatment and residual pulmonary disability in pulmonary tuberculosis: under-recognised phenomena.
PLoS One 2013 Nov 29;8(11):e80302. doi: 10.1371/journal.pone.0080302. eCollection 2013.

BACKGROUND: In pulmonary tuberculosis (PTB), morbidity during treatment and residual pulmonary disability can be under-estimated. METHODS: Among adults with smear-positive PTB at an outpatient clinic in Papua, Indonesia, we assessed morbidity at baseline and during treatment, and 6-month residual disability, by measuring functional capacity (six-minute walk test [6MWT] and pulmonary function), quality of life (St George's Respiratory Questionnaire [SGRQ]) and Adverse Events ([AE]: new symptoms not present at outset). Results were compared with findings in locally-recruited volunteers. RESULTS: 200 PTB patients and 40 volunteers were enrolled. 6WMT was 497 m (interquartile range 460-529) in controls versus 408 m (IQR 346-450) in PTB patients at baseline (p <0.0001) and 470 m (IQR 418-515) in PTB patients after 6 months (p = 0.02 versus controls). SGRQ total score was 0 units (IQR 0-2.9) in controls, versus 36.9 (27.4-52.8) in PTB patients at baseline (p <0.0001) and 4.3 (1.7-8.8) by 6 months (p <0.0001). Mean percentage of predicted FEV1 was 92% (standard deviation 19.9) in controls, versus 63% (19.4) in PTB patients at baseline (p<0.0001) and 71% (17.5) by 6 months (p <0.0001). After 6 months, 27% of TB patients still had at least moderate-severe pulmonary function impairment, and 57% still had respiratory symptoms, despite most achieving 'successful' treatment outcomes, and reporting good quality of life. More-advanced disease at baseline (longer illness duration, worse baseline X-ray) and HIV positivity predicted residual disability. AE at any time during treatment were common: itch 59%, arthralgia 58%, headache 40%, nausea 33%, vomiting 16%. CONCLUSION: We found high 6-month residual pulmonary disability and high AE rates. Although PTB treatment is highly successful, the extent of morbidity during treatment and residual impairment could be overlooked if not specifically sought. Calculations of PTB-related burden of disease should acknowledge that TB-related morbidity does not stop at 6 months. Early case detection and treatment are key in minimising residual impairment.

29 **Raveh A, Delekta PC, Dobry CJ, Peng W, Schultz PJ, Blakely PK, Tai AW, Matainaho T, Irani DN, Sherman DH, Miller DJ.**
Discovery of potent broad spectrum antivirals derived from marine actinobacteria.
PLoS One 2013 Dec 5;8(12):e82318. doi: 10.1371/journal.pone.0082318. eCollection 2013.

Natural products provide a vast array of chemical structures to explore in the discovery of new medicines. Although secondary metabolites produced by microbes have been developed to treat a variety of diseases, including bacterial and fungal infections, to date there has been limited investigation of natural products with antiviral activity. In this report, we used a phenotypic cell-based replicon assay coupled with an iterative biochemical fractionation process to identify, purify, and characterize antiviral compounds produced by marine microbes. We isolated a compound from *Streptomyces kaviengensis*, a novel actinomycetes isolated from marine sediments obtained off the coast of New Ireland, Papua New Guinea, which we identified as antimycin A1a. This compound displays potent activity against western equine encephalitis virus in cultured cells with half-maximal inhibitory concentrations of less than 4 nM and a selectivity index of greater than 550. Our efforts also revealed that several antimycin A analogues display antiviral activity, and mechanism of action studies confirmed that these *Streptomyces*-derived secondary metabolites function by inhibiting the cellular mitochondrial electron transport chain,

thereby suppressing de novo pyrimidine synthesis. Furthermore, we found that antimycin A functions as a broad spectrum agent with activity against a wide range of RNA viruses in cultured cells, including members of the *Togaviridae*, *Flaviviridae*, *Bunyaviridae*, *Picornaviridae*, and *Paramyxoviridae* families. Finally, we demonstrate that antimycin A reduces central nervous system viral titers, improves clinical disease severity, and enhances survival in mice given a lethal challenge with western equine encephalitis virus. Our results provide conclusive validation for using natural product resources derived from marine microbes as source material for antiviral drug discovery, and they indicate that host mitochondrial electron transport is a viable target for the continued development of broadly active antiviral compounds.

30 **Rosewell A, Ropa B, Randall H, Dagina R, Hurim S, Bieb S, Datta S, Ramamurthy S, Mola G, Zwi AB, Ray P, MacIntyre CR.**
Mobile phone-based syndromic surveillance system, Papua New Guinea.
Emerg Infect Dis 2013 Nov;19(11):1811-1818. doi: 10.3201/eid1911.121843.

The health care system in Papua New Guinea is fragile, and surveillance systems infrequently meet international standards. To strengthen outbreak identification, health authorities piloted a mobile phone-based syndromic surveillance system and used established frameworks to evaluate whether the system was meeting objectives. Stakeholder experience was investigated by using standardized questionnaires and focus groups. Nine sites reported data that included 7 outbreaks and 92 cases of acute watery diarrhea. The new system was more timely (2.4 vs. 84 days), complete (70% vs. 40%), and sensitive (95% vs. 26%) than existing systems. The system was simple, stable, useful, and acceptable; however, feedback and subnational involvement were weak. A simple syndromic surveillance system implemented in a fragile state enabled more timely, complete, and sensitive data reporting for disease risk assessment. Feedback and provincial involvement require improvement. Use of mobile phone technology might improve the timeliness and efficiency of public health surveillance.

31 **Soli KW, Kas M, Maure T, Umezaki M, Morita A, Siba PM, Greenhill AR, Horwood PF.**
Evaluation of colorimetric detection methods for *Shigella*, *Salmonella*, and *Vibrio cholerae* by loop-mediated isothermal amplification.
Diagn Microbiol Infect Dis 2013 Dec;77(4):321-323. doi: 10.1016/j.diagmicrobio.2013.09.009. Epub 2013 Sep 28.

We evaluated loop-mediated isothermal amplification end-point detection methods for *Salmonella*, *Shigella*, and *Vibrio cholerae*. Detection sensitivities were comparable to real-time PCR methods. The colorimetric dyes hydroxynaphthol blue and SYBR Green I showed increased sensitivity when compared to visual and automated turbidity readings. End-point colorimetric dyes promise great utility in developing settings.

32 **Stanisic DI, Javati S, Kiniboro B, Lin E, Jiang J, Singh B, Meyer EV, Siba P, Koepfli C, Felger I, Galinski MR, Mueller I.**
Naturally acquired immune responses to *P. vivax* merozoite surface protein 3α and merozoite surface protein 9 are associated with reduced risk of *P. vivax* malaria in young Papua New Guinean children.
PLoS Negl Trop Dis 2013 Nov 14;7(11):e2498. doi: 10.1371/journal.pntd.0002498. eCollection 2013.

BACKGROUND: *Plasmodium vivax* is the most geographically widespread human malaria parasite. Cohort studies in Papua New Guinea have identified a rapid onset of immunity against vivax-malaria in children living in highly endemic areas. Although numerous *P. vivax* merozoite antigens are targets of naturally acquired antibodies, the role of many of these antibodies in protective immunity is yet unknown. METHODOLOGY/PRINCIPAL FINDINGS: In a cohort of children aged 1-3 years, antibodies to different regions of Merozoite Surface Protein 3α (PvMSP3α) and Merozoite Surface Protein 9 (PvMSP9) were measured and related to prospective risk of *P. vivax* malaria during 16 months of active follow-up. Overall, there was a low prevalence of antibodies to PvMSP3α and PvMSP9 proteins (9-65%). Antibodies to the PvMSP3α N-terminal, Block I and Block II regions increased significantly with age while antibodies to the PvMSP3α Block I and PvMSP9 N-terminal regions were positively associated with concurrent *P. vivax* infection. Independent of exposure (defined as the number of genetically distinct blood-stage infections acquired over time (molFOB)) and age, antibodies specific to both PvMSP3α Block II (adjusted incidence rate ratio (aIRR) = 0.59, p = 0.011) and PvMSP9 N-terminus (aIRR = 0.68, p = 0.035) were associated with protection against clinical *P. vivax* malaria. This protection was most pronounced against high-density infections. For PvMSP3α Block II, the effect was stronger with higher levels of antibodies. CONCLUSIONS: These results indicate that PvMSP3α Block II and PvMSP9 N-terminus should be further investigated for their potential as *P. vivax* vaccine antigens. Controlling for molFOB assures that the observed associations are not confounded by individual differences in exposure.

33 **Sumardi, Hertiani T, Sasmito E.**
Ant plant (*Myrmecodia tuberosa*) hypocotyl extract modulates TCD4+ and TCD8+ cell profile of doxorubicin-induced immune-suppressed Sprague Dawley rats in vivo.
Sci Pharm 2013 Jun 21;81(4):1057-1069. doi: 10.3797/scipharm.1302-03. eCollection 2013.

Myrmecodia tuberosa Jack (Rubiaceae) has been used as part of traditional Indonesian remedies for a wide range of therapeutic usages in West Papua. Our preliminary study revealed the significant potency of these plant extracts and fractions as an immunomodulator by an in vitro technique on Balb/c mice. This study explored the effect of *M. tuberosa* hypocotyl ethanol extract on the TCD4+ and TCD8+ cell profiles of doxorubicin (Dox)-induced immune-suppressed Sprague Dawley (SD) rats by an in vivo method. Dried powder of *M. tuberosa* hypocotyl was macerated in 95% ethanol. Following solvent evaporation in a vacuum, the ethanol extract (EE) was partitioned to yield an n-hexane fraction (FH) and residue (FNH). FNH was further partitioned to yield ethyl acetate (FEtOAc) and water fractions (FW). The extract and fractions in the concentrations 10, 20, 50, and 100 µg/mL were tested on macrophage cells by the latex bead method, while the proliferation of lymphocyte cells was evaluated by the MTT assay. The total phenolic and flavonoid contents of those fractions were evaluated. The active fraction was administrated orally on Dox-induced SD rats for 28

days by an in vivo method to observe the TCD4+ and TCD8+ cell profiles. The in vivo assay showed that the FNH could maintain the number of TCD4+ cells, but not the number of TCD8+ cells. The ED50 observed was 24.24 mg/kg BW. Steroid/terpenoid compounds were detected in this fraction along with the phenolics and flavonoids. The FNH contained 3.548 ± 0.058% GAE of total phenolics and 0.656 ± 0.026% QE of total flavonoids. *M. tuberosa* hypocotyl extract is a potent immunomodulatory agent and may act as co-chemotherapy in Dox use.

34 **Tommbe R, MacLaren DJ, Redman-MacLaren ML, Mafile'o TA, Asugeni L, McBride WJ.**
Researching male circumcision for HIV prevention in Papua New Guinea: a process that incorporates science, faith and culture.
Health Res Policy Syst 2013 Nov 13;11:44. doi: 10.1186/1478-4505-11-44.

BACKGROUND: Undertaking HIV research in the culturally diverse Pacific nation of Papua New Guinea (PNG) requires careful consideration of social, cultural and religious beliefs and practices. Here, we share a detailed description of culturally informed research processes and lessons learned from the first ever study undertaken on male circumcision for HIV prevention at a faith-based university in PNG. METHODS: Male and female staff and students at Pacific Adventist University were invited to complete an anonymous self-administered questionnaire, and/or participate in a semi-structured interview or focus group discussion. Male participants were invited for clinical examination. Results were collated and disseminated to the university community in gender segregated sessions. The study deliberately partnered with student leaders and centralised social, cultural, and religious paradigms. Student leaders were interviewed about their experience of partnering in sensitive health research. RESULTS: The student leaders reported that pre-existing relationships, cultural ties, gendered sensitivity and regular communication reinforced trust between researchers, student leaders and participants, and helped the success of the study. The amount of time, complex logistics and social and cultural relationships between single and married staff and students were highlighted as challenges. CONCLUSIONS: Partnering with regional student leaders to plan and implement the study gave a legitimate and immediate mechanism for involving PNG staff and students in this sensitive health research. Gendered research processes utilised established social and cultural structures and ensured the safety of participants; all of these factors contributed to the acceptability of the study. Capacity was strengthened in PNG and Australian researchers to undertake sensitive HIV research in PNG. The study demonstrated that it is possible to conduct sensitive sexual health research at a faith-based university in PNG.

35 **Vallely LM, Homiehombo P, Kelly AM, Vallely A, Homer CS, Whittaker A.**
Exploring women's perspectives of access to care during pregnancy and childbirth: a qualitative study from rural Papua New Guinea.
Midwifery 2013 Oct;29(10):1222-1229. doi: 10.1016/j.midw.2013.03.011. Epub 2013 May 14.

OBJECTIVES: to explore women's perceptions and experiences of pregnancy and childbirth in a rural community in PNG. DESIGN: a qualitative, descriptive study comprising focus group discussions (FGDs) and in-depth interviews. SETTING: this study took place in a rural community in Eastern Highlands Province, PNG. PARTICIPANTS: 51 women participated in seven focus group discussions. In-depth interviews were undertaken with 21 women, including women recruited at the antenatal clinic, women purposively selected in the community and three key informants in the community. FINDINGS: the majority of women mentioned the benefits of receiving antenatal care at the health facility and the importance of a supervised, facility birth. Women faced numerous challenges with regard to accessing these services, including geographical, financial and language barriers. Cultural and customary beliefs surrounding childbirth and lack of decision making powers also impacted on whether women had a supervised birth. KEY CONCLUSIONS AND IMPLICATIONS FOR PRACTICE: distance, terrain and transport as well as decision making processes and customary beliefs influenced whether a woman did or did not reach a health facility to give birth. While the wider issue of availability and location of health services and health system strengthening is addressed shorter term, community-based interventions could be of benefit. These interventions should include safe motherhood and birth preparedness messages disseminated to women, men and key family and community members.

36 **van Leeuwen E, Mashuri A.**
Intergroup helping in response to separatism.
Pers Soc Psychol Bull 2013 Dec;39(12):1647-1655. doi: 10.1177/0146167213499613. Epub 2013 Aug 15.

Despite its prevalence and widespread media coverage, separatism as a phenomenon is barely covered in psychological investigations, and the majority's response to separatism has been completely ignored. We present two studies in which we investigated the notion that separatist movements threaten the continuation of the national identity, as well as the nation's economic position. Moreover, we hypothesized and found that members of the majority group respond to continuation threat by supporting government measures to help the separatist group. Javanese students who were induced to believe that existing separatist movements in West Papua (Study 1, N = 322) or Aceh (Study 2, N = 180) were currently increasing their efforts to gain independence were more willing to support these groups than participants who believed these movements were dormant. Moreover, this effect was mediated by continuation threat but not economic threat. These results demonstrate the possibility of a peaceful response to separatism threat.

37 **Wandra T, Ito A, Swastika K, Dharmawan NS, Sako Y, Okamoto M.**
Taeniases and cysticercosis in Indonesia: past and present situations.
Parasitology 2013 Nov;140(13):1608-1616. doi: 10.1017/S0031182013000863. Epub 2013 Aug 21.

The main aim of this study is to overview the past and present situations of human taeniases and cysticercosis in Indonesia and including future perspectives. Through joint projects from 1996, we have confirmed the occurrence of *Taenia saginata* (beef tapeworm) in Bali, of *Taenia solium* (pork tapeworm) mainly in Papua and sporadically in Bali, and of *Taenia asiatica* in North Sumatra. These taeniases were caused through eating uncooked pork and pig viscera for *T. solium* and *T. asiatica*, respectively, and beef for *T. saginata*. The distribution

of these tapeworms in Indonesia is basically highly restricted by the traditional cultural and religious backgrounds in each island. *T. saginata* is relatively common in Bali although people consume pork 'lawar' more than beef 'lawar'. Taeniases due to *T. saginata* or *T. asiatica* and *T. solium* and cysticercosis due to *T. solium* have also been sporadically reported in some other islands. Among these species, *T. solium* is exceptional since humans can be infected not only by larval stages (cysticerci) in pork but also by eggs released from human tapeworm carriers. Cysticercosis has been confirmed in Indonesia in humans, pigs and even dogs.

38 **Waramboi JG, Gidley MJ, Sopade PA.**
Carotenoid contents of extruded and non-extruded sweetpotato flours from Papua New Guinea and Australia.
Food Chem 2013 Dec 1;141(3):1740-1746. doi: 10.1016/j.foodchem.2013.04.070. Epub 2013 May 3.

Carotenoid contents of extruded and non-extruded flours of Papua New Guinean and Australian sweetpotato cultivars were studied, using spectrophotometry and high performance liquid chromatography (HPLC). The cultivars differed ($p<0.05$) in their total carotenoid and β-carotene contents, and the Original Beauregard cultivar had the highest total carotenoid and β-carotene contents among the cultivars. The spectrophotometry (84-1720 µg/g solids) method generally over-estimated the total carotenoid content compared to the more specific HPLC (23-355 µg/g solids) method. Extrusion significantly ($p<0.05$) decreased the ΔL(*) Hunter colour values, while the Δa(*), Δb(*), total colour change (ΔE), chroma (CR), and browning indices (BI) increased. With the extruder and screw configuration used, extrusion at 40% moisture and 300 rpm screw speed retained carotenoid maximally at more than 80%. This study reports, for the first time, carotenoids of flours from south Pacific sweetpotato cultivars, and carotenoid retention during extrusion.

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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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