Malaria control: generating evidence from local to global level

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Prof. Dr. Christian Lengeler, Prof. Dr. Marcel Tanner und Prof. Dr. Umberto d' Alessandro

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Prof. Dr. Eberhard Parlow
Dekan der Philosophisch-Naturwissenschaftlichen Fakultät

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List of abbrevations

ACT	Artemisinin-Based Combination Therapy
CBA	Controlled before-and-after study
CC	Cochrane Collaboration
CDC	Commonwealth Development Corporation
CENTRAL	Cochrane Central Register of Controlled Trials
CI	Confidence Interval
DDT	Dichlorodiphenyltrichloroethane
Deff	Design effect
DRSTP	Democratic Republic of São Tomé and Príncipe
EC	Emulsion Concentrate
ELISA	Enzyme-linked immunosorbent assays
EPI	Expanded Program on Immunization
Hb	Haemoglobin
HOP	Higaturu Oil Palms
ICC	Intra-cluster Correlation Coefficient
IPTi	Intermittent Preventive Treatment for Infants
IPTp	Intermittent Preventive Treatment during pregnancy
IRS	Indoor Residual Spraying
ITN	Insecticide Treated Net
ITS	Interrupted Time Series
LBW	Low Birth Weight
LLIN	Long-Lasting Insecticidal Net
LSDI	Lubombo Spatial Development Initiative

MMV	Medicines for Malaria Venture
MRAC	Medical Research Advisory Committee
NMCP	National Malaria Control Programme
PCR-RFLP	Polymerase Chain Reaction-Restriction Fragment Length Polymorphism
Pf	*Plasmodium falciparum*
Pm	*Plasmodium malariae*
PMI	(United States) President's Malaria Initiative
PPP	Public-Private Partnership
Pv	*Plasmodium vivax*
RBM	Roll Back Malaria
RCT	Randomised Controlled Trial
RDT	Rapid Diagnostic Test
RR	Risk Ratio
SC	Suspension Concentrate
TDR	Special Programme for Research and Training in Tropical Diseases
UNICEF	United Nations Children's Fund
WBC	White Blood Cell
WHO	World Health Organization
WMD	Weighted Mean Difference
WP	Wettable Powder

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Summary

In addition of the provision of effective treatment to each case, malaria control is heavily relying on vector control with either insecticide treated mosquito nets (ITNs) or indoor residual spraying (IRS). The effectiveness of ITNs in controlling malaria in many different settings has already been comprehensively documented. On the other hand, while IRS has a long and distinguished history in malaria control, its health effects have never been properly quantified.

The present thesis aimed at generating malaria control knowledge on local and global level. On a local level, new insights into the malaria burden in South-East PNG were generated. The work was carried out within an oil palm plantation, which allowed exploring options for malaria control, together with the management of the company. On a global level, this thesis aimed to quantify the effectiveness of IRS in reducing ill-health from malaria through a Cochrane review and to compare IRS to ITNs.

Local level – malaria in Papua New Guinea

To get an overview on malaria epidemiology within the Higaturu Oil Palms plantation (HOP) in Oro Province in PNG, different observational study approaches were used. In 2006, we carried out a cross-sectional study within six company villages, which included the determination of parasite rates by conventional microscopy, interviews and haemoglobin measurements. Passive surveillance data were collected from the 13 company aid posts for the years 2005 and 2006. Before the start of this study, malaria diagnosis was relying on clinical symptoms only. Since malaria symptoms are unspecific, we introduced rapid diagnostic tests (RDTs) in all aid posts. Finally, entomological data were collected by human landing catches.

Prevalence of malaria was high, with more than a third of the participants (33.5%, 95% confidence interval (CI): 30.1-37.0) found with a malaria positive blood slide. Overall, prevalence was highest in the age group 5-9 years (40.3%, 95% CI 0.32-0.49). More than half of the infections were caused by *Plasmodium falciparum* (59.5%), followed by *P. vivax* with 37.6% and *P. malariae* (6.6%). Haemoglobin levels were low, with a mean of 11.0 g/dl (95% CI 10.8-11.1) for men and 10.4 g/dl (95% CI 10.3-10.5) for women. *Plasmodium falciparum* infections were significantly associated with anaemia (Hb < 10 g/dl). At the aid posts, all malaria cases in 2005 and January-March 2006 were diagnosed by symptoms only, while from April 2006 onwards most cases were tested by rapid diagnostic tests. Between 2005 and 2006, 22,023 malaria cases were diagnosed at the aid posts and malaria

accounted for 30-40% of all clinical cases. Of the malaria cases, 13-20% were HOP employees. On average, an employee sick with malaria was absent for 1.8 days, resulting in a total of 9,313 workdays lost between 2005 and 2006. Sleeping outside of the house did not increase the risk for a malaria infection, neither did getting up before 7am. *Anopheles punctulatus* was the main vector founding the area.

Malaria was found to be a major health burden in the Higaturu Oil Palm plantation, posing a high risk to company staff and their relatives, including expatriates and other non-immune workers. Reducing the malaria risk is a highly recommended investment for the company.

Global level

The health effects of IRS were summarized and quantified in the frame of a Cochrane review. Studies considered for the review had to be either Randomized Controlled Trials (RCTs), Controlled Before-and After studies (CBA), or Interrupted Time Series (ITS). They had to include children and adults living in malarious areas and be carried out with one of the World Health Organization (WHO) recommended insecticides.

There was a great paucity of high-quality evidence. Only six out of 132 identified studies met the inclusion criteria (four RCTs, one CBA and one ITS) and not all key malariological outcomes were addressed within these studies. Also, the geographic spread of the included studies was limited.

For stable malaria settings, malaria incidence was significantly reduced in children aged one to five years (RR 0.86, 95% CI 0.77 to 0.95), while no difference was seen for children older than five years. For malaria prevalence no difference was seen between the IRS and the control group. With regard to anaemia, the haemoglobin levels were significantly lower in the control group than in the IRS group (WMD 0.61 g/dl; 95% CI IRS group 9.99 to 10.02; 95% CI no IRS group: 9.38 to 9.40).

When comparing IRS to ITNs, IRS showed a better protective effect in reducing malaria incidence for children aged one to five years (RR 0.88, 95% CI 0.78 to 0.98). No difference was seen for children older than five years. Prevalence rates were found to be equal within the IRS and ITN groups. No difference in haemoglobin levels was found (WMD 0.01; 95% CI IRS: 9.99 to 10.02; 95% CI ITN: 9.99 to 10.01).

In regard to unstable malaria settings IRS was shown to significantly reduce the incidence rate of malaria infections with a protective efficacy ranging from 24% to 86%. IRS also

significantly reduced the incidence of malaria when looking separately at *P.falciparum* and *P. vivax (P.falciparum:* RR 0.07, 95% CI 0.02 to 0.39; *P. vivax:* RR 0.21, CI 95% 0.10 to 0.55). Different results were seen when assessing the impact of IRS on malaria prevalence. No effect of IRS in reducing malaria prevalence was found in India. In children aged five to fifteen in Pakistan, IRS reduced the risk of getting infected with *P. falciparum* as well as with *P.vivax* - by 90% and 68%.

Conflicting results were seen comparing IRS to ITNs against malaria incidence, with one study showing a better protection with ITNs (RR 1.55, 95% CI 1.49 to 1.60) and one study detecting no difference between the two interventions.

Unfortunately, the aim of the review (to quantify the health effects of IRS) could not be achieved. A major conclusion of this work is the urgent need for high-quality evidence from two or three-arm RCTs. Ideally such trials should have one IRS arm, one ITN arm and an arm combining both interventions at high coverage. A control arm should not be planned for ethical reasons. This evidence will be crucial to support the long-term aim of malaria elimination/eradication.

Zusammenfassung

Zusätzlich zu einer wirksamen Behandlung im Falle einer Malaria Erkrankung stützt sich die Malariakontrolle auf die Bekämpfung des Vektors mit dem Gebrauch von insektizidbehandelten Mückennetzen (ITNs) und dem Besprühen von Wänden im Innern von Häusern mit Insektizid (Indoor Residual Spraying, IRS). Während die Wirksamkeit von ITNs, die Malaria unter verschiedensten Bedingungen zu kontrollieren, bereits ausführlich dokumentiert wurde, fehlt dieses Wissen noch immer für IRS. Obwohl IRS in der Malariakontrolle eine lange und bedeutende Rolle hat, wurde der Einfluss des Sprayens auf die Gesundheit noch nie richtig dokumentiert.

Ziel dieser Arbeit war es, den Wissensstand der Malariakontrolle sowohl auf lokaler als auch auf globaler Ebene zu erweitern. Auf lokaler Ebene konnten wir neue Erkenntnisse über die Malaria-Bürde im Südosten von Papua Neuguinea generieren. Die Arbeit wurde innerhalb einer Ölpalmenplantage ausgeführt, was es uns ermöglich hat, zusammen mit dem Management der Firma verschiedene Möglichkeiten der Malariakontrolle anzuschauen. Auf globaler Ebene hatten wir das Ziel mit Hilfe einer Cochrane Review den Effekt von IRS auf die Malaria bedingte Morbidität zu quantifizieren und mit ITNs zu vergleichen.

Lokale Ebene - Malaria in Papua Neuguinea

Mit Hilfe von verschiedenen beobachtenden Studiendesigns konnte ein Überblick über die Malaria-Epidemiologie innerhalb der Higaturu Oil Palms Plantagen (HOP) gewonnen werden. In 2006 haben wir in sechs verschiedenen Firmen-Dörfern eine Querschnittsstudie durchgeführt, in welcher wir Daten über die Parasitenrate gesammelt, Interviews durchgeführt und den Hämoglobinlevel gemessen haben. In den Jahren 2005 und 2006 wurde mithilfe von passiver Überwachung Daten von 13 Firmenkliniken gesammelt. Vor dem Beginn der Studie wurde Malaria anhand von klinischer Symptome diagnostiziert. Da diese Symptome aber sehr unspezifisch sind, haben wir Schnelltest für die Malariadiagnose (RDTs) eingeführt. Zu guter Letzt wurden entomologische Daten mithilfe von „human landing catches“ gesammelt.

Die Malariaprävalenz war hoch. Insgesamt wurde bei mehr als einem Drittel der Teilnehmer (33.5%, 95% CI 30.1 -37.0) Malariaparasiten im Blut entdeckt. Mit einer Prävalenz von 40.3% (95% CI 0.32-0.49) waren die 5-9 jährigen Kinder am meisten von der Malaria betroffen. Über die Hälfte der Infektionen wurden durch *Plasmodium falciparum* verursacht (59.5%), gefolgt von *P. vivax* mit 37.6% und *P. malariae* mit 6.6%. Die Hämoglobinlevels

waren tief mit einem Durchschnittswert von 11.0 g/dl (95% CI 10.8-11.1) für Männer und 10.4g/dl (95% CI 10.3-10.5) für Frauen. Es wurde ein signifikanter Zusammenhang zwischen *P. falciparum* und Anämie (Hb < 10g/dl) gefunden. Während in den Kliniken im Jahr 2005 und von Januar bis März im 2006 alle Malariafälle aufgrund von klinischen Symptomen diagnostiziert worden sind, wurden ab April 2006 RDTs benutzt. In den Kliniken wurden von 2005 bis 2006 22'023 Malariafälle diagnostiziert, was 30-40% von allen klinischen Fällen ausmachte. 13-20% der Malariafälle waren Angestellte von HOP. Innerhalb den drei Monaten in 2006 in welchen noch keine RDTs benutzt wurden, wurden monatlich 1'220 Malariafälle diagnostiziert. Nach der Einführung von RDTs in April wurden bis Ende 2006 monatlich nur noch 698 Malariafälle diagnostiziert. Ein erkrankter Arbeiter fehlte durchschnittlich 1.8 Tage pro Malariaepisode. Dies führte in den Jahren 2005 und 2006 zu 9'313 verlorenen Arbeitstagen für die Firma. Leute die im Freien schliefen oder vor sieben Uhr morgens aufstanden, hatten kein erhöhtes Risiko an Malaria zu erkranken. *Anopheles punctulatus* wurde als wichtigster Vektor in diesem Gebiet identifiziert.

Malaria wurde als eine bedeutende Gesundheitsbürde für die Higaturu Oil Palm Plantage identifiziert, welche ein grosses Risiko sowohl für die Angestellten als auch deren Angehörigen, inklusive andere nicht-immune Arbeiter, darstellt. Eine Investition in die Reduktion des Malariarisikos wird der Firma stark empfohlen.

Globales Level

Die Auswirkungen von IRS auf die Gesundheit wurden im Rahmen einer Cochrane Review zusammengefasst und quantifiziert. Nur randomisierte Kontrollstudien (RCT) sowie kontrollierte Vor-und-Nachher-Studien (CBA) und unterbrochene Zeitstudien (interrupted time series, ITS) wurden für die Studie berücksichtigt. Die Studien mussten sowohl Kinder und Erwachsene aus einem Malariabetroffenen Gebiet beinhalten als auch mit einem Insektizid durchgeführt worden sein, welches von der Weltgesundheitsorganisation (WHO) empfohlen wird.

Es wurde ein grosser Mangel an hochwertiger Evidenz festgestellt. Von 132 identifizierten genügten nur sechs Studien den Einschlusskriterien (vier RCTs, ein CBA und ein ITS) und nicht alle malariologisch-relevanten Resultate wurden angesprochen. Die geographische Ausbreitung der Studien war ebenfalls sehr limitiert.

In den Gebieten mit stabiler Malaria wurde die Malaria-Inzidenz in ein- bis fünfjährigen Kindern signifikant reduziert (RR 0.86, 95% CI 0.77 to 0.95). Für Kinder über fünf Jahre wurde kein Effekt festgestellt. Im Bezug auf die Prävalenz wurde kein Unterschied zwischen

der Gruppe mit IRS und der Kontrollgruppe festgestellt. Der Hämoglobinlevel in der Gruppe mit IRS war signifikant tiefer im Vergleich zu der Gruppe ohne IRS (WMD 0.61 g/dl; 95% CI IRS group 9.99 to 10.02; 95% CI no IRS group: 9.38 to 9.40).

Bei dem Vergleich von IRS mit ITNs erzielte IRS bei der Reduktion der Malaria-Inzidenz bei ein- bis fünfjährigen Kindern ein besseres Resultat als ITNs (RR 0.88, 95% CI 0.78 to 0.98). Für Kinder über fünf Jahre wurde hingegen kein Unterschied festgestellt. Die Prävalenz-Raten zwischen IRS und ITN Gruppen waren gleich. Es wurde kein Unterschied zwischen den Hämoglobinlevels gefunden (WMD 0.01; 95% CI IRS: 9.99 to 10.02; 95% CI ITN: 9.99 to 10.01).

In den Gebieten mit unstabiler Malaria reduzierte IRS die Inzidenz-Rate signifikant und erreichte eine „protective efficacy" zwischen 24% und 86%. Auch wenn man die Spezies *P. falciparum* und *P.vivax* separat betrachtet, wird die Inzidenz-Rate signifikant durch IRS reduziert (*P.falciparum:* RR 0.07, 95% CI 0.02 to 0.39; *P. vivax:* RR 0.21, CI 95% 0.10 to 0.55).

Beim Betrachten des Einflusses auf die Prävalenz wurden unterschiedliche Resultate vorgefunden. In Indien wurde durch IRS keine Reduktion der Prävalenz festgestellt. In Pakistan hingegen wurde für ein- bis fünfjährige Kinder sowohl das Infektionsrisiko für *P. falciparum*- als auch für *P. vivax* um 90%, respektive 68% reduziert.

Im Bezug auf die Inzidenz wurden bei dem Vergleich von IRS mit ITNs widersprüchliche Daten gefunden. Während eine Studie einen besseren Schutz durch ITNs aufzeigte (RR 1.55, 95% CI 1.49 to 1.60), fand eine andere Studie keinen Unterschied zwischen den Interventionen.

Unglücklicherweise wurde das Ziel der Review (den Effekt von IRS auf die Malaria bedingte Morbidität zu quantifizieren) nicht erreicht. Eine wichtige Schlussfolgerung dieser Arbeit ist, dass dringend qualitativ hochstehende Evidenz benötigt wird, welche durch zwei- oder drei-armige RCTs gewonnen werden sollte. Idealerweise sollten diese Studien aus einem Arm mit IRS, einem mit ITN und einem mit beiden Interventionen zusammen bestehen. Dabei sollte bei den Interventionen einen hohen Deckungsgrad erreicht werden. Aus ethischen Gründen sollte auf einen Kontrollarm verzichtet werden. Solche Evidenz wird entscheidend dafür sein, das langfristige Ziel der Malaria Elimination/Eradikation zu unterstützen.

1 Introduction

1.1 Malaria: Global burden, geographical distribution and its causative organism

Malaria is the pre-eminent parasitic infection in humans and about half of the world's population is at risk of getting infected (Greenwood *et al.* 2005; WHO & UNICEF 2008). Obtaining exact measures on the morbidity and mortality due to malaria is difficult (de Savigny & Binka 2004). Frequently, data are derived from health facilities, but this source is often of poor quality. For example in Africa, most deaths occur at home, without patients having seen a health facility before their death. Also geographical and physical access, socioeconomic status, sex, age, belief systems, quality and availability of health services, can influence the decision of a patient whether to visit a health facility or not (de Savigny & Binka 2004; Obrist *et al.* 2007). But also co-infections with other diseases or indirect contributors of mortality such as anaemia or malnutrition complicate the estimation of the real burden. Nevertheless, many attempts have been made and are still ongoing to produce more reliable burden of disease estimate, combining epidemiological, geographical and demographical data (Greenwood *et al.* 2005). These estimates suggest that yearly between 247 – 500 million people become ill and nearly one million people die from malaria (WHO & UNICEF 2008; Greenwood *et al.* 2008; Lewison & Srivastava 2008). This makes malaria one of the leading global killers, with only 13 diseases or injuries causing more deaths (World Health Organization 2004; Roll Back Malaria 2008b).

In areas of high malaria transmission, pregnant women and children under five suffer most from malaria (World Health Organization 2004). Pregnant women infected with malaria have a higher risk of developing severe anaemia and the percentage of malaria-related maternal deaths range from 0.5% to 23% (Desai *et al.* 2007). The risk of a baby to be born with low birth weight (LBW) is considerable higher in women with placental malaria and LBW is associated with a clear increase in infant mortality (Desai *et al.* 2007). More than 75% of all deaths by malaria occur in children (Breman 2001), mainly resulting from cerebral malaria and anaemia. And of those who survive severe malaria, up to 20% experience neurological sequelae including behavioural disorders, as well as other sequelae (Sachs & Malaney 2002). This is also hampering the economics of malaria endemic countries (for more details see chapter 1.6).

Malaria is present in 109 countries, mainly in the tropics and subtropics (Figure 1) (WHO & UNICEF 2008). Malaria transmission does not occur at temperatures below 16° C or above 33° C and at altitudes greater than 2000 m above se a level because the development of the

parasite within the mosquito can not take place (Cook & Zumla 2003). Sub-Saharan Africa bears the major disease burden and the risk of dying from malaria is considerable higher than in other parts of the world (WHO & UNICEF 2008). Ecology and the climate are the main factors defining the distribution of malaria. However, there are other factors which also play an important role such as the demography as well as the socio-economic and cultural characteristic of the population, the predominant vectors and parasites and their ability to mount resistance against anti-malarial drugs and insecticides, the functioning of the public health systems and finally the presence of control programmes.

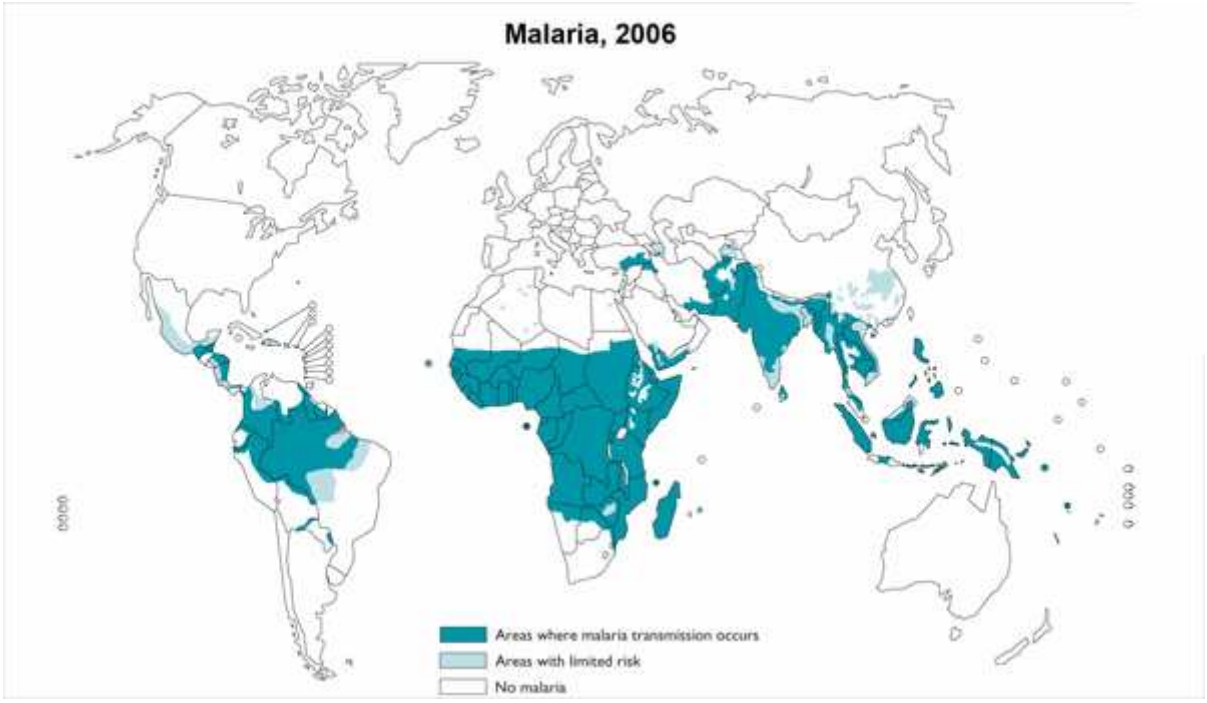

Figure 1: Geographical distribution of malaria in 2006 (Source: World Health Organisation: http://www.who.int/malaria/malariaendemiccountries.html)

1.2 Malaria – the parasite

Malaria is caused by protozoan parasites of the genus *Plasmodium* (Warrell & Gilles 2002). Of the four *Plasmodium* species that cause malaria in humans, *Plasmodium falciparum* is the most virulent one, responsible for the most deaths from malaria (Greenwood *et al.* 2008). *P. vivax* is less deadly, but it is also recognised to be responsible for a substantial health burden (Picot 2006; Genton *et al.* 2008; Karyana *et al.* 2008; Greenwood *et al.* 2008; Greenwood 2008). *P. ovale* and *P. malariae* are thought to cause relatively mild infections (Mueller *et al.* 2007). In some areas, for example Malaysia, a fifth species, *Plasmodium knowlesi*, also infects humans although it is primarily a parasite of the long-tailed macaque monkeys (Greenwood *et al.* 2008).

Malaria gets transmitted to humans when they are bitten by an infectious *Anopheles* mosquito, releasing the infective form of the parasites (sporozoites) into the human body (Figure 2). Within humans, the parasite enters the blood stream and then rapidly invades the liver, where it multiplies and transforms into merozoites. The merozoites then invade red blood cells. Within the blood stages, merozoites either undergo repeated cycles of multiplication or transform into gametocytes, which are taken up by a female *Anopheles* mosquito. Within the mosquito, the parasite undergoes further transformation and the sexual replication takes place. After approximately two weeks the mosquito becomes infectious for the humans and the cycle repeats itself (Warrell & Gilles 2002; Greenwood *et al.* 2008).

The life cycle of *Plasmodium* as it was just described has some specifics for each of the four species. During the blood stage of an infection, *P. falciparum* is able to stick to the endothelium and hence to be sequestered in internal organs, including the brain. It further needs a higher environmental temperature to develop within the mosquito than *P. vivax*, which explains the wider distribution of *P. vivax* compared to *P. falciparum*. *P.vivax* and *P. ovale* are able to remain dormant for months within the liver through the production of hypnozoites, which makes their infection difficult to eliminate. The ability to remain for years in the blood in a very low density is a feature of *Plasmodium malariae* (Greenwood *et al.* 2008).

The length of the erythrocytic cycle varies with the different species. It takes 48 hours in *P. falciparum* and *P. vivax* infections and 72 hours in *P. malariae* infections. The characteristic fever therefore occurs every third, respectively fourth day (Warrell & Gilles 2002).

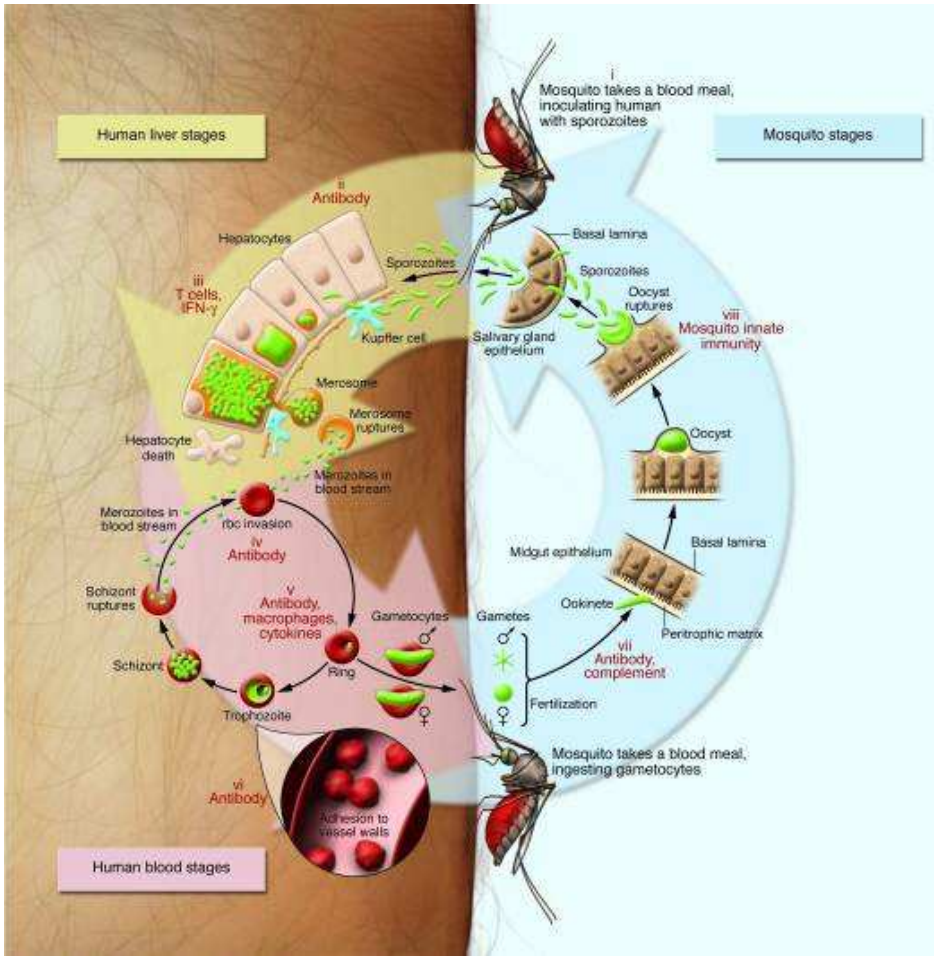

Figure 2: Life cycle of malaria-causing Plasmodium parasites in humans and the mosquito (Source: Greenwood et al. 2008)

1.3 Malaria in PNG

Papua New Guinea is characterized by its variability and complexity in culture, ecology and geography. This complexity is also reflected in the malaria situation. All four human pathogen malaria parasites are found. *Plasmodium falciparum* is the dominant species, followed by *P. vivax*, *P. malariae* and *P. ovale* (Muller *et al.* 2003).

After pneumonia, malaria ranks second amongst the main diseases that cause death in Papua New Guinea (Genton *et al.* 2003) and 90% of all people are at risk (RBM Partnership & WHO 2000). But there is a great variation in the relative importance of malaria in different areas of the country. The epidemiology of malaria in PNG ranges from complete absence of malaria through unstable low levels of transmission with recurring epidemics, to permanently high levels of transmission (Schuurkamp 1992; Genton *et al.* 1995a; Mueller *et al.* 2002), even reaching the highest transmission levels known outside of Africa. About 46% of all Papua New Guineans live in an altitude zone of 0 – 600 m (Muller *et al.* 2003), where malaria

is found to be highly endemic. Variation in the malaria epidemiology is not only seen between different regions but also between villages and even between clusters of houses. The variations in endemicity are mainly due to environmental differences (Genton *et al.* 1995a; Muller *et al.* 2003). But there are several other factors that also play a role in the malaria distribution. These include primarily drug and bednet usage, nutritional factors, and also migration (Muller *et al.* 2003).

PNG has a highly diverse Anopheline mosquito fauna. The group of mosquitoes which is mainly responsible for the transmission of malaria is the *Anopheles punctulatus* complex. This group consists of at least 11 species (Muller *et al.* 2003). Members that have been identified in PNG are *A. punctulatus*, *A. koliensis*, *A. farauti* s.s., *A. farauti* 2, *A. farauti* 3, *A. farauti* 4-6 and *A.* sp. near *punctulatus* (Cooper *et al.* 2002). The different species have a diverse distribution throughout the country and are known to differ significantly in their biting patterns (early vs. late, indoor vs outdoor biting), their larval habitats as well as in their vectorial capacity. These differences have an important impact on malaria control programs since (1) mosquito nets do not protect people so well against mosquitoes which bite before they go indoors, and (2) indoor residual spraying protects less against outdoor biters. Therefore, it might be necessary to adapt vector control programs for each region.

1.4 Malaria control

For a successful malaria control, a solid understanding of the local malaria epidemiology, as well as the social and economic circumstances is needed (Warrell & Gilles 2002).

With targeted vector control, changes in land use, agricultural practices and quality house construction, malaria could be eliminated from the United States and most of Europe during the first half of the twentieth century (Greenwood & Mutabingwa 2002). In 1939 the highly effective residual insecticide dichlordiphenyltrichlorethane (DDT) was developed, which gave hope of eradicating malaria. With the dual strategy of DDT spraying and choloroquine treatment, the World Health Organization carried out the global eradication programme in the 1950s and 1960s. It was initially very successful in many countries such as India, Sri Lanka and the former Soviet Union. But in highly endemic areas, above all in Africa, the eradication programme was never implemented and the situation remained as bad as before (Greenwood & Mutabingwa 2002). The high costs of the programme, resistance of many communities to repeated spraying of their houses and the emergence of DDT resistance led to programme failures. In 1969 the strategy of malaria eradication was officially abandoned. The malaria eradication program was a big success in that it freed over 30 countries from malaria and the risk of malaria was removed from about 20% of the world' population. However, it clearly did not come anywhere close to eradication of the disease and hence it

was unfortunately considered widely to be a failure (Greenwood 2008). This led to a neglect of malaria research and control from the early 1970s to the late 1990s. As an example, between 1975 and 1996 only 3 out of 1223 new drugs were antimalarials (Greenwood & Mutabingwa 2002), even though in the late 1970s the method of *in vitro* cultivation for *Plasmodium falciparum* was developed.

In 1992 the process of malaria control was re-started with the ministerial conference in Amsterdam. In 1998, the Roll Back Malaria partnership was launched with the aim of halving the deaths from malaria by 2010 (Yamey 2004). This was supported by the summit of African Heads of State in Abuja, Nigeria in 2000, where African leaders declared to ensure that by the year 2005:

- at least 60% of those suffering from malaria have prompt access to, and are able to correctly use, affordable and appropriate treatment within 24 hours of the onset of symptoms
- at least 60% of those at risk of malaria, particularly children under five years of age and pregnant women, will benefit from the most suitable combination of personal and community measures such as insecticide treated mosquito nets and other interventions and
- at least 60% of all pregnant women who are at risk of malaria have access to chemoprophylaxis or presumptive intermittent treatment (Roll Back Malaria & World Health Organization 2000).

In 2005, the target coverage was changed from 60% to 80% by 2010, even though many countries had not met the 2005 targets (Yamey 2004; WHO & UNICEF 2008).

At present, malaria research and control is high on the donors' agenda. The global financial investment into malaria control increased to over 2 billions per year in 2008, the main sources being the Global Fund to fight AIDS, Tuberculosis and Malaria (GFATM) as well as the United States President's Malaria Initiative (PMI) and the World Bank's Booster Program (World Health Organization 2008). However, the estimated global cost to control and eliminate malaria is much higher with an estimated average of US$ 5.9 billion needed per year from 2011 to 2020 (Roll Back Malaria 2008a).

Since the failure of the world eradication program in the 1960s, the dominant strategy was to control malaria, and eradication of malaria has never been considered again until October 2007, when Bill and Melinda Gates called for a new malaria eradication campaign. They claimed that new scientific advances and growing financial and political support for malaria

initiatives had made the goal of eradication realistic again (Anonymous 2007). This was also taken up by the World Health Organization (WHO), with its current objectives of reducing the disease burden and maintaining it at a low level, to eliminating the disease from a defined geographical area and, finally to eradicate the disease globally (World Health Organization 2008). However, the feasibility of eradicating malaria has been doubted by malaria experts (Roberts & Enserink 2007; Greenwood 2008; Tanner & deSavigny D. 2008). There is consensus that elimination in stable malaria transmission settings with the existing tools is impossible at present, due to lacking tools, weak health systems and insufficient knowledge on malaria transmission heterogeneity. Even if elimination was achieved in some areas, the possibility of reintroduction would be a constant threat, requiring highly effective surveillance and monitoring systems (Greenwood 2008; Tanner & deSavigny D. 2008). Elimination is most likely to be achieved currently in areas of low and unstable transmission or on islands (Greenwood 2008).

The current tools with which malaria should be controlled (and eventually eradicated) are the following (Roll Back Malaria 2008b):

1) Early diagnosis and prompt and effective treatment of malarial illness

Without treatment, acute mild episodes of malaria in non-immune and semi-immune people can progress rapidly into the severe and fatal forms of the disease. Therefore, identifying potential malaria cases early and treating them promptly with an efficacious drug remains the first and foremost intervention to reduce disease and death from malaria. By eliminating parasite reservoirs in the general population and by decreasing the duration of illness, prompt diagnosis and treatment also helps to reduce malaria transmission (Alilio *et al.* 2004).

Microscopy is the most widely used routine method for malaria diagnosis and is considered as the gold standard (Shrinivasan *et al.* 2000). It enables to specify and quantify the parasites seen in thick and thin blood smears. But, it requires a special expertise and an organized health system infrastructure (Duffy & Fried 2005; Bell *et al.* 2006). In many countries malaria diagnosis is purely made on a symptomatic basis as the laboratory equipment is insufficient (Hommel 2002). This leads to a large over-treatment, as symptoms of malaria are non-specific and similar to those of other infections like influenza, pneumonia, viral hepatitis or typhoid fever (Hommel 2002). It also leads to the under-treatment of other potentially fatal conditions, such as pneumonia.

A potential alternative to microscopy is the use of rapid diagnostic tests (RDTs). RDTs have the advantage to be quick, easy to perform and interpret and recent reviews have shown,

that their performance is comparable to that of expert microscopy (WHO 2000; Moody 2002; Soto *et al.* 2004; D'Acremont *et al.* 2009).

Current guidelines recommend that all fever episodes in African children should be treated presumptively with antimalarial drugs (D'Acremont *et al.* 2009). However, this is currently debated (D'Acremont *et al.* 2009; English *et al.* 2009) and there is still a long way to go towards universal testing of all fever episodes in malaria-endemic zones.

2) Vector control interventions

Indoor Residual Spraying (IRS): IRS has a long and distinguished history in malaria control. Using mainly DDT, malaria was eliminated or greatly reduced as a public health problem in Asia, Russia, Europe, and Latin America (Shiff 2002; Lengeler & Sharp 2003; Roberts *et al.* 2004). IRS continues to be used in many parts of the world, with the services provided by the public health system or by a commercial company (usually for the benefit of its employees). There is no IRS programme known to us in which beneficiaries were expected to contribute financially. A historical review of IRS in Southern Africa investigated the malaria situation before and after the introduction of IRS in South Africa, Swaziland, Namibia, Zimbabwe, and Mozambique, where it continues to protect 13 million people (Mabaso *et al.* 2004). Immediately after the implementation of control operations, spectacular reductions in malaria and vector densities were recorded, malaria endemicity was reduced, and in certain instances the intervention led to local elimination. Another historical paper reviewed the health impacts of 36 successful IRS programmes in 19 countries throughout sub-Saharan Africa (Kouznetsov 1977). The analyses compared parasite rates and other malariological outcomes before and after the implementation of IRS in each of the five major eco-epidemiological zones.

IRS operates both through repelling mosquitoes from entering houses and by killing female mosquitoes that are resting inside houses after having taken a blood meal. This implies that IRS is most effective against mosquito species that are resting indoors (so called endophilic mosquitoes). Spraying needs to be carried out between once and three times per year; the timing depends on the insecticide and the seasonality of transmission in a given setting. Reviewing the advantages and disadvantages of each insecticide is beyond the scope of this review and can be found in Najera (2001). IRS has the advantage of being able to make use of a much wider range of insecticide products in comparison to Insecticide-Treated Nets (ITNs) for which pyrethroids are the only class of insecticide currently used. The World Health Organization recommends a number of insecticides for IRS: DDT wettable powder (WP); malathion WP; fenitrothion WP; pirimiphos-methyl WP and emulsifiable concentrate

(EC); bendiocarb WP; propoxur WP; alpha-cypermethrin WP & SC; cyfluthrin WP; deltamethrin WP; etofenprox WP; and lambda-cyhalothrin WP (WHOPES 2007). This range of insecticides has important implications for the management of insecticide resistance and hence long-term sustainability (pyrethroid resistance has already been reported in many parts of tropical Africa and other parts of the world among populations of the major malaria vectors).

Insecticide-treated nets (ITN): Mosquito nets represent a physical barrier between humans and mosquitoes. Insecticides have either a repellent effect or kill the mosquito on contact (or both). Thus, mosquitoes are prevented from biting people and are killed if they try to do so (Lengeler & Sharp 2003). ITNs have been shown to be very effective in controlling malaria in many different settings (Lengeler 2004). The use of ITNs has led to a reduction of up to 50 % in the number of fever cases in children in Asia, Africa and Latin America. The anaemia status of children and pregnant women in Africa could also be improved significantly. Most importantly, ITNs could be shown to be very effective in reducing child mortality rates. Overall, ITNs showed a 18% protective efficacy against all-cause child mortality (Lengeler 2004). When coverage exceeded 50%, ITNs could be shown not only to be effective for the people sleeping under nets but also in reducing morbidity and mortality for people sleeping within a 300m distance of the treated net (Binka *et al*. 1998; Hawley *et al.* 2003). Currently, pyrethroids are the only class of insecticides being available for impregnating bed nets and research on new insecticides is urgently needed.

3) Malaria prophylaxis and intermittent preventive treatment during pregnancy

For pregnant women and their newborn children, malaria infections with *P. falciparum* or *P. vivax* pose a substantial risk. In areas of low transmission, malaria can result in stillbirths, spontaneous abortions, or maternal deaths. In highly endemic areas, malaria infections can cause placental parasitaemia and contribute to maternal anaemia, both of which may lead to low birth weight (LBW) and an increased risk of death in the newborn. It is estimated that 5-12% of all LBW are due to malaria in pregnancy (Newman *et al.* 2003).

These risks can be lowered with effective prevention of malaria, with chemoprophylaxis on a weekly base being the method of choice. But this method faces problems such as parasite drug resistance, contra-indications of certain drugs and poor patient compliance. In the search of an alternative, intermittent treatment during pregnancy (IPTp) was proposed. It involves the administration of a full curative-treatment dose of an effective antimalarial drug at predefined intervals during pregnancy. It was suggested as a simple and cost-effective

method in highly malarious areas (Greenwood 2004; Garner & Gulmezoglu 2006) and it is now policy in a number of endemic countries.

4) Surveillance, prediction of, and rapid response to epidemics
Among susceptible populations with low immunity, epidemics often lead to great morbidity and cause high fatality rates in all age groups (Kiszewski & Teklehaimanot 2004; Mueller *et al.* 2009). Therefore, it is paramount to be able to predict epidemics and ensure epidemic preparedness at all levels. For this purpose, it is necessary to establish an early warning system (e.g. based on meteorological data and population movements) which triggers the chain of events leading to an explosive increase in the number of cases. In parallel, countries need to establish emergency- preparedness systems that allow them to react promptly with vector control and treatment activities (Najera 1999).

Malaria vaccine
So far, no vaccine against malaria is available. However, promising results concerning the malaria vaccine RTS,S were published recently from three trials (Aponte *et al.* 2007.; Bejon *et al.* 2008; Abdulla *et al.* 2008). In these phase IIb randomized, controlled and double-blind trials, a protective efficacy against *P. falciparum* infections of 56% for infants was found (Bejon *et al.* 2008). The vaccine was also found to be safe and feasible for integration into the WHO Expanded Program on Immunization (EPI) (Bejon *et al.* 2008; Abdulla *et al.* 2008). The vaccine is now going to be tested within a large, multi-country phase III trial, giving hope that in future time it might be part of an integrated control strategy.

1.5 Malaria control in Papua New Guinea (PNG)

The first known attempt to control malaria in Papua New Guinea (PNG) was carried out as early as 1901 on the north coast by mass treating (immigrant) people with quinine. After the identification of the vectors, local larval control measures were undertaken. However, this was largely done in urban areas and malaria in the indigenous population was regarded as largely beyond control (Spencer 1992).

During the Second World War, much information on malaria epidemiology and parasitology was collected by the Australian and American military. They implemented larval control measures by draining swamp areas, introducing *Gambusia affinis* fish and by chemical applications. In 1943, DDT (dichlorodiphenyltrichloroethane) became available and was widely used during the last phase of the war (Spencer 1992).

During the world malaria eradication period of the World Health Organization (1957-1968), a plan was formulated for an eradication campaign using dieldrin to cover the whole of PNG progressively over a period of 14 years. However, in 1959 the promoters changed the insecticide to DDT to overcome the short residual effect of dieldrin. While successes were achieved, it became evident that eradication with DDT and mass drug administration alone wouldn't be possible. In 1969, control programs aiming at the reduction of malaria incidence replaced eradication. The insecticide was changed to a mix of Malathion and DDT, since bedbugs became resistant to DDT, which was worrying the local inhabitants. Another problem was the damage of the sago-palm roofing by the moth larvae *Herculia nigrivitta* following DDT spraying. The feeding habits of these larvae protected them from DDT, whereas all their predators were killed by the insecticide.

Furthermore, not all control measures were carried out carefully and had success, leading to a lower trust of the public and morale within the control service (Spencer 1992). Outdoor activities such as sago making, gardening, social gatherings and cooking were identified to be associated with being at risk of malaria and were considered to hamper control. By 1978 the malaria control strategy was changed again. The new aims were 1) to reduce malaria mortality and mortality, 2) to reduce the effects of the disease on socioeconomic development and 3) achieve elimination wherever feasible.

Overall, DDT was sprayed for approximately 30 years and various modifications were tried such as shortening of the period between spray rounds, mixing of dieldrin with DDT, distribution of pyrimethaminized salt, or additional mass drug administration. However, spraying was never able to completely interrupt the transmission of malaria in areas with a high degree of malaria endemicity (Spencer 1992). After the cessation of vector control in 1984, easy access to antimalarial drugs, especially chloroquine, became the mainstay of malaria control (Muller *et al.* 2003). Mosquito nets were also used for protection and some of the first studies worldwide on ITNs were carried out in PNG, showing effects on mosquito population as well as and the prevalence and incidence of *P. falciparum* infections in children (Graves *et al.* 1987; Charlwood & Graves 1987; Muller *et al.* 2003).

As a result of control measures, marked changes in the epidemiology of malaria occurred during the last 30 years (Cattani *et al.* 1986; Muller *et al.* 2003; Mueller *et al.* 2005). Before the start of the extensive vector control programmes in 1957, *Plasmodium vivax* was the predominant species followed by *P. falciparum* and *P. malariae* (Cattani *et al.* 1986; Mueller *et al.* 2005). Nowadays, *P. falciparum* is predominant everywhere, in areas covered by the control programme and in areas not covered (Cattani *et al.* 1986; Genton *et al.* 1995b). The

relative increase of *P. falciparum* is thought to have occurred as an immediate consequence of the cessation of spraying and also due to the spread of drug resistance. *P. falciparum* resistance to Chloroquine is very common in PNG. It was first noted in 1976 and there was a great reduction in the effectiveness of chloroquine by the early 1990s. *P. vivax* resistance is also present, but to a much lesser extend (Muller *et al.* 2003). Control measures might also have affected the immune status of the population and caused a shift towards older ages of peak prevalence. However, since the cessation of vector control the peak prevalence has shifted back to younger children (Mueller *et al.* 2005).

Currently, PNG aims to halve the number of deaths and illness caused by malaria between 2001 and 2010. Following strategies were implemented by the National Malaria Control Programme (NMCP): 1) improvement of diagnosis and treatment, 2) implementation of vector control through ITNs, IRS, and – where feasible – environmental modification and 3) information, education and communication. For the vector control the aim is to have by 2010 80% of the population living in endemic areas protected by an ITN, and in the highland regions (prone to epidemics) IRS being conducted annually (WHO & UNICEF 2005). Targets were difficult to achieve before 2003 because of financial constraints. Having received funds from the Global Fund to Fight AIDS, Tuberculosis and Malaria (GFATM) in 2003, the targets of the NMCP were adjusted for 2008: 1) more than 80% of the population in malaria-endemic areas should be consistently using LLINs, 2) over 70% of suspected malaria cases should be laboratory-confirmed by rapid diagnostic tests or microscopy, 3) the case rate should be reduced from 504/100,000 in 2001 to 300/100,000 and 4) the mortality rate should be reduced from 12.8/100,000 in 2001 to 7/100,000. However, until 2007 only 53,500 treated nets were delivered, resulting in a coverage of 23.5%, which is still far away of the target of 80%. Close to 25,000 people were protected by IRS (WHO & UNICEF 2008).

1.6 Malaria control within the private sector

Malaria represents not only a huge health but also a major economic burden. In Africa alone, the estimated direct and indirect costs of malaria exceed US$ 12 billion annually (Breman *et al.* 2004). Malaria negatively influences economic growth, reducing it for example by 1.3% annually in endemic countries between 1965 and 1990. In 1995, the average gross domestic product in non-malarious countries was five times higher than in countries affected with malaria (Gallup & Sachs 2001).

Apart from households, malaria also impacts on private sector companies. The most immediate effect of malaria on a company is its impact on the workforce and the resulting cost of caring for sick employees. Employees who are sick with malaria are not working

efficiently and are likely to take time off to recover. In a worst case, employees die and replacements need to be recruited and trained (Global Health Initiative 2006).
A survey of the World Economic Forum (WEF) of over 8000 business leaders from over 100 countries showed that about three-quarter of them perceived malaria as having an impact on their business, and that about 40% perceived this impact to be severe. It also showed that several major businesses had already taken action against malaria (Global Health Initiative 2006). Within the Global Malaria Action Plan, RBM recognises the private sector as an important partner in scaling up malaria interventions (Roll Back Malaria 2008b). However, even though some successful experiences of malaria control by private companies exist, the literature available on impact and effectiveness is scarce. Yet, in recent years, new initiatives such as the Global Business Coalition (http://www.gbcimpact.org) as well as the Global Health Initiative of the World Economic Forum (http://www.weforum.org/en/initiatives/globalhealth/index.htm) were formed to support and link companies in their fight against malaria. Currently, a review on the engagement and successes of the private sector in malaria control is being carried out at the World Economic Forum (Achoki T., personal communication).

1.7 Systematic reviews and the Cochrane Collaboration

Annually, over two million articles are published in the biomedical literature in over 20'000 journals, leaving policy makers, health care providers and researchers with an unmanageable amount of information. Systematic reviews summarise this overwhelming amount of facts and ascertain whether scientific findings are convincing and can be generalised across populations, settings, and treatment variations (Mulrow 1994; Green 2005). Systematic reviews seek to comprehensively identify all literature on a given topic and review all the evidence is a sound way. Systematic reviews can include a meta-analysis, a statistical approach for merging the results of several studies into a single estimate if certain statistical conditions are met (mainly the homogeneity of the results). This is in contrast to the term "review" which is just the general term for attempts to consolidate the results and conclusions from a number of publications (Sackett *et al.* 1996; Green 2005).

The Cochrane Collaboration (CC) holds a very prominent role within the health sector in producing systematic reviews (www.cochrane.org). The CC was formally formed in 1993 and named after the person who inspired it, Archie Cochrane (a British public health researcher 1909-1988). The CC is an international not-for-profit organisation aiming to improve decision making by preparing, maintaining and promoting systematic reviews of the effects of healthcare interventions (Jadad *et al.* 1998; Clarke 2002). It is financially supported by over 650 organisations, including health service providers, research funding agencies, departments of health, international organizations, universities and industry (Clarke 2002).

Over 15,000 people, mainly review authors, are involved in about 100 countries. Authors work within one of the 51 Cochrane review groups, which provide editorial support, administration and infrastructure. Cochrane Centres provide training, support and methodological advice to the review groups. The Cochrane collaboration includes 11 methods groups, which are dealing with topics such as applicability, reporting bias, statistics, and health economics. In addition, a consumer network exists, helping to promote the interests of users of health care. The main instrument to disseminate the work of the Cochrane collaboration is the Cochrane Library, a database of reviews and other resources available either on a CD-ROM or on the internet (Clarke 2002; Clarke 2007).

Cochrane reviews all have the same format and are written in a highly structured manner (White 2002; Higgins & Green 2008). Unlike other reviews, the Cochrane reviews are reviewed very extensively. The Cochrane editorial group is involved at the beginning and a proposal has to be written and accepted by the Cochrane review group. Cochrane reviews aim to search comprehensively for all evidence available on a particular topic. For this, an extensive search has to be carried out, including grey and unpublished literature, irrespective of the language. In a second step, a quality appraisal of all identified studies has to be done. Once the review is finished and accepted by the Cochrane group, it will be published electronically within the Cochrane library.

Today, Cochrane reviews of interventions are considered to be the gold standard for assessing the effectiveness of health care interventions. This is also expressed by the rising number of reviews written: in 1995, 36 full reviews were completed, rising to 2000 reviews in 2004 (Clarke 2007) and 5676 reviews in January 2009.

Rationale for the current PhD thesis

Despite the fact that Papua New Guinea suffers heavily under the burden of malaria, research on malaria control is largely focused on Africa. Furthermore, it is known that malaria is not only a health, but also an economic burden. Several studies on the epidemiology of malaria were carried out within the research areas of the Institute of Medical Research e.g. around Madang and the Wosera area, whereas other areas of PNG were neglected. To the best of our knowledge, no data is published to-date on the malaria epidemiology within the Popondetta area, Oro province. Furthermore, there is an urgent need for more studies assessing the health as well as the economic risk posed by malaria to companies based in endemic settings and the possibility for such companies to initiate control measures.

In a second part compile all available evidence of the effectiveness of IRS in reducing ill-health from malaria, one of the malaria control tools used in PNG. While the effectiveness of ITNs has already been comprehensively summarised in two Cochrane reviews (one for the general population and one for pregnant women (Lengeler 2004; Gamble *et al.* 2004), no such systematic assessment has been done for IRS. Two reviews outlined the cost and health effects of IRS (Curtis & Mnzava 2001; Lengeler & Sharp 2003), but neither was conducted systematically or assessed the methodological quality of the included studies. Since ITNs and IRS are in many ways similar interventions, more information is needed urgently on their comparative impact. To this effect we conducted a Cochrane review on IRS, following a similar methodology to the one on ITNs.

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2 Aims and objectives

Note: the intention of this work was to assess the benefits and costs of an integrated malaria control program in a large oil palm plantation in Papua New Guinea, focusing on house screening. The work was to be implemented within the Higaturu Oil Palm Plantations (HOPPL), a company of the Commonwealth Development Corporation (CDC). Unfortunately, in 2006, HOPPL was sold to Cargill Corp. Following this change of leadership, HOP set new priorities and beginning of April 2007 the company decided to terminate the house screening intervention that was to be the core of this project.

Hence, the revised objectives of the present PhD thesis were as follows:

1. To provide an overview of the malaria epidemiology within a commercial oil palm plantation in the Oro Province (Papua New Guinea) and to quantify the burden of malaria and its implication for the company.

2. To systematically review the impact of indoor residual spraying (IRS) on key malariological parameters and to compare the relative impacts of IRS and ITNs in the frame of a Cochrane review.

3 Malaria – a major health problem within an oil palm plantation around Popondetta, Papua New Guinea

Bianca Plüss[1*], Ivo Müller[2], Damien Levi[3], Graham King[3], Thomas A Smith[1], Christian Lengeler[1]

[1] Swiss Tropical Institute, P.O. Box, 4002 Basel, Switzerland

[2] Institute of Medical Research, P.O. Box 60, Goroka, Papua New Guinea

[3] CTP (PNG) Ltd., Higaturu Oil Palms, P.O. Box 28, Popondetta, Papua New Guinea

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3.1 Abstract

Background

For companies operating in malaria endemic countries, malaria represents a substantial risk to workers and their dependants, and can lead to significantly reduced worker productivity. This study provides an overview of the malaria epidemiology within an oil palm plantation in Popondetta, south-eastern Papua New Guinea, its implication for the company with its employees and their families and the potential for control.

Methods

In 2006, we carried out a cross-sectional study within six company villages, which included the determination of parasite rates by conventional microscopy, interviews and haemoglobin measurements. Passive surveillance data were collected from the 13 company aid posts for the years 2005 and 2006.

Results

Malaria prevalence was found to be high: all-age prevalence was 33.5% (95% CI 30.1-37.0) in 723 individuals. Plasmodium falciparum was the dominant species, followed by Plasmodium vivax and Plasmodium malariae. Children between five and nine years of age were most affected (40.3%, 95% CI 0.32-0.49). Haemoglobin levels were found to be low; 11.0 g/dl (95% CI 10.8-11.1) for men and 10.4 g/dl (95% CI 10.3-10.5) for women, respectively. Plasmodium falciparum infections were significantly associated with anaemia (Hb < 10 g/dl). At the aid posts, all malaria cases in 2005 and January-March 2006 were diagnosed by symptoms only, while from April 2006 onwards most cases were tested by rapid diagnostic tests. Between 2005 and 2006, 22,023 malaria cases were diagnosed at the aid posts and malaria accounted for 30-40% of all clinical cases. Of the malaria cases, 13-20% were HOP employees. On average, an employee sick with malaria was absent for 1.8 days, resulting in a total of 9,313 workdays lost between 2005 and 2006. Sleeping outside of the house did not increase the risk of a malaria infection, neither did getting up before 7am.

Conclusions

Malaria was found to be a major health burden in the Higaturu Oil Palm plantation, posing a high risk for company staff and their relatives, including expatriates and other non-immune workers. Reducing the malaria risk is a highly recommended investment for the company.

3.2 Background

Papua New Guinea (PNG) is characterized by its variability and complexity in culture, ecology and geography. This complexity is also reflected in the malaria situation (Muller *et al.* 2003). Malaria ranks first amongst the diseases causing illness and death in Papua New Guinea (WHO & UNICEF 2005), although there is a great variation in the relative importance of malaria for people in different areas. The epidemiology of malaria in PNG ranges from complete absence of malaria, through unstable low levels of transmission with recurring epidemics, to permanently high levels of transmission (Genton *et al.* 1995; Mueller *et al.* 2002), even reaching the highest transmission levels known outside of Africa. About 46% of all Papua New Guineans live in an altitude zone of 0 – 600 m above sea level (Muller *et al.* 2003), where malaria is highly endemic.

Malaria represents not only a health, but also an economic burden. In Africa alone, the estimated direct and indirect costs of malaria exceed US$ 12 billion annually (Breman *et al.* 2004). Malaria negatively influences economic growth, reducing it for example by 1.3% annually in endemic countries between 1965 and 1990. In 1995, the average gross domestic product in non-malarious countries was five times higher than in countries affected with malaria (Gallup & Sachs 2001). The most immediate effect of malaria on a company is its impact on the workforce and the resulting cost of caring for sick employees. Employees sick with malaria are not working efficiently and are likely to take time off to recover. In a worst case, employees die and replacements need to be recruited and trained (Global Health Initiative 2006).

In PNG, oil palm is a major agro-industry. In 2001, over 100,000 hectares of oil palms were planted. Since 2000, palm oil has been the most important agricultural export industry in PNG, with nearly 400,000 tons exported in 2002, amounting to approximately 136 million USD (Koczberski *et al.* 2001; FAO 2008). Commercial plantations such as oil palm plantations offer suitable environmental conditions for Anopheles mosquitoes breeding and people living and working in such plantations are thus likely to be at high risk for malaria (Chang *et al.* 1997). However, such plantations also provide relatively good housing and infrastructure for their workers, making it relatively easy to deploy screening or indoor residual spraying, and improvement in case management.

Higaturu Oil Palms plantations (HOP) is one of the major employers in Oro Province (south-east PNG). It provides housing for its employees and their dependants within the plantation. HOP is located in the area around Popondetta, the capital of Oro province, on a coastal plain

at the foot of the Owen-Stanley range. So far, and to the best of our knowledge, no data have ever been collected on the epidemiology of malaria for this part of the country. Therefore, this study aims to provide (1) an overview of the malaria epidemiology in Oro province, and (2) to quantify the problem of malaria within a commercial oil palm plantation, its implications for the company and the potential for control. Few such studies have been carried out and this economic risk (in addition to the health risk) urgently requires to be better quantified.

3.3 Methods

Study site

The study was conducted within the oil palm plantations of CTP (PNG) Ltd. (trading as Higaturu Oil Palms - HOP), close to Popondetta, the capital of Oro Province in Papua New Guinea. HOP processes palm fruits from an area of about 22,997 hectares. The company owns 8,997 hectares which are divided into five estates (namely Javuni, Sumbiripa, Ambogo, Mamba and Embi). Smallholders own another 14,000 hectares. While the smallholders are responsible for maintaining and harvesting the palms, Higaturu buys their fruits and processes them into palm oil and palm kernel oil.
People living in the 14 villages within the plantation consist of HOP employees and their dependants, for whom housing is provided by the company. Every village is equipped with a small health centre, staffed with one Community Health Worker or nursing officer. A bigger health centre with a laboratory is situated in Siroga village, which is easily accessible by car or foot.

The cross-sectional study took place in six villages (namely Epa, Irigi, Sumbiripa, Irihambo, Javuni and Moale), located within three of the five estates. Two of the estates (Mamba and Embi) were excluded as they were difficult to reach due to their geographical distance. Mamba estate is located in the mountains and hence is not ecologically comparable to the other estates. For the passive surveillance system, all aid posts were included.

Ecologically the study villages are very similar to each other, all being surrounded by oil palms. Within the study area, rainfall usually shows some seasonality with a wet season from November to April and a drier season from May/June to October. During our study period the average rainfall was 189 mm, with 243 mm in April and 135 mm in May (Figure 1).

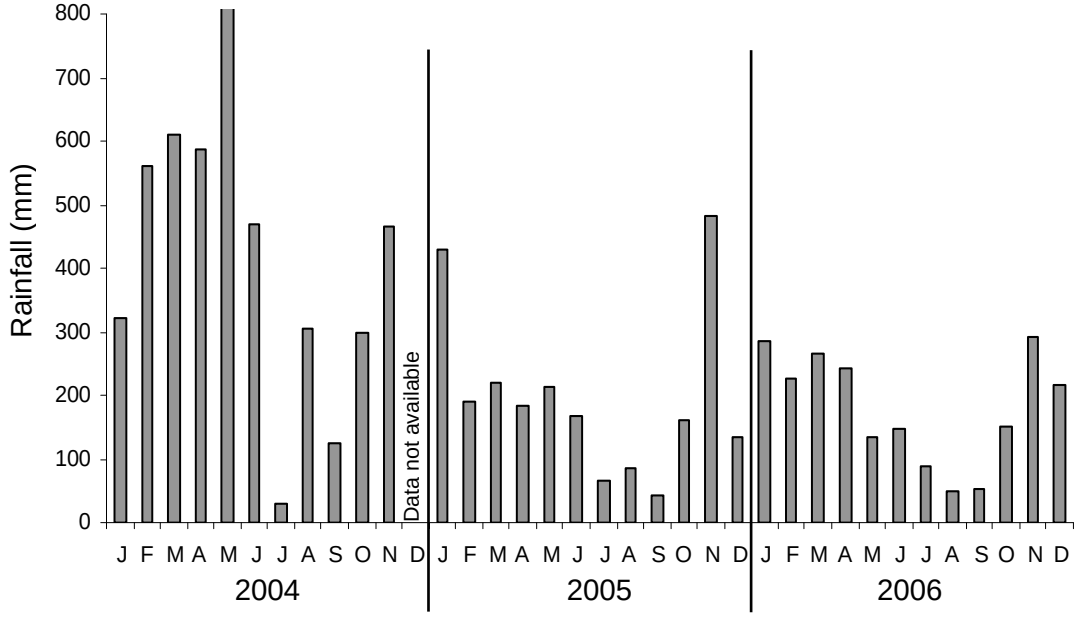

Figure 1: Average monthly rainfall within the study area from 2004 to 2006.

A company village consists of 100 houses of which 87-98% are so-called “labour houses”, wooden two-bedroom houses with a cooking place outside on the veranda and its own ablution block and toilet next to the house (Figure 2).

Figure 2: A typical worker’s house.

Cross-sectional parasitology surveys

The cross-sectional surveys were conducted between April and May 2006. Every village was visited once. Households to be included were selected by random sampling from the existing list of houses provided by the company.

Inclusion/exclusion criteria. Every person living within the selected villages and households was eligible for the study. Consent procedures are described below. Persons ill at the time of the survey, as well as visitors to the household who had lived less than four weeks in the village, were excluded from the study.

Measurements. From each participant we measured the axillary temperature with an electronic thermometer. At the same time, a questionnaire was administered on the use of bed nets, the history of malaria sickness in the previous two weeks, the use of anti-malarial drugs and health facility attendance, travel behaviour, evening leisure activities and sleeping habits (outside or inside their house). The interview was performed in English or Melanesian pidgin by bilingual interviewers. At the same time as the interview, a blood slide including a thick and a thin film was prepared. Thin blood films were fixed with methanol. All smears were stained with 2.5% buffered Giemsa (pH 7.2) for 35 minutes and examined afterwards microscopically for 100 fields under oil immersion (x 1000 magnification) before being declared negative. In positive films, parasite species were identified and densities recorded as the number of parasites/200 white blood cells (WBC). Densities were converted to the number of parasites/µl of blood assuming 8,000 WBC/µl.

Haemoglobin levels were measured from the same finger prick as from the blood slide, using a HaemoCue machine (HemoCue B-Hemoglobin, Angholm, Sweden). From each individual the spleen size was determined according to Hackett's grading by a local nurse.

Routine data assessment

Malaria incidence data routinely collected by the health staff of the HOP aid posts were available from January 2005 to December 2006. Until April 2006, all patients presenting with fever were considered as malaria cases. In April 2006 Rapid Diagnostic Tests (RDTs, ICT Malaria Combo Cassette, R&R Marketing, South Africa) were introduced in all aid posts within the Higaturu Oil Palm estates. Before the introduction of the RDTS, a three-day training course on the theory and usage of the RDTs was provided to every member of the health staff of HOP. After the introduction of RDTs, patients were classified as having malaria

only if the RDT result was positive. Compared to microscopy the RDT sensitivity was 89%, which is excellent.

For every patient visiting a HOP aid post, the date of the visit, data of joining the company (employee, dependant or outsider) and the diagnosis were collected.
In 2006, for an ad hoc sample of employees, the numbers of days missing from work due to malaria were recorded.

Data entry and analysis

The data from the questionnaires were entered in an Access 2002 database (Microsoft Corp., Redmond, US). To check for data entry mistakes, the entered data were checked and corrected by one person, while a second person was reading out the entries of the original questionnaires. The blood slides were read twice by two different microscopists and the results entered into two different Excel databases (Microsoft Corp., Redmond, US). A Kappa test for checking for consistency between the two datasets showed an expected agreement of 78.4% (k=0.43). In case one microscopist only found few parasites (< 3 parasites/200WBC) and the other none, the slides were considered to be positive. Slide results were excluded from the analysis if one microscopist found a high parasites density (>12 parasites/200 WBC) and the second one found none. Differences in categorical variables were tested using the Pearson chi-square test. Differences were regarded as significant if the p-value of the test statistic was ≤5%.

Changes of haemoglobin values were calculated using linear regression, adjusting for age, gender and mosquito net use. Difference between males and females in haemoglobin levels were tested with Wilcoxon test, as the Bartlett's test for the differences in the variance was significant. Participants were classified as having anaemia if haemoglobin levels were under 10 g/dl. For obtaining odds ratios, relevant variables (age, sex, village, sleep outside on the veranda, sleep outside in a shelter, sleeping under a bed net, work and getting up before 7am) were added one-by-one in the model and a likelihood ratio tests was performed to test for significance.
Work days lost for the company were calculated by taking the arithmetic mean of the days not worked per employee due to malaria.

Unfortunately, it was not possible to get the results of RDT testing from all the clinics because these statistics were not available to us. In order to have at least an estimate of testing positivity rate, a sample of 90 successive patients in one clinic (Siroga) who were

clinically diagnosed with malaria were also tested with a RDT in order to calculate the RDT positivity rate.

All statistical analyses were performed in Intercooled Stata 9 (StataCorp, Texas, US).

Informed consent

The community was informed about the aims and the methods of the study before the start of the data recording and sample collection by the company health care workers. Consent was then sought individually from all study participants or their guardians. This was done by explaining again the aim and procedure of the study at the beginning of the interview. A signature of each participant on their understanding and willingness to participate was then collected. All enrolled individuals retained their right to withdraw from the study at any stage.

The survey methodology was approved by the PNG Medical Research Advisory Committee (approval number MRAC No. 06.07).

3.4 Results

Cross-sectional survey: general description

A total of 843 individuals were included in the survey (Table 1). The study participants consisted of 423 (50.2%) females and 419 (49.8%) males and were grouped into the following five age categories: 0-4, 5-9, 10-19, 20-39 and >40 years. There was a big excess of individuals aged 20-39 years (40.9% of the participants), which were made up largely of workers, as expected in this population. Over half of the participants (57.1%) came from Oro province, and overall 81.7% of the people had an Oro, a Morobe or a mixed Oro/Morobe origin. 28.2% (238/843) of the interviewee were permanently employed by the company and of those, 89.4% (211/236) were working within the field department. Employees within the field department work in the plantations, mainly harvesting ripe palm fruits. Only 6.8% (57/843), of the participants were hired as casuals, of whom 87.7% (50/57) were women.

Table 1: Description of cross-sectional survey participants

Variable		N (%)	Male	Female
Sex	Male	423 (50.2)		
	Female	419 (49.8)		
	Total	842		
Age	0-4	169 (20.1)	89 (21.1)	79 (18.9)
	5-9	148(17.6)	76 (18.0)	72 (17.2)
	10-19	117 (13.9)	47 (11.1)	70 (16.7)
	20-39	344 (40.9)	167 (39.6)	177 (42.2)
	>40	64 (7.6)	43 (10.2)	21 (5.0)
	Total	842	422	419
Origin	Oro	480 (57.1)	218 (51.8)	261 (62.4)
	Morobe	160 (19.1)	97 (23.0)	63 (15.1)
	Oro/Morobe	46 (5.5)	24 (5.7)	22 (5.3)
	Other provinces	157(18.3)	82(19.5)	72(17.2)
	Total	840	421	418
Work	Permanent	238 (28.2)	205 (86.1)	33 (13.9)
	Casual	57 (6.8)	7 (12.3)	50 (87.7)
	Not working	548 (65.0)	211 (38.6)	336 (61.4)
	Total	843	423	419
Work department	Field	211 (89.4)	178 (87.7)	33 (100.0)
	Security	12 (5.1)	12 (5.9)	0 (0.0)
	Transport	7 (3.0)	7 (3.5)	0 (0.0)
	Mill	4 (1.7)	4 (2.0)	0 (0.0)
	Growers	1 (0.4)	1 (0.5)	0 (0.0)
	HR	1 (0.4)	1 (0.5)	0 (0.0)
	Finance	0 (0.0)	0 (0.0)	0 (0.0)
	Total	236	203	33
Work casual	Collect mama lusfrut	22 (38.6)	0	22 (44.0)
	Weeding	13 (22.8)	1 (14.3)	12 (24.0)
	Empty Fruit Bunch (EFB) distribution	8 (14.0)	1 (14.3)	7 (14.0)
	Fertilizer application	6 (10.5)	0 (0.0)	6 (12.0)
	Harvest helper	3 (5.3)	2 (28.6)	1 (2.0)
	Other	5 (8.8)	3 (42.9)	2 (4.0)
	Total	57	7	50

Population movement was low with only 7.9% (66/839) individuals reporting to have been travelling within the last four weeks. 70.9% of 839 participants slept under a mosquito net the previous night. However, many people reported, that the treatment status of their nets was poor, with many of the nets not having been retreated within one year of usage.

Parasitological results

A total of 723 blood slides were collected, of which 242 were positive for malaria (33.5%, 95% confidence interval [CI]: 30.1-37.0). This result documents the high level of endemicity in this area. Between the three estates, the malaria prevalence differed significantly ($\chi 2=$

24.85, 2 df, p<0.001), ranging from 20.3% (95% [CI] 0.14-0.26) in Ambogo to 43.1% (0.37-0.49) in Sumbiripa (Table 2). At village level, the malaria prevalence ranged from 17.7% (95% CI 0.09-0.27) in Epa to 44.6% (95% CI 0.36-0.53) in Sumbiripa village ($\chi 2$= 25.80, 5 df, p<0.001) (Table 2). There was no significant difference between prevalence among males and females ($\chi 2$=0.10, 1 df, p=0.319). Overall, prevalence was highest in the age group 5-9 years (40.3%, 95% CI 0.32-0.49) (Figure 3). The same pattern was seen with *P. falciparum* infections, with 27.9% (95% CI 0.20-0.36) of the 5 to 9 years old children infected (Figure 4). For *P. vivax*, no difference between the age-groups was found (Figure 4).

Table 2: Malaria prevalence and bed net usage, by estates and by villages (cross-sectional survey).

Estate	**Number examined**	**Prevalence all species n infected; % (CI 95%)**	**Prevalence *P. falciparum* n infected; % (CI 95%)**	**Prevalence *P. vivax* n infected; % (CI 95%)**	**Bed net usage (%)**
Ambogo	177	36; 20.3 (0.14-0.26)	28; 15.8 (0.10-0.21)	6; 3.4 (0.01-0.06)	84.3
Sangara	279	91; 32.6 (0.27-0.38)	56; 20.0 (0.15-0.25)	35; 12.5 (0.09-0.16)	69.7
Sumbiripa	267	115; 43.1 (0.37-0.49)	60; 22.5 (0.17-0.27)	50; 18.7 (0.14-0.23)	63.3
Village					
Epa	68	12; 17.7 (0.09-0.27)	9; 13.2 (0.05-0.21)	2; 2.9 (-0.01-0.07)	97.5
Irigi	109	24; 22.0 (0.14-0.30)	19; 17.4 (0.10-0.25)	4; 3.7 (0.00-0.07)	75.8
Moale	154	48; 31.2 (0.24-0.38)	28; 18.1 (0.12-0.24)	23; 14.8 (0.09-0.20)	65.6
Javuni	125	43; 34.4 (0.26-0.43)	28; 22.4 (0.15-0.30)	12; 9.6 (0.04-0.15)	75.0
Irihambo	137	57; 41.6 (0.33-0.50)	33; 24.1 (0.17-0.31)	21; 15.3 (0.09-0.21)	57.6
Sumbiripa	130	58; 44.6 (0.36-0.53)	27; 20.8 (0.14-0.28)	29; 22.3 (0.15-0.29)	68.9

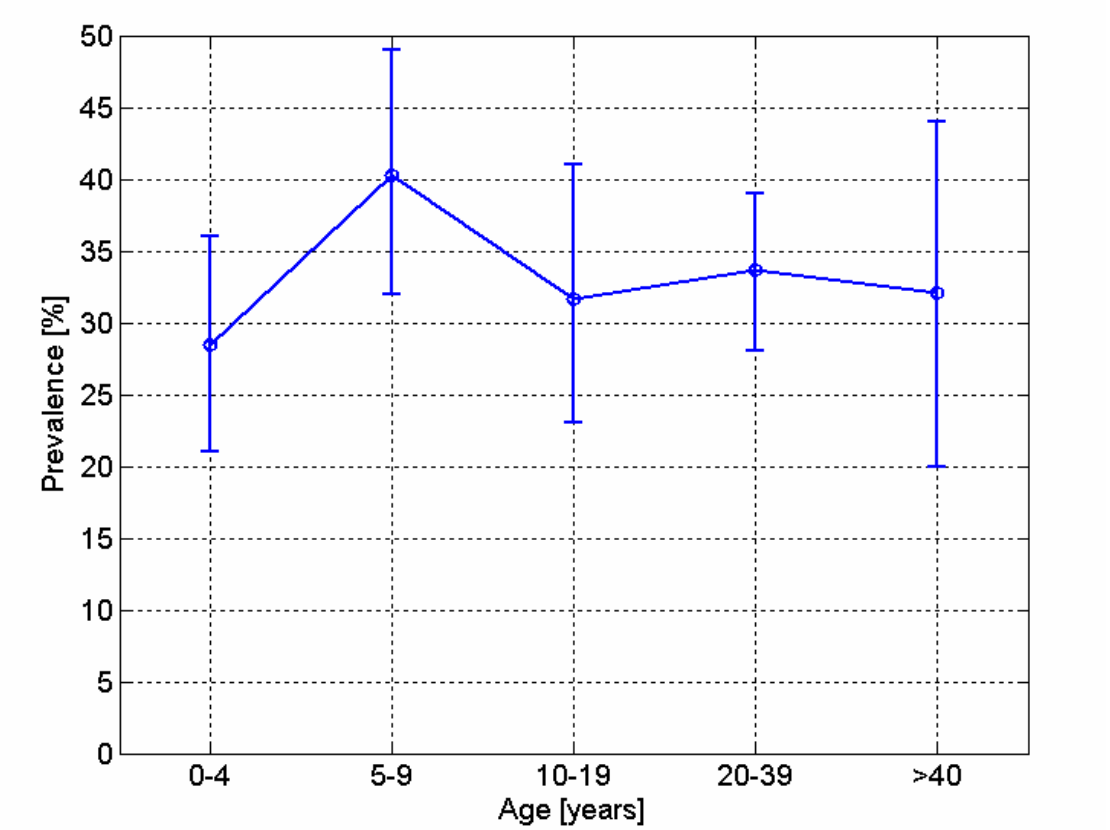

Figure 3: Malaria prevalence (all species) by age group, with 95% confidence intervals

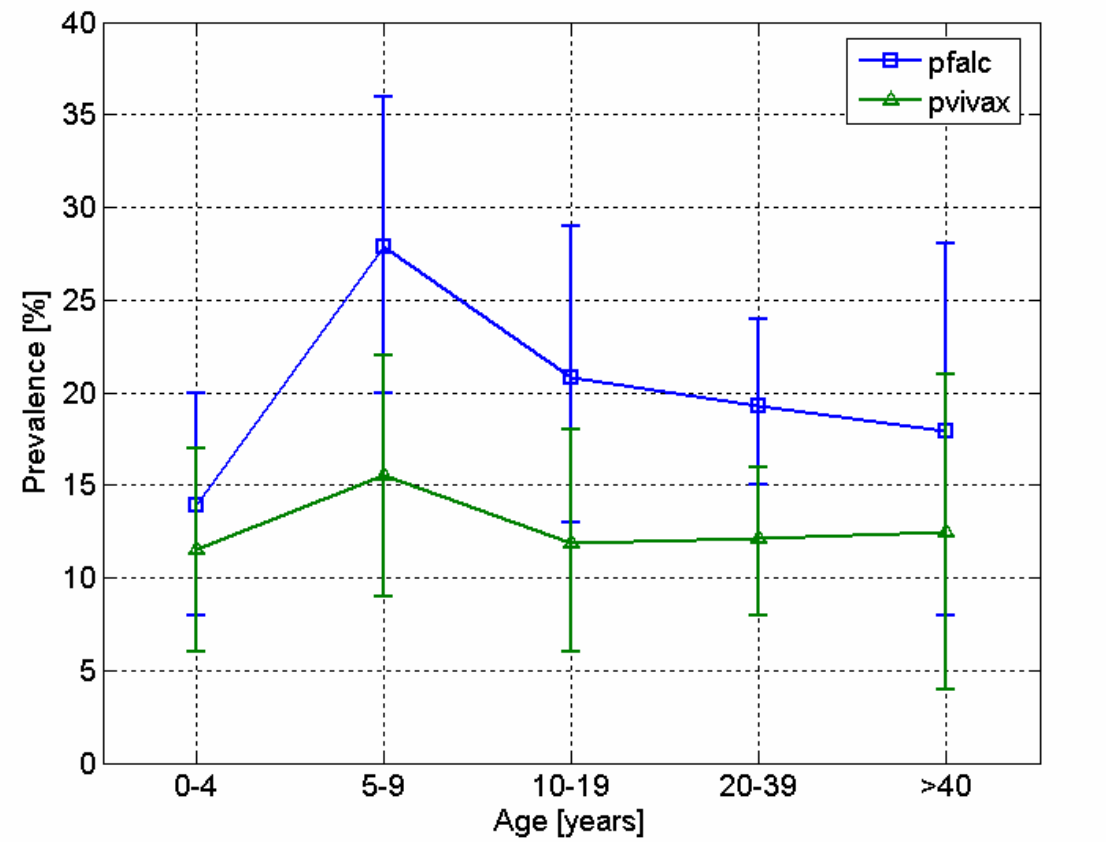

Figure 4: Malaria prevalence by species and age group, with 95% confidence intervals.

With 59.5% (144/242) of all positives, *P. falciparum* was the dominant species, followed by *P. vivax* (91/242, 37.6%) and *P. malariae* (16/242, 6.6%). No Plasmodium ovale was found. A high number of the positive slides (26/242, 10.7%) were mixed infections. The parasite densities of the infections were low, with 91.1% (194/213) of the slides having <500 parasites/µl. No difference in parasite densities between age groups was found ($\chi 2$= 20.18, 12 df, p=0.064). When investigating differences by species, 11.1% (14/144) of *P. falciparum* infections were >500 parasites/µl, compared to 4.4% (4/91) of the *P. vivax* infections (significant difference $\chi 2$= 10.48, 2 df, p=0.005).

Reported malaria episodes and spleen rates

25 participants presented with fever at the survey and a quarter (26.1%) of the 842 participants reported having had malaria during the past two weeks. No correlation between measured or reported fever and malaria parasitaemia was found: eight of the 20 (40.0%) people presenting with fever and having a blood slide taken, had a positive blood slide ($\chi 2$= 0.41, 1 df, p=0.521). Of the participants with reported fever during the last two weeks and having a malaria slide taken, only 37.6% (70/186) had a positive slide ($\chi 2$= 1.86, 1 df, p=0.173).

Almost everybody who reported having had malaria within the last two weeks visited a health centre (99.5%) and took anti-malarials (96.2%).

Overall, 59.7% of the participants had an enlarged spleen with an average size of 3.2 on the Hackett scale, but no association with a current malaria infection was seen, neither at individual ($\chi 2$= 0.78, 1 df, p=0.377) nor at village level. Looking at age groups, the average Hackett score was lowest for the youngest age group (0-4 years) with 2.4, and it rose with age to 3.8 for the oldest age group (>40 years). Despite the fact that this was reported differently by Genton *et al.* within the Wosera (Genton *et al.* 1995), a high rate of enlarged spleens in adults is a particular feature of malaria in PNG. Often, enlarged spleens in adults are linked to hyperreactive malarial splenomegaly (Muller *et al.* 2003). Though usually present within mid altitude zones (Muller *et al.* 2003), this syndrome might also occur within this study area.

Haemoglobin levels were low with a significantly higher level for men (11.0 g/dl, 95% CI 10.8-11.1) than for women (10.4 g/dl, 95% CI 10.3-10.5) ($\chi 2$= 24.73, 1 df, p<0.001). In children aged 5-9 years, the haemoglobin levels were significantly decreased with a malaria infection: by 1.0 g/dl (95% CI -1.43 – -0.47, p<0.001) for any species and 1.2 g/dl (95% CI -1.66 - -0.63, p<0.001) for *P. falciparum* infections. A significant association between *P. falciparum* and anaemia was found ($\chi 2$= 4.41, 1 df, p=0.036).

Behavioural aspects

There was no difference between employees and dependants in the risk of getting infected with malaria (Table 3). Neither were sleeping outside during the night (on the veranda or on a shelter) nor getting up before 7am associated with a higher risk of a malaria infection. On an individual level, sleeping under a bed net showed no protection against malaria (OR 1.2, 95% CI 0.8-1.7). However, there was a clear trend towards less malaria with higher bed net coverage between villages (Table 2). For every percentage increase in net coverage, the odds of malaria cases decreased by 3% (OR 0.97, 95% CI 0.96-0.99).

Table: 3 Risk factors for getting a malaria infection (cross-sectional survey).

Variable	**Total (n)**	**Infected (%)**	**Odds ratio**	**CI 95%**	**p-value (LRT)**
Village					
Moale	154	48 (31.2)	1		0.00
Epa	68	12 (17.7)	0.5	0.2-1.0	0.00
Irigi	109	24 (22.0)	0.6	0.3-1.1	
Javuni	125	43 (34.4)	1.1	0.7-1.9	
Irihambo	137	57 (41.6)	1.5	0.8-2.6	
Sumbiripa	130	58 (44.6)	1.2	1.1-3.2	
Sex			0.9	0.6-1.3	0.51
Age					
0-4	130	37 (28.5)	1		0.14
5-9	129	52 (40.3)	1.9	1.1-3.2	
10-19	101	32 (31.7)	1.1	0.6-2.0	
20-39	306	103 (33.7)	1.2	0.6-2.3	
>40	56	18 (32.1)	1.0	0.4-2.4	
Work permanent (vs not working (=dependants))	215	74 (34.4)	1.0	0.5-1.8	1.00
Work casual (vs not working (=dependants))	50	18 (36.0)	1.0	0.5-2.1	
Sleep under a bed net (yes vs no)	507	160 (31.6)	1.2	0.8-1.7	0.39
Sleep on veranda (yes vs no)	136	44 (32.4)	1.1	0.4-1.3	0.56
Sleep in a shelter (yes vs no)	63	25 (39.7)	0.7	0.4-1.3	0.25
Get up before 7am (vs getting up after 7am)	538	181 (33.6)	0.9	0.8-1.1	0.51

Incidence of routinely diagnosed malaria infections

Malaria was the most common disease diagnosed by HOP health staff in 2005 and 2006. In 2005 all malaria patients seen at the aid posts were diagnosed by clinical symptoms only. Of all patients seen at the 13 HOP aid posts in 2005, 39% (12,083/30,864) were diagnosed with malaria. Of the 12,083 malaria patients, 26% (3159/12083) were employees. On average, 1007 patients were diagnosed with malaria every month at the 13 aid posts. Malaria cases were found to be evenly distributed throughout the year 2005 (Figure 5), hence showing no seasonality.

In 2006, fewer patients were diagnosed with malaria than in 2005, especially after the introduction of RDTs in April. The RDT positivity rate, calculated for a sample of 90 patients, was found to be 48%. Overall, 9,940 patients were sick with malaria, still representing 29% (9,940/34,216) of all diagnosis made at the aid posts. 20% (2,015/9,940) of the malaria patients were employees. The malaria burden was highest in the beginning of the year, and then, after the introduction of RDTs in April a sharp decline until August was seen. During the three months in 2006 before the introduction of RDTs, 1,220 malaria cases were clinically diagnosed every month, of which 17% (211/1,220) were employees. After the introduction of RDTs in April and until the end of 2006, the monthly average of overall malaria cases decreased to 698. 153 (22%) of these malaria cases were employees. Malaria cases showed a seasonal peak in October, which did not reach the level of 2005, before RDT introduction (Figure 5).

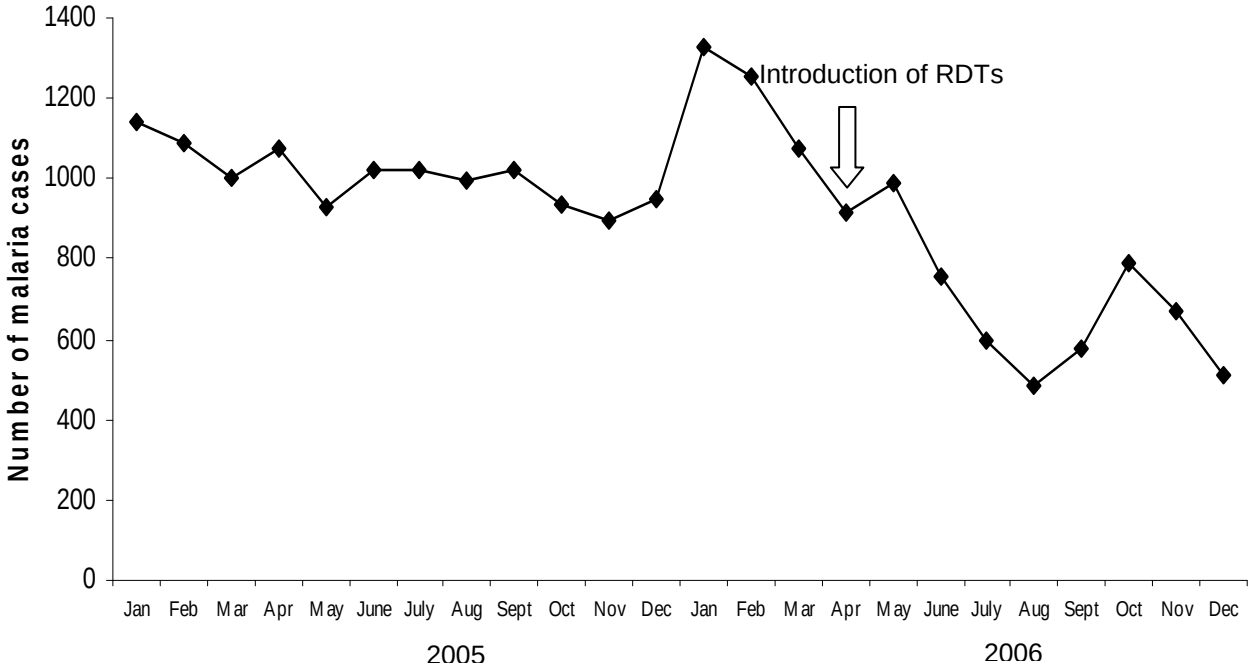

Figure 5: Monthly routine malaria incidence data from 2005 to 2006.

Days lost for the company

According to HOP administration, an employee sick with malaria was absent from work for an average of 1.8 days. Since 3,159 employees were sick with malaria in 2005 and 2,015 employees in 2006, this resulted in 5,686 lost days in 2005 and 3,627 lost days in 2006.

3.5 Discussion

To our knowledge, this work presents one of the few publications on the impact of malaria within an agro-industrial operation in endemic countries and the first one in PNG. Furthermore, it provides for the first time malariological information for the region around Popondetta, south-east PNG.

Clearly, malaria was found to be a major health problem within the CTP Plantation. The overall prevalence rate of 33.5% found among workers and their families is high. Furthermore, children and adults are both affected and this suggests there is little acquired immunity. This level of endemicity is similar to that found in other surveys done in the lowlands of PNG outside agro-industrial operations. Genton *et al.* (1995) and Cattani *et al.* (1986) found prevalence rates of 60% in the Wosera (East Sepik) and 35.0% to 42.7% surrounding Madang, respectively. The prevalence data of Genton *et al.* and Cattani *et al.* have been collected between 10 and 20 years ago and it is likely that the overall rate of transmission has decreased since. Furthermore, an oil palm plantation represents an artificial environment with its own ecosystem, which is likely to have a different transmission rate than a natural habitat. Comparisons with other oil palm plantations within PNG on the same altitudes would have been interesting, but such data are not available. Given the particular malaria epidemiology of PNG, which is by far the highest in the region, comparison with other Asian and Pacific countries is meaningless.

HOP offers housing to their employees within the geographical boundaries of the plantation. This special setting, with only employees and their dependants being allowed to live within the plantation, resulted in 40% of the participants being aged between 20 and 39. This is clearly not representative for the general population of the country and hence caution needs to be exerted when generalizing our results to the rest of the population.

Ripe palm fruits are collected by company trucks, creating deep wheel tracks on the ground. With enough rainfall such tracks present perfect breeding sites for Anopheles punctulatus (Cooper *et al.* 2002), which were found by human landing catches within the study area (Cooper R.D. personal communication). Though we conducted only one cross-sectional survey, the age peak for malaria infections (5-9 years old) was found to be characteristic for a highly endemic area (Genton *et al.* 1995; Muller *et al.* 2003). Such an intense malaria transmission can pose a high risk for all residents, especially for expatriates and other non-immune workers (for example coming from the cooler highlands). In one of the few documented examples, a large joint venture in Mozambique lost 13 expatriate employees due to malaria within two years (Sachs & Malaney 2002). Undoubtedly, malaria endemicity weighs down the attractiveness of industrial or agro-industrial sites.

Almost everybody who was feeling sick with presumptive malaria within the last two weeks had visited a health centre and received anti-malarials. This finding was not surprising, as employees and their dependants have free and ready health services provided by the company. This good access to treatment explains that we have not come across of any reported deaths from malaria.

In 2005, with the exception of one clinic, all malaria cases were diagnosed purely on a symptomatic basis. Unfortunately, symptoms of malaria are non-specific and many other diseases can present with the same clinical picture, including harmful diseases such as dengue, HIV or hepatitis B. But also many harmless viral infections present similar to a malaria infection. Thus, a purely symptomatic diagnosis of malaria can lead to a vast over-diagnosis of malaria cases (Font *et al.* 2001; Greenwood *et al.* 2005; Wang *et al.* 2006). This was illustrated by the fact that after the introduction of RDTs in April 2006 the number of reported malaria cases decreased steadily. But even with systematic testing with RDTs, still about 10,000 patients were diagnosed with malaria in 2006. This high level of morbidity represents a considerable cost of treatment (estimated cost per treatment: USD 0.18) and a substantial loss of productivity. These episodes resulted in 5686 lost days for the company in 2005, amounting to lost wages of over USD 60,000 (King G., personal communication). In 2006, the company lost 3627 working days due to malaria. The costs associated with malaria illness will increase significantly as PNG moves towards introducing the significantly more expensive Coartem® (artemether-lumefantrine) as the national first-line treatment.

More than half of the infections were caused by *P. falciparum* (57.4%), followed by *P. vivax* with 36.2%. Plasmodium falciparum infections are well known for their impact on morbidity and mortality, but also *P. vivax* infections are considered to be responsible for a substantial health burden (Greenwood *et al.* 2005; Picot 2006; Genton *et al.* 2008; Karyana *et al.* 2008). For an oil palm plantation, where approximately 60-75% of the employees do a physically strenuous work, a healthy workforce is crucial for a high productivity. The negative impact of malaria is also demonstrated by the low average haemoglobin level, which is a further drain on the energy of workers.

Virtually all inhabitants within the study area live within so-called “labour houses” (Figure 2), wooden two-bedroom houses, in which room temperatures can get very high, including at night. As a result, people sometimes sleep outside on their veranda or in a self-built shelter, where they are fully exposed to biting mosquitoes. Surprisingly, sleeping on a veranda or sleeping on a shelter was not found to be a risk-factor for a malaria infection. The most likely

explanation for this finding is that the mosquitoes prevalent within the plantation, Anopheles punctulatus (RD Cooper, personal communication) are exo- but also endophagic (Charlwood *et al.* 1986) and can easily enter and bite occupants inside the non mosquito-proofed houses.

The two main tools to prevent malaria infections are insecticide-treated mosquito nets (ITNs) and indoor residual spraying (IRS). Within HOP, nets are sold for half their commercial price, which explains the high usage rate of 70%. On an individual level no significant protection against malaria could be detected but this is expected in a situation with a homogeneously high level of net use. The village comparison showed a trend of decreasing number of malaria infections with increasing bed net level, confirming that the nets actually do have an impact. Unfortunately, the insecticide re- treatment level of the nets was poor since most nets were not retreated after 2004. Therefore, the introduction of LLINS, which are designed to maintain their efficacy against mosquitoes for at least three years without any re-treatment necessary, would be highly recommended (WHO Global Malaria Programme 2008). The high level of net protection also suggests that endemicity would be even higher if there was no protection.

Indoor residual spraying requires 1) an adequate knowledge of vector behaviour and occurrence, and (2) a highly structured programme including well-trained personnel, properly used insecticides, good logistics and scheduling and a high level of sustained financing (Lengeler & Sharp 2003). These terms would clearly be doable by a commercial oil palm plantation and thus, IRS would also be a feasible option for malaria control in the HOP setting. With housing provided by the plantation, upgrading the housing conditions including house screening would be a more long-term solution of malaria control. This approach has already shown great success within military barracks in Pakistan and India, reducing the malaria incidence up to 72% (Lindsay *et al.* 2002).

3.6 Conclusions

Malaria was found to be a major health burden to the Higaturu Oil Palms, posing a high risk for all inhabitants, indigenous workers as well as non-immune workers. Detrimental effect included direct cost of treatment, days away from work and reduced physical ability. These losses are substantial enough to warrant the implementation of energetic and long-lasting malaria control measures.

Competing interests

GK was the General Manager of the Higaturu Oil Palms plantations (HOP) from March 2005 to October 2007. DL is employed as Health Extension officer at HOP. BP, IM, TS and CL declare that they have no competing interests.

Authors' contributions

BP participated in the design of the study, conducted the field work, analysed and interpreted the data and drafted the manuscript. IM and CL were involved in designing and implementing the study and writing of the manuscript. TS assisted in the data analysis and writing of the manuscript. DL and GK were facilitating the overall coordination of the fieldwork and helped with the acquisition of the data.
All authors read and approved the final manuscript.

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4 Malaria vectors of Papua New Guinea

R. D. Cooper[1], D. G. E. Waterson[2], S. P. Frances[1], N. W. Beebe[3,4], B. Pluess[5] and A. W. Sweeney[3]

[1] Australian Army Malaria Institute, Gallipoli Barracks, Enoggera, Queensland, 4052, Australia.
[2] Pesticide Management, NSW Department of Primary Industries, PO Box 1, Sydney Markets, New South Wales, 2129, Australia.
[3] School of Biological Sciences, University of Queensland, Goddard Building, St Lucia, QLD 4072, Australia.
[4]CSIRO Entomology, Long Pocket Laboratories, Indooroopilly, QLD 4068, Australia.
[5] Public Health & Epidemiology, Swiss Tropical Institute, Socinstr. 57, CH 4002 Basel, Switzerland.

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4.1 Abstract

Understanding malaria transmission in Papua New Guinea (PNG) requires exact knowledge on what *Anopheles* species are transmitting malaria and is complicated by the cryptic species status of many of these mosquitoes. To identify the malaria vectors in PNG we studied *Anopheles* specimens from 232 collection localities around human habitation throughout PNG (using CO_2 baited light traps and human bait collections). A total of 22,970 mosquitoes were individually assessed using a *Plasmodium* sporozoite enzyme-linked immunosorbent assays to identify *Plasmodium falciparum*, *Plasmodium vivax* and *Plasmodium malariae* sporozoite proteins. All mosquitoes were identified to species by morphology and/or PCR. Based on distribution, abundance and their ability to develop sporozoites, we identified 5 species as major vectors of malaria in PNG these included: *Anopheles farauti* s.s, *Anopheles hinesorum* (incriminated here for the first time), *Anopheles farauti* 4, *Anopheles koliensis* and *Anopheles punctulatus*. *Anopheles longirostris* and *Anopheles bancroftii* were also incriminated in this study. Surprisingly, *An*. *longirostris* showed a high incidence of infections in some areas. A newly identified taxon within the Punctulatus Group, tentatively called *An*. *farauti* 8, was also found positive for sporozoite protein. These latter three species, along with *Anopheles karwari* and *Anopheles subpictus*, incriminated in other studies, appear to be only minor vectors, while *Anopheles farauti* 6 appears to be the major vector in the highland river valleys (>1500m). The 9 remaining *Anopheles* species found in PNG have been little studied and their bionomics are unknown; most appear to be uncommon with limited distribution and their possible role in malaria transmission has yet to be determined.

4.2 Introduction

Identification of malaria vectors and their role in transmission is a prerequisite to establishing why areas are malarious and how transmission can be interrupted (Pampana, 1969). The recent advent of molecular biology and polymerase chain reaction (PCR) has enabled species identification issues associated with isomorphic species complexes to be resolved and now entomological parameters can be reliably ascribed to accurately identified specimens.

In Papua New Guinea (PNG) the members of the Punctulatus Group, which originally included *Anopheles farauti* s.s., *Anopheles koliensis* and *Anopheles punctulatus*, have until recently been considered the major vectors of malaria and were regularly found infected with sporozoites (Peters and Standfast, 1960; Burkot *et al.*, 1987). With the application of allozyme electrophoresis, DNA hybridisation and PCR identification techniques this group, which includes the Farauti Complex, is now known to consist of 11 species in PNG (Foley *et al.*, 1993; Cooper *et al.*, 2002; Bower *et al.*, 2008), of these *An. punctulatus*, *An. koliensis*, *An. farauti* s.s., *An. farauti* 4 have been incriminated as vectors (Cooper and Frances 2002; Benet *et al.*, 2004) based on spatially limited studies from one or two localities. Outside of the Punctulatus Group four species have been incriminated: *Anopheles karwari*, *Anopheles subpictus*, *Anopheles bancroftii* and *Anopheles longirostris* (Afifi *et al.*, 1980; Hii *et al.*, 2000). Thus of the 20 species currently recognised in PNG, eight have been found to harbour sporozoites, the other 12 have been little studied or received scant attention either because they are uncommon species with limited distributions or they appear to have little or no association with humans.

Much of the recent work on the identification of malaria vectors in PNG has been restricted to the Maprik and Madang regions on the north coast of PNG (Afifi *et al.*, 1980; Burkot *et al.*, 1987; Hii *et al.*, 2000; Benet *et al.*, 2004). During the 1990's a series of surveys were conducted to identify the anophelines and to map their distribution throughout PNG (Cooper and Frances, 2002; Cooper *et al.*, 2002; Cooper *et al.*, 2006). These surveys resulted in collections being made from 793 localities across PNG and constitute the most extensive and comprehensive distribution study on anophelines in that country. Using species-specific genomic DNA probes (Cooper *et al.*, 1991; Beebe *et al.*, 1994, 1996) and a complementary PCR diagnostic technique (Beebe and Saul, 1995) these surveys have now established the range of the anopheline fauna throughout PNG. Specimens collected in these surveys were processed by enzyme-linked immunosorbent assay to identify the vector species across their entire range, the results of this study are reported here.

4.3 Material and methods

Surveys were made throughout the main island of PNG, the climate and topography of which is described in Cooper *et al.* (2002). From the original 793 collection sites (Cooper *et al.*, 2002, 2006) only those where adult collections were made within 500 m of human habitation were used in this study. This represented 232 trap nights using CO_2 baited light traps (Rohe and Fall 1979) and by 26 human landing collections (in most cases from 1900 to 2200 h) as described in Cooper *et al.* (2002, 2006). In the field, specimens were initially identified by morphology and then preserved either in liquid nitrogen or desiccated on silica gel. In the laboratory the head and thorax were removed from all specimens belonging to the Punctulatus Group and this material was assayed for sporozoite protein, the abdomens were used for species identification. This was done using polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) analysis of the internal transcribed spacer region 2 (ITS2) of the ribosomal DNA using the methods and primers of Beebe and Saul (1995), while *Msp*1 digestion of the ITS1 resolved *An. farauti* 8 from *An. farauti* s.s..

The enzyme-linked immunosorbent assays (ELISA) were performed using monoclonal antibodies to detect circumsporozoite protein of *Plasmodium falciparum*, *Plasmodium vivax* 210 (and 247 variant) and *Plasmodium malariae* and the methods of Wirtz *et al.* (1987). Sporozoite positive specimens were scored as those with absorption values greater than twice the mean (n=5) negative control value (Beier *et al.*, 1988). For species other than the Punctulatus Group the whole specimen was used in the ELISA.

4.4 Results

Collections were made from 258 locations throughout PNG (Fig 1), from these 22,970 specimens were collected and processed by ELISA for sporozoites. Eleven species were identified from this material including 9,962 *An*. *farauti* s.s., 1,189 *An*. *hinesorum* (formerly *An*. *farauti* 2), 3 *An*. *torresiensis* (formerly *An*. *farauti* 3), 1,535 *An*. *farauti* 4, 245 *An*. *punctulatus* and 8,600 *An*. *koliensis*, all specimens were individually identified by PCR. Additionally, 793 *An*. *longirostris*, 476 *An*. *bancroftii*, 116 *An*. *subpictus* and 13 *An*. *karwari* were identified by morphology. All species were found positive for sporozoites except for *An*. *torresiensis*, *An*. *subpictus* and *An*. *karwari* (Table 1).

Anopheles punctulatus is under reported in this study, this species is not readily attracted to CO_2 baited traps which was the main collection method used to sample anopheline populations.

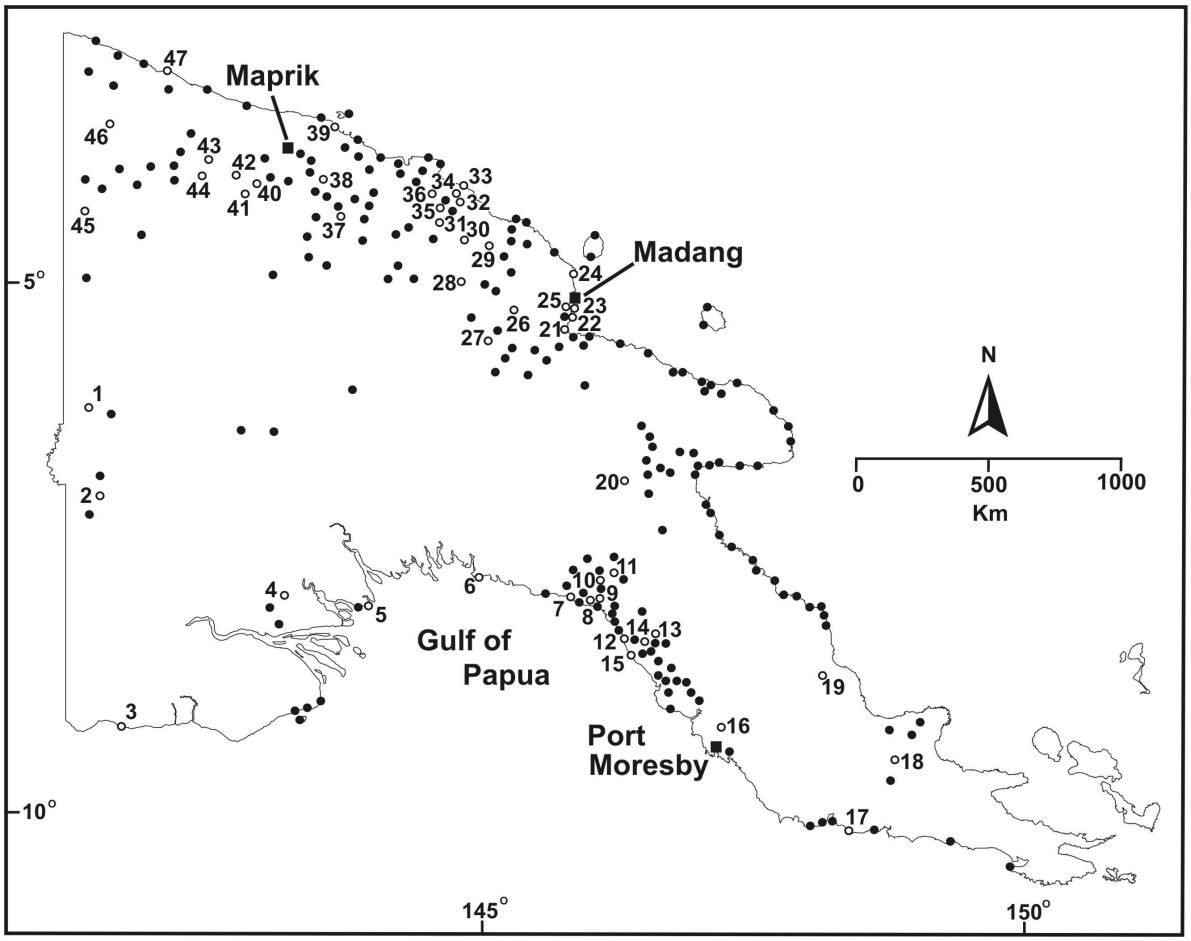

Fig. 1 Map of Papua New Guinea indicating sites where anophelines were collected (closed circles) and sites where anophelines positive for sporozoite protein were found (open circles). The numbers associated with open circles correspond to those in Table 1.
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An additional species within the Farauti Complex has recently been recognised and tentatively named *An*. *farauti* 8 (Bower *et al.*, 2008). This species displays the same ITS2 PCR-RFLP as *An*. *farauti* s.s., but occurs inland while *An*. *farauti* s.s. is restricted to the coast. The two species can be separated using the ITS1 region and the digest enzyme Msp1. Two specimens of *An*. *farauti* s.l. positive for *P*. *falciparum* sporozoites were collected >50 km inland and upon re-examination were found to be *An*. *farauti* 8.

From a number of collection sites a high incidence of infections were reported in *An*. *longirostris*; this species is considered a minor vector at best, to confirm that the results were correct and not due to contamination all samples were rerun with the same results being recorded.

Table 1: Number of sporozoites found per species

Species	No.	Pf	Pv	Pm	Mixed Pf/Pv	Total	%	No. Sites Sampled	No. Sites pos	Site no. from Fig. 1
An. farauti s.s.	9692	9	23	7	0	39	0.40	110	11	3,5,6,8,9, 12,15,17, 33,39,47
An. farauti 2	1,189	6	3	1	0	10	0.84	61	8	1,2,4,10, 14,20,25,26
An. farauti 3	3	0	0	0	0	0	0	1	0	
An. farauti 4	1,535	12	3	0	0	15	0.98	46	4	27,40,41,42
An. farauti 8	308	2	0	0	0	2	0.65	6	2	11, 13
An punctulatus	245	0	2	0	1	3	1.22	10	2	19, 46
An. koliensis	8,600	13	22	2	3	40	0.46	123	16	7,10,11,16,18,21,28,29, 30,31,32,35,36,37,38,45
An. longirostris	793	69	1	0	0	70	8.83	21	8	1,21,22,23,24,25,44,43
An. bancroftii	476	1	0	0	0	1	0.21	8	1	34
An. subpictus	116	0	0	0	0	0	0	9	0	
An. karwari	13	0	0	0	0	0	0	2	0	
Totals	22,966	112	54	10	4	180	0.78	391	52	

4.5 Discussion

Historically *An*. *farauti* s.l., *An*. *koliensis* and *An*. *punctulatus* (the original members of the Punctulatus Group) have been considered the major vectors of malaria in PNG due to their wide distribution, ability to occur in high densities, association with humans and their ability to develop human malaria parasites. Accordingly these species have received the most attention when implementing and evaluating control strategies.

However issues with the reliability of the morphological characters used to separate these three species and the recognition of additional isomorphic species within this group questions the vectorial status of these species. To overcome these problems in this study all specimens belonging to the Punctulatus Group were individually identified either by DNA hybridisation or by PCR. As a consequence *An*. *punctulatus* and *An*. *koliensis* were confirmed as major vectors of malaria in PNG due to their ability to develop the parasite, their wide spread distribution and their abundance.

Within the Farauti Complex seven species are now recognised as occurring in PNG (Cooper *et al.*, 2002; Bower *et al.*, 2008). Within this complex three species have now emerged which are of considerable importance with regards to malaria transmission - *Anopheles farauti* s.s., *An*. *hinesorum* and *An*. *farauti* 4. *Anopheles farauti* s.s was found here to be the major coastal vector throughout PNG supporting earlier findings of Cooper and Frances (2002) from Buka and Bougainville Islands and Benet *et al.* (2004) from Madang. *Anopheles hinesorum* is widely distributed throughout PNG (Cooper *et al.*, 2002) and here for the first time has been incriminated as a vector in a number of locations throughout its range. *Anopheles farauti* 4 is common throughout the northern part of the country, it has recently been incriminated as a vector in the Madang region (Benet *et al.*, 2004), and here we confirm the vectorial status of this species in other parts of its range (Fig. 1 sites 27, 40, 41 and 42). *Anopheles farauti* 4 presents a particular problem where its distribution overlaps with *An*. *hinesorum* and *An*. *koliensis* as it shares morphological markers with both these species, markers that were previously considered diagnostically important (Cooper *et al.*, 2002).

A fourth species, *An*. *farauti* 8, has recently been recognised within the Farauti Complex (Bower *et al.*, 2008), in the present study it was also found to be a vector of malaria parasites. This species appears to have a limited distribution occurring inland on the eastern edge of the Gulf of Papua (Fig. 1 sites 11 and 13) however within this range it was common and it may play an important role in malaria transmission locally within this region.

Four species outside of the Punctulatus Group have also been incriminated as malaria vectors in PNG: *An*. *subpictus*, *An*. *karwari*, *An*. *bancroftii* and *An*. *longirostris*.

Anopheles longirostris has been little studied; a preference for particular breeding sites appears to limit its association with humans resulting in mixed reports of its affinity for human blood. Metselaar (1957), van den Assem and van Dijk (1958), Peters and Christian (1960) and Cooper *et al.* (2006) reported it biting humans while Lee and Woodhill (1944) and Charlwood *et al.* (1985) indicated that it was primarily zoophilic. Mackerras and Roberts (1947) found that *An*. *longirostris* were reluctant to feed on malaria infected patients and of those that fed none developed *P. vivax* though one developed *P. falciparum*. Burkot *et al.* (1988), working in the Madang area, processed 3,000 *An*. *longirostris* for sporozoite protein and found none positive, though sporozoite protein was regularly found in specimens of *An*. *farauti* s.l., *An*. *koliensis* and *An*. *punctulatus* collected at the same time and from the same area. *Anopheles longirostris* was reported as sporozoite positive by van den Assem and van Dijk (1958) with no further reports until the early 1990s when Hii *et al.* (2000) found five positive specimens in a sample of 2,265 collected over a three year period in the Maprik area. In the present study infected *An*. *longirostris* were collected from 8 of its 20 collection sites; five closely related sites in the Madang area (sites 21 – 25) had very high numbers of infected specimens- site 21: 5 of 82, site 22: 8 of 111, site 23: 8 of 164, site 24: 37 of 197 and site 25: 4 of 146, the majority of infections were *P*. *falciparum* (61) with one *P*. *vivax*. At three sites *An*. *longirostris* was the dominant species collected while at the other two sites (21 and 25) appreciable numbers of other species were collected and infections were also found in 5 of 312 *An*. *koliensis* at site 21 and 3 of 94 *An*. *hinesorum* at site 25. Collections at sites 22 and 23 were made by human landing catches while those from sites 21, 24 and 25 were by CO_2 baited light traps; all collections were made at the end of the wet season in May 1995. Malaria transmission is intense in the Madang area with parasites rates of 37.5 – 42.5% for all ages and 53.6 - 56.7% in the 1-9 yr age group, *P*. *falciparum* makes up 70-76% of infections and a high proportion of cases are asymptomatic (Cattani *et al.*, 1986). Such transmission parameters may support the high incidence of sporozoite positive mosquitoes, but why such high infection levels should occur in a rather obscure species such as *An*. *longirostris* and not in one of the main vector species which also can occur in the area is unclear; it is possibly that climatic factors at the time of these surveys created favourable breeding conditions for *An*. *longirostris* at the expense of other anopheline species. Somboon *et al.* (1993) reported a factor in the blood of some bovine (25%, 4/16 tested) and pigs (8.3%, 1/12 tested) that cross reacted with the *P*. *falciparum* monoclonal antibody resulting in false positives. Cattle are rare in the Madang area though pigs were quite common around the villages where the *An*. *longirostris* collections were made. However as none of the collections

were made directly off pigs but were made either by human landing catches or by CO_2 baited light traps which attract unfed, host seeking mosquitoes, it is unlikely that the phenomenon reported by Somboon *et al.* (1993) could be responsible for the high incidence of infections recorded here. The findings here indicate that under certain climatic conditions *An*. *longirostris* may be an important local vector.

Anopheles bancroftii has a wide distribution throughout PNG but does not appear to be abundant anywhere within its range, it appears to be an indifferent feeder on humans and has rarely been collected biting humans in large numbers. It has been incriminated as a vector of malaria parasites by de Rook in 1929 (Lee *et al.*, 1987), Metselaar (1957) in Papua Province on Indonesian side of New Guinea and in PNG by Hii *et al.* (2000), and in this study.

Anopheles subpictus is an Southeast Asian immigrant, it has been collected from a number of locations in PNG (Cooper *et al.*, 2006) but is only common along the coast from Port Moresby west around the Gulf of Papua, and from within this region it has been incriminated as a vector on two occasions (Bang *et al.*, 1947; Afifi *et al.*, 1980). From collections made in the Madang area 754 *Anopheles subpictus* were dissected for sporozoites but all were negative although in the same collections sporozoite positive specimens of *An*. *farauti* s.l., *An*. *koliensis* and *An*. *punctulatus* were found (Afifi *et al.*, 1980). *Anopheles subpictus* is a species complex consisting of four species (Suguna *et al.*, 1994) one of which is a recognised vector of malaria in parts of coastal Southeast Asia. The specific identity of the PNG material in relation to other members of the complex is unknown. In this study only a small number were collected for analysis and none were found sporozoite positive.
Anopheles karwari, like *An*. *subpictus*, is a Southeast Asian introduction, and its present distribution is restricted to the north of PNG. Metselaar (1955) first incriminated it as a vector in Papua Province, Indonesia where he observed a sporozoite rate of 3.1%. In PNG it was first recorded as a vector of malaria by Afifi *et al.* (1980) but at the time was misidentified as *An*. *subpictus*. Hii *et al.* (2000) found it common in the Maprik area and recorded 14 sporozoite positive specimens in a collection of 13,134 anophelines made by human landing and light trap catches over a three year period. Very few specimens were collected in this present survey and none were positive for sporozoites.

Anopheles subpictus, *An*. *karwari*, *An*. *bancroftii* and *An*. *longirostris* because of their loose association with humans, their limited and patchy distribution and their normally low

abundance can only be considered as minor vectors of malaria. They may at times have some local importance under the right climatic and ecological conditions.

Of the remaining ten *Anopheles* species found in PNG very little is known about their bionomics and their potential to transmit malaria. Species such as *An*. *clowi*, *An*. *papuensis* and *An*. *farauti* 5 appear to be quite rare with limited distributions and are unlikely to be involved in malaria transmission. *Anopheles* sp near *punctulatus* is an uncommon species that so far has only been found in a few remote areas of PNG; *An*. *annulipes* s.l., both the coastal and highlands populations, are zoophilic species; *An*. *torresiensis*, *An*. *meraukensis*, *An*. *novaguinensis* and *An*. *hilli* are all restricted to the southern plains of Western Province, a vast, sparsely populated region where few malaria vector studies have been performed. None of these species have been studied in any detail but owing to the fact that they appear to be uncommon with limited distribution their role, if any, in malaria transmission would be at a local level.

Anopheles farauti 6, the large farauti of Lee (1946), is found exclusively in the highland river valleys of New Guinea (>1500m) (Cooper *et al.*, 2002). It was studied by Peters and Christian (1960) who noted that it was the most common *Anopheles* species biting humans in the Waghi Valley where they recorded a sporozoite rate of up to 2.2 % in this species. *Anopheles farauti* 6, confirmed by PCR-RFLP, was the most common anopheline biting humans in the Baliem Valley, a similar highland valley in Papua Province, Indonesia (Cooper *et al.*, 2002). Malaria is unstable in these highland river valleys with low levels of transmission and periodic epidemics (Bangs *et al.*, 1999; Mueller *et al.*, 2005). *Anopheles farauti* 6 is the only member of the farauti complex commonly found at these altitudes and is most likely an important malaria vector within its restricted range.

This study if anything highlights the lack of knowledge with regards to the bionomics of the anophelines of PNG and their role in malaria transmission. Table 2 presents an overview of the currently recognised species and their vectorial status which with some species is based on very limited information. As indicated a number of species - *An*. *torresiensis*, *An*. *meraukensis*, *An*. *novaguinensis*, *An.* sp. nr*. punctulatus* and *An*. *hilli* - have been too inadequately studied. Their limited distribution is partly responsible for this, however even in their restricted areas they may have the potential to occur in large numbers and may play an important, if local, role in malaria transmission. The findings in this study with regards to *An*. *longirostris* highlight the role that even a rather uncommon and obscure species can play in malaria transmission.

Table 2: Vectorial status of the currently known *Anopheles* species in Papua New Guinea.

Species	Vectorial Status	Distribution/ Abundance	Comments	Reference
An. koliensis	major	widespread / common	established vector three genotypes possible vectorial differences	Hii et al. 2000, Benet et al. 2004, this paper
An. punctulatus	major	widespread / common	established vector	Hii et al. 2000, Benet et al. 2004, this paper
An. farauti s.s.	major	widespread /common coastally	established vector	Cooper and Frances 2002, Benet et al. 2004, this paper
An. hinesorum	major	widespread / common	several genotypes, zoophilic on Buka and Bougainville Is. PNG	Cooper and Frances 2002, this paper
An. farauti 4	major	limited / common	common throughout northern PNG only	Benet et al. 2004, this paper
An. longirostris	minor	wide spread / uncommon	possible species complex	Hii et al. 2000, this paper
An. farauti 8	minor	limited / uncommon	recently discovered, species not studied	Bower et al. 2008, this paper
An. bancroftii	minor	widespread / uncommon	possible species complex, with varying host preferences	De Rook 1929, Hii et al. 2000, this paper
An. farauti 6	minor	limited / common in the highlands (>1500m)	the large farauti of Lee 1946, incriminated on circumstantial evidence	Peters and Christian 1960
An. subpictus s.l.	minor	limited / uncommon	species complex – PNG species unknown	Bang et al. 1947, Afifi et al. 1980
An. karwari	minor	limited / uncommon	previously misidentified as *An. subpictus*	Metselaar 1955, Afifi et al. 1980, Hii et al. 2000
An. meraukensis	none	limited / uncommon	species not studied – potential unknown experimentally infected in Australia	Mackerras and Roberts 1947
An. novaguinensis	none	limited / uncommon	species not studied – potential unknown	
An. torresiensis	none	limited / uncommon	species not studied – potential unknown	
An. sp. nr. punctulatus	none	limited / uncommon	species not studied – potential unknown	
An. hilli	none	limited / uncommon	vector in Cairns, Australia; in PNG species not studied – potential unknown	Roberts and O'Sullivan 1948
An. annulipes s.l.	none	limited / uncommon	two genotypes in PNG both zoophilic experimentally infected in Australia	Mackerras and Roberts 1947
An. farauti 5	none	limited / rare	species not studied	
An. clowi	none	limited / rare	species not studied	
An. papuensis	none	limited / rare	species not studied	

Further, the use of PCR is now the standard for verifying species identification and thus the species role in malaria transmission, but this technology is also raising new issues. *Anopheles farauti* 8, a recently discovered species based on molecular evidence, is here incriminated as a vector of malaria in PNG but as yet nothing is know of its behaviour or its distribution in PNG. The *An. koliensis* taxon is now known to contain three independently evolving rDNA genotypes suggesting the presence of three cryptic species (Benet *et al.*, 2004; NW Beebe unpublished data) which differ in biting behaviour, a character important in determining control strategies. *Anopheles longirostris* and *An*. *bancroftii* also appear to be complexes of independently evolving rDNA genotypes, indicating the presence of several cryptic species (Beebe *et al.*, 2001; NW Beebe unpublished data). *Anopheles hinesorum*, incriminated for the first time in this study, is a complex of several rDNA genotypes, the members of two of these genotypes appear not to feed on humans (Cooper and Frances 2002; NW Beebe unpublished data). Until we fully understand the bionomics of the extant and newly emerging species and genotypes we will never have a complete understanding how malaria is transmitted in PNG nor how it can be controlled or eliminated.

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5 Cochrane review: Indoor residual spraying for preventing malaria

Bianca Pluess[1], Frank C Tanser[2], Christian Lengeler[1], Brian L Sharp[3]

[1]Public Health and Epidemiology, Swiss Tropical Institute, Basel, Switzerland

[2]Africa Centre for Health and Population Studies, University of KwaZulu-Natal, Mtubatuba, South Africa

[3]Malaria Research Lead Programme, Medical Research Council, Durban, South Africa

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5.1 Abstract

Background

Primary malaria prevention on a large scale is essentially achieved through two main vector control interventions: indoor residual spraying (IRS) and insecticide-treated mosquito nets (ITNs). Up to now the effectiveness of IRS in reducing ill-health has never been properly quantified.

Objectives

To quantify the impact of IRS alone, and to compare the relative impacts of IRS and ITNs, on key malariological parameters.

Search methods

We searched the Cochrane Infectious Diseases Group Specialized Register (November 2008), CENTRAL (The Cochrane Library 2008, Issue 4), MEDLINE (1966 to November 2008), EMBASE (1974 to November 2008), LILACS (1982 to November 2008), mRCT (November 2008), reference lists, and conference abstracts. We also contacted researchers in the field, organizations, and manufacturers of insecticides (June 2007).

Selection criteria

Cluster randomised controlled trials (RCTs), controlled before-and-after studies (CBA) and interrupted time series (ITS) of IRS compared to no IRS or ITNs. Trials including studies examining the impact of IRS on special groups not representative of the general population, or using insecticides and dosages not recommended by the World Health Organisation (WHO) were excluded.

Data collection and analysis

Two authors independently reviewed trials for inclusion. One author extracted data, assessed methodological quality and analysed the data. Where possible, we adjusted the confidence intervals (CIs) for the clustering effect.

Results

Four RCTs, one CBA and one ITS met the inclusion criteria. Two of the four RCTs used adequate methods to generate allocation sequence and concealment. In unstable malaria settings, IRS significantly reduced the incidence rate of malaria infections with the protective efficacy ranging from 24% to 86%. No clear evidence was seen in reducing prevalence. Contradicting results were found assessing IRS versus ITNs in protecting against malaria incidence rates. Only one included trial measured the impact of IRS on incidence and prevalence in a stable malaria setting. For children under five, IRS was more effective than ITNs or no IRS in reducing incidence. Comparing IRS vs no IRS or vs ITNs, there was no difference in the prevalence rates (RR 1.01, 95% CI 0.84 to 1.22; 555 participants, 1 trial).

Authors' conclusions

The number of high-quality trials is too low to provide solid evidence. IRS seems effective in reducing malaria incidence in unstable malaria settings. For stable malaria settings no conclusion about impact can be derived. There is an urgent need for more high-quality comparative trials between ITNs and IRS, as well as to quantify their combined effects.

5.2 Background

There were an estimated 247 million malaria cases among 3.3 billion people at risk in 2006, causing nearly a million deaths, mostly of children under 5 years. 109 countries were endemic for malaria in 2008, 45 within the WHO African region (WHO 2008). Ninety per cent of all malaria cases occur in sub-Saharan Africa, in areas of stable endemic transmission, and around 20% of all deaths in children have been attributed directly to malaria (Snow 1999). The disease causes widespread premature death and suffering, imposes financial hardship on poor households, and holds back economic growth and improvements in living standards. The rapid spread of resistance first to chloroquine and now to sulfadoxine-pyrimethamine has greatly increased the cost and difficulty of malaria case management, particularly in Africa (RBM 2005). Estimates have suggested that malaria costs the African countries US$12 billion annually and may considerably retard economic development (Sachs 2002).

Primary prevention of malaria is essentially achieved through two main vector control interventions: indoor (house) residual insecticide spraying (IRS); and insecticide-treated (mosquito) nets (ITNs). The health effects of ITNs have been comprehensively summarized in two Cochrane Reviews, one for general populations (Lengeler 2004) and one for pregnant women (Gamble 2006).

IRS has a long and distinguished history in malaria control. Using mainly dichloro-diphenyl-trichlorethane (DDT), malaria was eliminated or greatly reduced as a public health problem in Asia, Russia, Europe, and Latin America (Schiff 2002; Lengeler 2003; Roberts 2004). IRS continues to be used in many parts of the world, with the services provided by the public health system or by a commercial company (usually for the benefit of its employees). There is no IRS programme known to us in which beneficiaries were expected to contribute financially.

A historical review of IRS in Southern Africa investigated the malaria situation before and after the introduction of IRS in South Africa, Swaziland, Namibia, Zimbabwe, and Mozambique, where it continues to protect 13 million people (Mabaso 2004). Immediately after the implementation of control operations, spectacular reductions in malaria parameters and vector densities were recorded, and in certain instances the intervention led to local elimination. Another historical paper reviewed the health impacts of 36 successful IRS programmes in 19 countries throughout sub-Saharan Africa (Kouznetsov 1977). The analyses compared parasite rates and other malariological outcomes before and after the operation in each of the five major eco-epidemiological zones and demonstrated substantial epidemiological benefits. Unfortunately, most of these studies simply documented time trends of malaria parameters. This is also the case for the most recent programme impact assessments (Sharp 2002; Tseng 2008; Teklehaimanot 2009).

IRS operates both through repelling mosquitoes from entering houses and by killing female mosquitoes that are resting inside houses after having taken a blood meal. This implies that IRS is most effective against mosquito species that are resting indoors (so called endophilic mosquitoes). Spraying needs to be carried out between once and three times per year; the timing depending on the insecticide and the seasonality of transmission in a given setting. Reviewing the advantages and disadvantages of each insecticide is beyond the scope of this review and can be found in Najera (2001).

IRS has the advantage of being able to make use of a much wider range of insecticide products in comparison to ITNs, for which pyrethroids are the only class of insecticide currently used. The World Health Organization (WHO) recommends a number of insecticides for individual residual spraying: DDT wettable powder (WP); malathion WP; fenitrothion WP; pirimiphos-methyl WP and emulsifiable concentrate (EC); bendiocarb WP; propoxur WP; alpha-cypermethrin WP & SC; cyfluthrin WP; deltamethrin WP; etofenprox WP; and lambda-cyhalothrin WP (WHOPES 2007). This extended range of insecticides has important benefits for the management of insecticide resistance and hence the long-term sustainability of vector control (pyrethroid resistance has already been reported in many parts of tropical Africa and other parts of the world among populations of the major malaria vectors). The potentially adverse effects of insecticides used for IRS, especially DDT, is an important issue but one that is beyond the scope of this review.

Insecticide spraying is often done at very large scale, and thus randomised controlled trial designs may not always be feasible. However, controlled before-and-after studies are clearly feasible, as are interrupted time series. We plan to include these three study designs while excluding simple pre-test and post-test studies with no concurrent controls, as the many potential biases make interpretation a problem. In order to take into account the differences of design, the primary analyses will be stratified by study design. In all identified studies the allocation is expected to be by clusters rather than by individuals, since IRS is only effective if a large proportion of the population is protected.

Two reviews have outlined the cost and health effects of IRS (Curtis 2001; Lengeler 2003) including a comparison of IRS against ITNs, but neither was conducted systematically or assessed the methodological quality of the included studies. Yukich *et a*l. (2008) presented standardized cost and cost-effectiveness assessments for the major ITN distribution models as well as for two IRS programmes in Southern Africa. Here we aim to quantify the health benefits of IRS and to compare how IRS and ITNs in their ability to prevent ill-health from malaria.

5.3 Objectives

To quantify the impact of IRS alone, and to compare the relative impacts of IRS and ITNs, on key malariological parameters.

5.4 Methods

5.4.1 Criteria for considering studies for this review

Types of studies

1. **RCTs and quasi-RCTs**, randomised by cluster (cluster RCTs) and with three or more units per arm; because of the mode of action of IRS we did not expect to find trials with individual randomisation.
2. **Controlled before-and-after studies** with (1) two or more units per arm, (2) a contemporaneous control group, (3) monitoring of at least one transmission season before and after the intervention and (4) at least 60% coverage in the intervention arm.
3. **Interrupted time series**, with (1) a clearly defined point in time when the intervention occurred, (2) monitoring of at least two transmission seasons before and after the intervention and (3) at least 60% coverage in the intervention arm.

Types of participants

Children and adults living in rural and urban malarious areas.

Excluded: studies examining the impact of IRS on soldiers, refugees, industrial workers and other special groups not representative of the general population.

Types of interventions

Interventions

IRS carried out with insecticides recommended by the World Health Organization at the correct dosage (WHO 2006; WHOPES 2007). Selected insecticides should not have been used where site-specific insecticide resistance has been reported by the authors or in other available literature. To this effect, we searched for publications on insecticide resistance for each included trial site.

For the comparison with ITNs, we used the same inclusion criteria as in Lengeler 2004: mosquito nets treated with a synthetic pyrethroid insecticide at a minimum target dose of: 200 mg/m2 for permethrin and etofenprox; 30 mg/m2 for cyfluthrin; 20 mg/m2 for alphacypermethrin; and 10 mg/m2 for deltamethrin and lambdacyhalothrin.

Controls

- Should not have received another insecticide-based malaria intervention.

- Should not have received a malaria-co-intervention(s) that differed from the intervention arm.

- ITNs only for the comparison IRS versus ITNs. For this comparison we made a difference between situations in which ITNs were distributed to a population previously protected by IRS (which was obviously stopped for the time of the study) from situations in which the distribution of ITNs represents the first vector control intervention.

Types of outcome measures

Child (< 10 years) mortality from all causes as determined by a prospective demographic surveillance system. Severe disease: site-specific definitions, based on the WHO guidelines (WHO 2000). The definition includes demonstration of parasitaemia. Cerebral malaria is defined as coma or prostration and/or multiple seizures. The cut-off for severe, life-threatening anaemia is set at 5.1 g/L. Uncomplicated clinical malaria episodes: measured using site-specific definitions, including measured or reported fever, with or without parasitological confirmation - as long as the case definition was similar in all trial arms. Results from both passive and active case detection were considered. Parasite prevalence: obtained using site-specific method for estimating parasitaemia, usually thick and/or thin blood smears. In the case of repeated cross-sectional surveys, we used the mean of all the measures and adapted the denominator to be equal to the average sample size of the surveys (to avoid inflating artificially the denominator with repeated surveys of the same individuals). High density malaria prevalence: same as for parasite prevalence but with a site-specific parasitological cut-off. Anaemia: haemoglobin levels measured in g/dL. Standard anthropometric measures: weight-for-age, height-for-age, weight-for-height, skinfold thickness, and/or mid-upper arm circumference. Splenomegaly: measured using Hackett's scale from 1 to 5.

5.4.2 Search methods for identification of studies

We attempted to identify all relevant studies regardless of language or publication status (published, unpublished, in press, and in progress). For details see Table 1.

Databases

On 28 November 2008 we searched the following databases using the search terms and strategy described in Table 1: Cochrane Infectious Diseases Group Specialized Register; Cochrane Central Register of Controlled Trials (CENTRAL) as published in *The Cochrane*

Library; MEDLINE; EMBASE; and LILACS. We also searched the *meta*Register of Controlled Trials (*m*RCT) using 'insecticide$' and 'malaria' as search terms.

Table 1: Detailed search strategy

Search set	**CIDG SR^/LILACS^^**	**CENTRAL/MEDLINE^^**	**EMBASE^^**	**LILACS^^**
1	malaria	malaria	malaria	malaria
2	insecticide*	insecticide*	insecticide*	insecticide*
3	indoor residual spray*	indoor residual spray*	indoor residual spray*	indoor residual spray*
4	IRS	house spray*	IRS	IRS
5	house spray*	IRS	house ADJ spray$	house spray*
6	2 or 3 or 4 or 5	MOSQUITO CONTROL/INSTUMENTATION/METHODS	VECTOR CONTROL	2 or 3 or 4 or 5
7	1 and 6	INSECTICIDES/THERAPEUTIC USE	INSECTICIDE	1 and 6
8		PYRETHRINS/ADMINISTRATION AND DOSAGE	2-7/OR	
9		2-8/OR	1 and 8	
10		1 and 9		
^Cochrane Infectious Diseases Group Specialized Register ^^Upper case: MeSH or EMTREE heading; Lower case: free text term				

Agencies and manufacturers

We contacted the following agencies, which have funded malaria control studies, for unpublished and ongoing trials: World Bank; Rockefeller Foundation; UNICEF; World Health Organization; PAHO; and USAID. We also contacted the following manufacturers of insecticides: Bayer; BASF; Sumitomo; and Syngenta (June-July 2007). In June 2007 we also searched the US Armed Forces Pest Management Board web site for relevant trials, as well as all other sources that we identified in the process of the search.

Reference lists

We checked the reference lists of all studies identified by the above methods.

5.4.3 Data collection and analysis

Study selection

BP screened the results of the search strategy for potentially relevant studies and retrieved full articles. BP and FT independently assessed all identified studies for inclusion in the review, using an eligibility form based on the inclusion criteria. We scrutinized each report to avoid study duplication. We attempted to contact the study authors for clarification if it was unclear whether a study met the inclusion criteria or if there were issues with the study

design. CL was asked to resolve any differences in opinion. We explain below the reasons for excluding studies ("Characteristics of excluded studies").

5.4.4 Assessment of methodological quality

BP/FT and CL independently evaluated the methodological quality of each included study. We attempted to contact the study authors if key information was missing or unclear, and resolved any disagreements through discussion.

RCTs and quasi-RCTs

BP assessed the risk of bias of each included trial using the Cochrane Collaboration's risk of bias tool (Higgins 2008). We followed the guidance tool to make judgements on the risk of bias in six domains: sequence generation; allocation concealment; blinding (of participants, personnel, and outcome assessors); incomplete outcome data; selective outcome reporting; and other sources of bias. We categorized these judgements as 'yes' (low risk of bias), 'no' (high risk of bias), or 'unclear'.

Controlled before-and-after studies

We followed a strategy published elsewhere (Adinarayanan 2007); BP and FT independently assessed the quality of the included CBA study using a variety of criteria that we considered important and had specified *a priori*. These included: high intervention coverage in the community of interest (defined as at least 60% IRS coverage), presence of some type of comparison group with no intervention, reporting of outcomes for the entire community. We also attempted to identify concurrent control activities carried out at the same time or just before the IRS intervention by screening the primary study report and other relevant literature.

Interrupted time series

We used the criteria published elsewhere (EPOC 2002) to assess the study quality of the one included study. Criteria included protection against secular changes, sufficient data points to enable reliable statistical inference, protection against detection bias, and completeness of the data set. We also attempted to identify concurrent control activities carried out at the same time or just before the IRS intervention by screening the primary study report and other relevant literature.

5.4.5 Data extraction

BP independently extracted the data from each study into standardized data extraction forms. Again, we attempted to contact the corresponding author in any case of unclear or missing data.

RCTs and quasi-RCTs

We extracted data according to the intention-to-treat principle: if any individuals allocated to a treatment group was analysed as if the person had effectively received the intervention. If there was discrepancy between the number of units/participants randomised and the number of units/participants analysed we calculated the percentage losses to follow-up in each group and reported this information. In trials that compared ITNs with IRS, we assessed the differences in coverage between the different groups and presented this information in a table.

Cluster RCTs: Where results have been adjusted for clustering, we extracted the point estimate and the 95% confidence interval. If the results were not adjusted for clustering, we extracted outcome data as for individual RCTs and corrected the data in the analysis (see 5.4.6). We always recorded the number of clusters, the average size of clusters, and the unit of randomizations (eg household, village or other). The statistical methods used to analyse the trials are described below in section 5.4.6.

Controlled before-and-after studies

We extracted data using the same methods as for the RCTs, but we added information on the comparability of baseline characteristics and the time period of data collection.

Interrupted time series

We extracted data using the same methods as for the RCTs, but we added information on the comparability of baseline characteristics and additional information relating to the assessments made before and after the initiation of the intervention, using the approach recommended by EPOC 2002.

5.4.6 Data analysis

Individual and cluster RCTs

We had planed to meta-analyse the data from RCTs and quasi-RCTs using Review Manager 4.2 and to stratify the analyses according to whether the trial had an individual or cluster allocation. However, no stratification was necessary as all trials were cluster randomised trials. Results are presented with 95% confidence intervals. Only reports of RCTs that had adjusted for the cluster effect (or could be done post-hoc in this review) were included in the analysis. We presented a narrative or tabulated summary of data for the other study designs.

Cluster RCTs with two or three arms and at least three clusters per arm were used for the comparisons of IRS versus no intervention or IRS versus ITNs. The two other study designs (CBA and ITS) were used only for the comparison of IRS versus no intervention. We did not plan to summarize trials with these alternative designs in a meta-analysis.

Cluster RCTs: These trials require a more a complex analysis than that for individual RCTs (Hayes 2000). Observations on participants in the same cluster tend to be correlated and that intra-cluster variation must be accounted for during the analysis. If this correlation is ignored in the analysis the measure of effect remains a valid estimate but the associated variance of the estimate would be underestimated, leading to unduly narrow confidence intervals. In a meta-analysis trials analysed without allowing for the design effect would receive too much weight.

When the results have been adjusted for clustering, we combined the adjusted measures of effect in the analysis. For dichotomous outcomes expressed as risk the results can be adjusted for clustering by multiplying the standard errors of the estimates by the square root of the design effect, where the design effect is calculated as DEff=1+(m-1)*ICC. This requires information such as the average cluster size (m) and the intra-cluster correlation coefficient (ICC). Unfortunately, the ICC was not reported by trial authors. Therefore, we carried out a sensitivity analysis using a range of values for the ICC to determine if the conclusions of the analysis would change as the ICC increased (Table 2 and Table 3).

Table 2: Impact of ICC[a] on confidence interval: IRS vs control in Curtis (1998)

Prevalence of malaria infections (any species); children aged 1 to 6 years			
Study	**Risk ratio**	**ICC**	**95% Confidence interval**
Curtis 1998 (Tanzania)	0.92	0	0.83 to 1.01
		0.01	0.83 to 1.02

Footnotes
[a] ICC: Intra-cluster correlation coefficient

Table 3: Impact of ICC[a] on confidence interval: IRS vs control in Rowland (2000)

Prevalence of *P. falciparum*, children aged 5 to 15 years			
Study	**Risk ratio**	**ICC**	**95% Confidence interval**
Rowland 2000 (Pakistan)	0.09	0	0.09 to 0.09
		0.01	0.08 to 0.10
		0.2	0.06 to 0.14
		0.3	0.06 to 0.16
		0.4	0.05 to 0.17
Prevalence of *P. vivax*, children aged 5 to 15 years			
Study	**Risk ratio**	**ICC**	**95% Confidence interval**
Rowland 2000 (Pakistan)	0.32	0	0.30 to 0.33
		0.01	0.26 to 0.38
		0.2	0.13 to 0.76
		0.3	0.11 to 0.92
		0.4	0.09 to 1.09

For dichotomous outcomes expressed as rates, we applied the methods described in (Bennett 2002) using a rate ratio calculated from the mean incidence rates for each treatment group. In the case of the study by Misra 1999 the authors used a geometric mean of the incidence rates and we applied the method described by Bennett 2002 using a rate ratio based on the geometric mean incidence rates for the outcomes "*P. vivax* and *P. falciparum* combined".

Heterogeneity: With enough trials we would have assessed heterogeneity by (1) inspecting the forest plots to detect overlapping confidence intervals, (2) applying the chi-squared test with a P value of 0.10 indicating statistical significance, and (3) implementing the I^2 test with a value of 50% denoting moderate levels of heterogeneity. However, the number of trials was so low that combining of trials was not possible.

We stratified the presentation of the results into two groups on the basis of the entomological inoculation rate (EIR < 1 and = 1), as well as on the basis of the main types of vectors. Where possible the analyses were stratified by parasite species (*P. falciparum* and *P. vivax)*. Finally, consideration was given to the fact that in some areas the vector control activities have gone on for many years before the reported study, while in some other situations the investigated study introduced the vector control activities. While we have at present no way to assess the effect of this difference, areas having had vector control for a long time are clearly different from areas with no previous activities in many different aspects (entomological and human health parameters).

Sensitivity analysis: There weren't sufficient trials to conduct a sensitivity analysis to investigate the robustness of the results.

Controlled before-and-after studies

We analysed the study in the same manner as RCTs and presented the results in tables.

Interrupted time series

We analysed the study in the same manner as RCTs and presented the results in tables.

Assessment of risk of bias in included studies

see 5.4.4.

5.5 Results

5.5.1 Results of the search

We identified 132 potentially relevant studies. Of these we excluded 126 studies (for details of reasons see below and "Characteristics of excluded studies"). The remaining six studies m*et al* l the inclusion criteria. These trials are described below (for details see also "Characteristics of included studies").

5.5.2 Included studies

Trial design and location

Out of the six included studies, four were RCTs (Curtis 1998; Misra 1999; Mnzava 2001; Rowland 2000), and in all the allocation was by cluster (by villages, geographical blocks and sectors comprising several villages). One study was a CBA (Molineaux 1980) and one was an ITS (Sharp 2007).

Four trials were conducted in sub-Saharan Africa: Tanzania (Curtis 1998), South Africa (Mnzava 2001), Nigeria (Molineaux 1980) and Mozambique (Sharp 2007) and one was conducted in Pakistan (Rowland 2000) and one in India (Misra 1999).

Participants

The trials included either all ages (Misra 1999; Mnzava 2001; Molineaux 1980; Sharp 2007 for 1st year) or specific age groups (different groupings among children aged 1 to 15 years)(Curtis 1998; Rowland 2000; Sharp 2007 for subsequent years).

Intervention

RCT:

One trial compared the impact of IRS versus the provision of ITNs to all inhabitants (Mnzava 2001). Two trials had three arms and compared the impact of IRS to the impact of ITN and to an untreated control zone (Curtis 1998; Misra 1999). One trial studied the impact of IRS in comparison to a control area without any intervention (Rowland 2000). For IRS, all studies used pyrethroids as insecticide. Two of them used deltamethrin (dosage = 20 mg/m^2), and the two others used lambdacyhalothrin (30mg/m^2) and alphacypermethrin (25mg/m^2). Since there is no evidence to suggest that there is any difference between these insecticides in terms of impact and hence they were grouped for analysis. Two trials didn't specifically report the spray coverage (Mnzava 2001; Curtis 1998), for the other two (Misra 1999; Rowland 2000) it ranged from 92.2% to 96%. For treating ITNs, lambdacyhalothrin (10 mg/m^2 and 20mg/m^2), deltamethrin (25 mg/m^2) and permethrin (200mg/m^2) were used. Again, available evidence (Lengeler 2004) does not suggest any difference in impact between these three pyrethroids. Coverage rates with ITNs ranged in two trials from 85.4% to 100% (Misra 1999;

Mnzava 2001), while one trial didn't report coverage (Curtis 1998) but it was very high since nets were given for free to the whole population (Curtis C. personal communication).

In one of the three trials comparing IRS with ITNs (Mnzava 2001) IRS was done in the ITN areas before these were distributed. In the study area in KwaZulu-Natal there is a long history of IRS (around 50 years). Within the study area, all the houses in the ITN arm were sprayed in September 1996 before distributing the ITNs in January 1997. In subsequent years, house spraying was deliberately withdrawn in blocks with bed nets. In India (Misra 1999) there was a similar situation because of the long history of IRS in the study area, which started in 1953. IRS is also nowadays still a mainstay of malaria control in the study area.

CBA:

The impact of IRS was compared to a control area without any intervention (Molineaux 1980). Propoxur (2g/m²) was used as insecticide. The spray coverage ranged from 74% to 100%. There was no history of IRS in the area before this trial.

ITS:

In the study of Sharp 2007 the change over time due to IRS was examined over the time period of 3 years before and 4 years after the introduction of IRS. The insecticide used was bendiocarb (400mg/m²) and no usage coverage was mentioned. No history of IRS has been reported within the area before this study.

Some additional characteristics of the trials are given in Table 4.

Table 4: Trial characteristics of major factors influencing the impact of IRS

Trial	EIR[a]	Insecticide	Insecticide resistance	Main vector	Dominant wall type	Co-intervention(s)	Pre-trial control measures
Pakistan (Rowland)	< 1	Alpha cypermethrin	No	*A. stephensi*	Mud/brick	Treatment of fevers	IRS (for 25 years)
India (Misra)	< 1	Deltamethrin	yes[b]	*A.culicifacies*	Mud/brick	IEC[d]	IRS (for 60 years)
South Africa (Mnzana)	< 1	Deltamethrin	No[c]	*A.arabiensis*	N/A	IEC[d]	IRS (for 50 years)
Tanzania (Curtis)	> 1	Lambda cyhalothrin	No	*A.gambiae* *A.funestus* *A.arabiensis*	Mud	None	Clearing of malaria infections
Nigeria (Molineaux)	> 1	Propoxur	No	*A.gambiae* *A.arabiensis* *A.funestus*	Clay	None	None
Mozambique (Sharp)	> 1	Bendiocarb	No	*A.arabiensis* *A.funestus*	N/A	Treatment of slide - positive participants	None

Footnotes

[a] Transmission intensity (EIR: Entomological inoculation rate - indicates how many infectious mosquito bites a person receives on average per year)

[b] Mortality range for WHO susceptibility test: 74.4% to 96.5%

[c] Within other areas in Kwa-Zulu Natal, *A. funestus* was shown to be resistant to deltamethrin. However, there was no evidence that pyrethroid-resistant *A. funestus* were present in the area during the reported study
[d] IEC: Information, Education and Communication

Outcomes

Prevalence and incidence of malaria infections were the main outcomes that we could assess. Five studies looked at the prevalence rates of parasitaemia. Of these, two RCTs (Misra 1999; Rowland 2000) and one ITS (Sharp 2007) were conducted in unstable malaria settings and one RCT (Curtis 1998) and one CBA (Molineaux 1980) in stable malaria settings.

The incidence rate of malaria infections was assessed in four studies. Of these, three RCTs were conducted in unstable malaria settings (Rowland 2000; Misra 1999; Mnzava 2001) and one CBA in a stable malaria setting.

Impact on infant parasitological conversion rates was measured by Molineaux 1980, while incidence of re-infection and anaemia as additional outcomes were collected within stable malaria settings by Molineaux 1980 (CBA) and Curtis 1998 (ITS).

Infant mortality rates were measured by Molineaux (1980) but unfortunately not in a suitable control area, and hence this outcome could not be used. See Table 5 for details.

Table 5: Outcomes of studies

Study	**Study design**	**Comparison**		**Incidence of re-infection**	**Incidence of infections**	**Prevalence of infection**	**Anaemia**	**Infant parasite conversion rate**
		IRS vs control	**IRS vs ITN**					
Tanzania (Curtis 1998)	RCT	X	X	X	X	X	X	
South Africa (Mnzava 2001)	RCT		X		X			
Pakistan (Rowland 2000)	RCT	X			X	X		
India (Misra 1999)	RCT	X	X		X	X		
Nigeria (Molineaux 1980)	CBA	X			X	X		X
Mozambique (Sharp 2007)	ITS					X		

5.5.3 Excluded studies

126 studies were excluded due to the following reasons (for details see table "Characteristics of excluded studies" at the end of the paper):

- 40 didn't have enough units/arm (minimum required: RCT: 3 clusters per arm, CBA: 2 clusters per arm)
- 20 didn't have control sites which were comparable with the intervention sites
- 8 used an insecticide or a dosage not recommended by WHO
- 12 were only reviews or conference abstracts and did not provide enough data
- 28 were ITS which didn't provide enough data for pre- or post-intervention assessment
- 14 were ITS using a mix of interventions
- 2 trials didn't collect contemporaneous data for the control and intervention sites
- 5 measured non-eligible outcomes
- 1 trial included refugees as study participants
- 2 studies used a non - experimental approach (modelling)
- 1 RCT had a randomised allocation of the intervention that was not acceptable
- 2 studies had an IRS coverage under 60%
- 1 trial experienced a population movement of over 10%
- 1 trial sprayed with DDT in an area with documented DDT resistant *Anopheles.*

5.5.4 Risk of bias in included studies

For an overview of the risk of bias see Figure 1 and Figure 2.

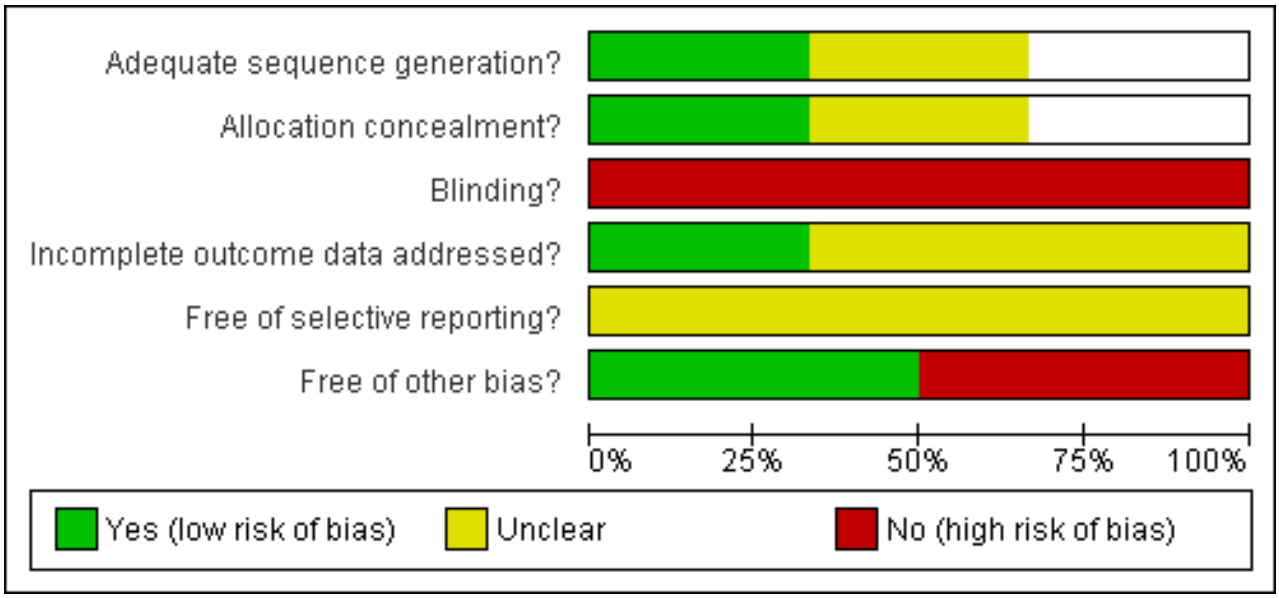

Figure 1: Methodological quality graph: review authors' judgements about each methodological quality item presented as percentages across all included studies.

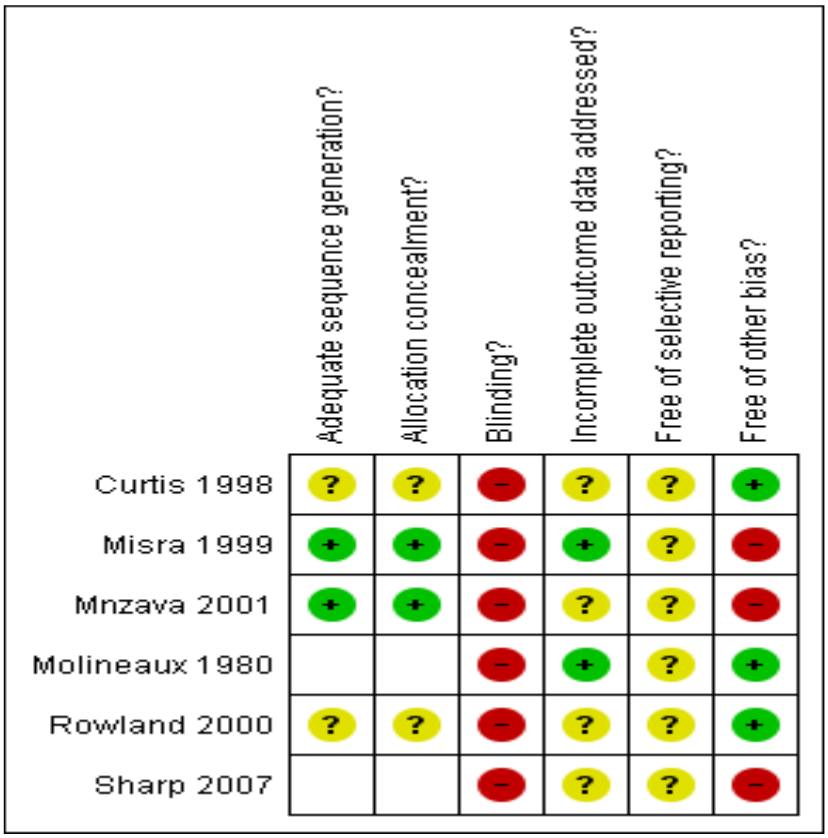

Figure 2: Methodological quality summary: review authors' judgements about each methodological quality item for each included study.

Allocation

Two of the four RCTs (Misra 1999; Mnzava 2001) generated allocation sequences by public drawing/tossing of coins. The risk of bias with these methods is low and allocation concealment is ensured by the fact that the allocation was made in public. The remaining two

RCTs (Curtis 1998; Rowland 2000) don't specify how the randomisation was done and how allocation concealment was secured. Therefore the risk for bias can't be assessed.

Blinding

Obviously, blinding is neither possible for IRS nor for ITNs and this criteria should therefore not be considered for assessing study quality. Given the type of interventions and the nature of the outcomes, the risk of bias resulting from the absence of blinding is thought to be minimal.

Incomplete outcome data

Two of the six included trials (Misra 1999; Molineaux 1980) reported changes in the number of participants over time. In one trial (Misra 1999) the losses to follow-up were under 10% and therefore the risk of bias was considered to be low. In the other trial (Molineaux 1980), there was a large migration with 15% to 20% of the population changing per year. The migration was described in detail within the study. Furthermore, the study did an analysis to check for the risk of bias due to the population movements and it was considered to be low.
The other trials didn't report on losses to follow-up. Since they were calculating "person-time-at-risk" as denominators the rates were accurate but the risk of selection bias over time could not be estimated.

Selective reporting

For none of the studies there was enough information to permit a reliable judgment of the risk of this type of bias.

Other potential sources of bias

In one trial (ITS, Sharp 2007) the spraying was interrupted for 2 years during the intervention phase, before being re-started. In addition, all age categories were sampled in December 1999, but subsequent surveys were confined to children two to 14 years of age. Unfortunately the authors did not give any details by age in the 1999 survey and hence we could not investigate the effect of that change. Whereas the first issue (interruption of spraying) obviously reduced the effect of spraying, the other issue (change in measured age group) might also lead to a lower apparent spraying efficacy.

Mnzava (2001) compared IRS versus ITNs. However, houses in bed net blocks had already been sprayed by the time the nets were distributed in 2007. Even though there was immediately an effort by investigators to re-plaster these houses to cover the insecticide on the walls, the effect of ITNs might be overestimate due to the dual protection for a limited

time. In subsequent years, house spraying was deliberately withdrawn in blocks with bed nets.

In the study of Misra (1999) a high incidence of plastering mud on the walls of houses was reported (Misra 1999). This most likely reduced the effectiveness of IRS and hence underestimated its real effect. Unfortunately the authors don't provide detailed data on the replastering to allow a judgment on its impact.

5.5.5 Effects of interventions

Randomised control trials

1. Stable malaria setting

Only one RCT assessed the impact of IRS in a stable malaria setting. Curtis (1998) compared the impact of IRS with lambdacyhalothrin versus ITNs treated with lambdacyhalothrin versus a control group with no intervention in a highly endemic malaria setting in Tanzania. The main study outcomes were (1) incidence of re-infection (active case detection in children aged one to five years, passive surveillance in all age-groups), (2) prevalence of infection (children aged one to five years) and (3) anaemia (children aged one to five years)(Table 5).

IRS vs no IRS:

IRS was shown to be effective in protecting children from re-infection with malaria over a 11 months period (protective efficacy: 54%)(Table 6). Malaria incidence (assessed by passive surveillance) was significantly reduced in children aged one to five years (RR 0.86, 95% CI 0.77 to 0.95 (unadjusted for clustering). For children older than five years no difference was seen in regard to malaria incidence (Table 7). For malaria prevalence no difference was seen between the IRS and the control group (Table 8). With regard to anaemia, the haemoglobin levels were significantly lower in the control group than in the IRS group (WMD 0.61 g/dl; 95% CI IRS group 9.99 to 10.02; 95% CI no IRS group: 9.38 to 9.40) (Table 9).

Table 6: RCTs: Impact on Incidence of re-infection in children aged one to five years (stable malaria only)

Comparison	Study	IRS[a]	no IRS[a]	Risk ratio	95% Confidence interval[b]	Protective efficacy
IRS versus no IRS:	Curtis 1998 (Tanzania)	468/3840	1014/3840	0.46	0.42 to 0.51	54%
IRS vs ITN:	Curtis 1998 (Tanzania)	468/3840	384/3840	1.22	1.06 to 1.40	-22%

Footnotes
[a] Denominator are person-weeks
[b] Not adjusted for clustering

Table 7: RCTs: Impact on parasite incidence

Comparison	Age groups	Study	IRS	Controls (no IRS or ITNs)	Rate ratio	95% Confidence Interval	Protective Efficacy
STABLE MALARIA (EIR >1), IRS versus no IRS							
Any infection	Children 1 to 5 years	Curtis 1998 (Tanzania)	228/413	304/471	0.86	0.77 to 0.95[b]	14%
	Older than five years	Curtis 1998 (Tanzania)	382/1007	365/984	1.02	0.91 to 1.15[b]	-2%
STABLE MALARIA (EIR >1), IRS versus ITN							
Any infection	Children 1 to 5 years	Curtis 1998 (Tanzania)	228/413	255/405	0.88	0.78 to 0.98[b]	12%
	Older than five years	Curtis 1998 (Tanzania)	382/1007	346/893	0.98	0.87 to 1.10[b]	2%
UNSTABLE MALARIA (EIR <1), IRS versus no IRS							
Any infection	all ages	Misra 1999 (India)	1497/19170	2195/21363	0.76	0.73 to 0.78[a]	24%
		Rowland 2000 (Pakistan)	6/12000	24/12000	0.14	0.06 to 0.33[a]	86%
P. falciparum	all ages	Misra 1999 (India)	834/19170	1244/21363	0.76	0.69 to 0.81[b]	24%
		Rowland 2000 (Pakistan)	25/12000	177/6000	0.07	0.02 to 0.39[a]	93%
P. vivax	all ages	Misra 1999 (India)	663/19170	951/21363	0.78	0.70 to 0.86[b]	22%
		Rowland 2000 (Pakistan)	112/6000	47/12000	0.21	0.10 to 0.55[a]	79%

UNSTABLE MALARIA (EIR <1), IRS versus ITNs						
Any infection	all ages	Misra 1999 (India)	834/19170	494/16885	1.55	1.49 to 1.60[a]
Any infection	all ages	Mnzava 2001 (South Africa)	2094/7649	1038/5450	1.44	0.77 to 2.70[c]

Footnotes
[a] adjusted for clustering
[b] not adjusted for clustering
[c]adjusted for matching, but not for clustering

Table 8: RCTs: Impact on parasite prevalence

Comparison	Age groups	Study	IRS	no IRS or ITN	Risk ratio	95% Confidence interval	Protective efficacy
1. STABLE MALARIA (EIR < 1), IRS vs no IRS:							
Any infection	Children 1 to 5 years	Curtis 1998 (Tanzania)	191/262	232/293	0.92	0.83 to 1.02[a]	8%
2. STABLE MALARIA (EIR < 1), IRS vs ITNs:							
Any infection	Children 1 to 5 years	Curtis 1998 (Tanzania)	191/262	211/293	1.01	0.90 to 1.13[a]	-1%
3. UNSTABLE MALARIA (EIR < 1), IRS vs no IRS							
Any infection	All age groups	Misra 1999 (India)	167/26085	237/26589	0.71	0.59 to 0.87[b]	29%
	Children 5 to 15 years:	Rowland 2000 (Pakistan)	72/12000	289/12000	0.25	0.08 to 0.80 (ICC = 0.01) 0.05 to 1.29 (ICC = 0.02)	75%
P. falciparum	All age groups	Misra 1999 (India)	127/26085	197/26589	0.66	0.53 to 0.82[b]	34%
	Children 5 to 15 years	Rowland 2000 (Pakistan)	5/1232	28/719	0.10	0.08 to 0.10[a]	90%
P. vivax	All age groups	Misra 1999 (India)	40/26085	40/26589	1.02	0.66 to 1.58[b]	-2%
	Children 5 to 15 years	Rowland 2000 (Pakistan)	30/1232	54/719	0.32	0.26 to 0.38[a]	68%
4. UNSTABLE MALARIA (EIR < 1), IRS vs ITNs							
Any infection	All ages	Misra 1999 (India)	167/26085	102/26849	1.69	1.32 to 2.15[b]	-69%
P. falciparum	All ages	Misra 1999 (India)	127/26085	114/25904	1.10	0.86 to 1.42[b]	-10%
P. vivax	All ages	Misra 1999 (India)	40/26085	29/26849	1.41	0.88 to 2.29[b]	-41%

Footnotes
[a] Adjusted for clustering, ICC= 0.01
[b] Not adjusted for clustering

Table 9: RCTs: Impact on Haemoglobin levels (stable malaria only)

Comparison	Study	Haemoglobin (in g/dl) (95% CI) n=number of participants	Haemoglobin (in g/dl) (95% CI) n=number of participants	WMD[a]
IRS vs no IRS	Curtis 1998 (Tanzania)	10.01 (9.99 to10.02) n = 752	9.39 (9.38 to 9.40) n = 850	0.61
IRS vs ITN	Curtis 1998 (Tanzania)	10.01 (9.99 to10.02) n = 752	10.00 (9.99 to 10.01) n = 909	0.01

Footnotes
[a] WMD: Weighted Mean Difference

IRS vs ITN:

For the incidence of re-infection, ITNs were shown in one RCT (Curtis 1998) to have a significantly greater protective effect than IRS in the Tanzanian trial (Risk ratio IRS:ITN = 1.22, CI 1.06 to 1.40) (Table 6). For children aged one to five years IRS showed a better protective effect than ITNs in reducing malaria incidence (RR 0.88, 95% CI 0.78 to 0.98 (unadjusted for clustering). No difference was seen for children older than five years (Table 7). Prevalence rates were found to be equal within the IRS and ITN groups (Table 8). No difference in haemoglobin levels were found (WMD 0.01; 95% CI IRS: 9.99 to 10.02; 95% CI ITN: 9.99 to 10.01) (Table 9).

2. Unstable malaria setting

Three clustered RCTs were done in unstable malaria settings (EIR < 1), of which one trial in Pakistan (Rowland 2000) assessed the impact of IRS vs a control group without intervention, one trial in India assessed IRS versus ITNs versus a control group (Misra 1999) and the last trial in South Africa assessed IRS versus ITNs only (Mnzava 2001). Two of the studies had incidence and prevalence as their main outcomes (Misra 1999 Rowland 2000), while one study assessed only incidence as outcome (Mnzava 2001).
All the three countries in which the trials have been contacted, have a long history of IRS. In Pakistan (Rowland 2000) IRS is the primary method of malaria control. But also India (Misra 1999) and KwaZulu Natal (Mnzava 2001) have a long history of IRS with 50 years and 60 years of spraying, respectively. The effect of these different durations of spraying before the start of the trials on the measures of impact could not be explored in the present analysis.

IRS vs no IRS:

IRS was shown to significantly reduce the incidence rate of malaria infections with the protective efficacy ranging from 24% to 86% (Table 7). IRS also significantly reduced the incidence of malaria when looking separately at *P.falciparum* and *P. vivax (P.falciparum:* RR 0.07, 95% CI 0.02 to 0.39, 1 trial (Rowland 2000); *P. vivax:* RR 0.21, CI 95% 0.10 to 0.55, 1 trial (Rowland 2000)*.*

Different results were seen when assessing the impact of IRS on malaria prevalence (Table 6). Overall, and for the all ages group, Misra 1999 did not find any effect of IRS in reducing malaria prevalence in India. However, looking at the effect of IRS on *P. falciparum* prevalence alone, in one year (1997) a statistically significant effect was seen, but not in the following year (1998). The opposite was found for *P. vivax*, where there was a significant effect in the second but not in the first year. In children aged five to fifteen in Pakistan, IRS reduced the risk of getting infected with *P. falciparum* as well as with *P.vivax* - by 90% and 68% *(*Rowland 2000, Table 8).

IRS vs ITN:

Slightly different results were seen when comparing IRS to ITNs against malaria incidence rates (Table 7). Whereas Misra 1999 found a significant difference between IRS and ITNs (RR 1.55, 95% CI 1.49 to 1.60), Mnzava 2001 did not find a significant difference (RR 1.44, CI 95% 0.77 to 2.70 - unadjusted for clustering)
Only one trial in India (Misra 1999) compared IRS to ITNs for the outcome of prevalence. ITNs were shown to give a better protection against any infection in 1998 but not in 1997 (Table 8).

Controlled before and after (CBA) and interrupted time-series (ITS) studies

Stable malaria setting

Two trials (one CBA in Nigeria Molineaux 1980 and one ITS in Mozambique Sharp 2007) were done in a stable malaria setting. They compared spraying with Propoxur (Molineaux 1980) and lambdacyhalothrin (Sharp 2007) to a control area. Both of them were looking at the impact on prevalence data, whereas Molineaux 1980 also assessed the infant parasitological conversion (incidence) rates.

IRS vs no IRS:

In both trials, the malaria prevalence was reduced where IRS was applied. However, Molineaux 1980 found only a significant difference for the prevalence rates during the wet season in which IRS showed a protective efficacy of 26% (Table 10). In Mozambique (Sharp 2007), the first year after spraying showed a clear drop in prevalence (2001), while in 2002 and 2003 IRS had stopped for operational reasons and prevalence went up again. In 2004 and 2005 IRS was resumed and prevalence had dropped again (Table 11).
Molineaux 1980 found that the infant parasitological conversion rates had a stronger reduction in the areas with IRS compared to areas without (Table 12). These authors also measured infant mortality rates (IMR) but unfortunately without measuring it in a control area.

They then derived evidence of impact from the close correlation between reduced infant parasitological conversion rates and IMR. It is unfortunate that because of this limitation these unique mortality data can not be used in our analysis.

Table 10: CBA: IRS versus no IRS: Crude parasite prevalence rates (seasonal average)

Comparison	Study	IRS	no IRS	Risk ratio	95% Confidence interval	Protective efficacy
STABLE MALARIA IRS vs no IRS: dry season: any infection	Molineaux 1980 (Nigeria)	700/2310	405/1261	0.94	0.85 to 1.04	6%
STABLE MALARIA IRS vs no IRS: wet season: any infection	Molineaux 1980 (Nigeria)	809/2310	599/1261	0.74	0.68 to 0.80	26%

Table 11 ITS: Prevalence of P. falciparum

Study	Year	1999	2000	2001[a,b]	2002[b]	2003[b]	2004	2005
Sharp 2007 (Mozambique)	**% (n)**	73 (101)	79 (120)	32 (130)	51 (117)	59 (118)	39 (120)	23 (114)
	95% CI[c]	62 to 82	70 to 86	23 to 43	38 to 64	49 to 69	28 to 52	15 to 34

Footnotes
[a] Start of spraying in February 2001 and data collection took place in June 2001
[b] Interruption of spraying between second half of 2001 to second half of 2003
[c] CI: Confidence interval

Table 12: CBA: Infant parasitological conversion ratesa of P.falciparum (stable malaria)

Study	Treatment	year		
		1971[b]	**1972[c]**	**1973[c]**
Molineaux 1980 (Nigeria)	IRS	0.012	0.002	0.002
	no IRS	0.016	0.005	0.009

Footnotes
[a] Infant parasitological conversion rate: -ln(1-p)/t, where p= conversion rate and t= time
[b] Before intervention
[c] After intervention

5.6 Discussion

Since the 1950s, IRS has been used widely in many areas of the world, especially in Asia, Latin America and Southern Africa. IRS with DDT and other insecticides has been one of the main interventions leading to the elimination of malaria in about half of the world's regions (Lengeler 2003). This was for example the case in much of southern Europe, North America, Japan, Central Asia and Latin America. Very low levels of malaria transmission have been achieved and maintained in countries as different as India, Tadjikistan and Colombia. Hence the effectiveness of this intervention is beyond doubt. Unfortunately, the epidemiological effect has never been quantified properly, so that a comparison with other malaria control interventions, for example with ITNs, is impossible. As a result, an accurate comparative cost-effectiveness assessment is also impossible.

With the exception of Southern Africa (South Africa, Namibia, Botswana, Swaziland Zimbabwe) and the Ethiopian and Madagascar highlands, the implementation of IRS in the highly endemic areas of sub-Saharan Africa has been limited in geographical extent and usually only for a limited time period. Recently, IRS was introduced in southern Mozambique, Equatorial Guinea, Zambia, Ghana, Sao Tome, Zanzibar. There is also a new interest for IRS since 2007 in the wake of the United States President's Malaria Initiative (PMI). For many of the other endemic countries of SSA, vector control has been upscaled from 2000 onwards through the increased deployment of ITNs. In this context, two important questions have emerged: (1) what are the comparative advantages, including feasibility, cost and impact of ITNs and IRS; and (2) is there any benefit in combining both IRS and ITNs together to increase impact, especially in view of the goal of malaria elimination declared in 2007.

While there are now relatively good data on comparative feasibility and cost (see review by Yukich 2008) this review demonstrates a great paucity of high-quality evidence in the comparative assessment of health impact. There are too few high-quality randomised controlled studies on the health effects of IRS, with too few health outcomes and not enough geographical coverage. Only six out of 132 identified studies met the inclusion criteria (four RCTs, one CBA and one ITS) and not all key malariological outcomes were addressed within these studies. Unfortunately, none of the studies investigated the potential of IRS for reducing child mortality rates. In some ways, this results is not entirely surprising considering the fact that (1) IRS started to be implemented on a large scale after the invention of DDT in 1943 (and hence before the conduct of the first RCT in 1948), and (2) IRS with DDT was outstanding in its health effects from the start and there was no strong rationale for public health officials in the 1950s and 1960s to test formally its effects.

Currently, our evidence on the second question (the impact of the combination of IRS with ITNs) stems only from very limited descriptive evidence and properly conducted RCTs are urgently required.

Overall, the formal quality of the six included trials was considered to be satisfying. Two of the four RCTs used appropriate methods for sequence generation and allocation concealment, whereas the other two trials didn't mention their procedure. However, given the nature of the intervention and the fact that it is allocated by cluster, this is unlikely to lead to bias in the results. Due to the nature of the intervention, blinding is not possible, but no risk of bias is expected because of this.

A much bigger issue for the validity of the results is the implementation of the interventions, which has been sub-optimal in some settings. IRS application was for example discontinued in one setting (Sharp 2007). In three (Misra 1999; Mnzava 2001; Rowland 2000) of the four RCTs, the control and/or ITN arms of the trials had a long previous history of IRS, and spraying was simply suspended for the duration of the trial. Obviously, the entomological baseline situation was not any more one of an untouched area. In addition, insecticide was sprayed shortly before ITN distribution in the ITN arms in India and Tanzania. Despite the best efforts by investigators to minimize the effects of this by re-plastering the walls, this is still likely to have had an independent effect on the outcomes. Unfortunately, it is impossible to quantify that effect.

Only two different classes of insecticides (carbamates and pyrethroids) were used in the reviewed trials, to which the mosquitoes were fully susceptible in all settings. Insecticide resistance is an obvious threat to the effectiveness of IRS. However, unlike 50 years ago, when DDT was the only insecticide on hand, there are now 12 different insecticides within four different classes available for IRS. This gives the possibility to alternate the insecticide and to switch to others insecticides in case resistance appears. This is a clear advantage over ITNs, for which only one class of insecticide is available. On the other hand, ITNs still offer a physical barrier to the vector, even if the insecticide doesn't work anymore, whereas for IRS the protection through the insecticide will be strongly reduced when resistance occurs.

The four randomised controlled trials meeting the inclusion criteria were distributed between Asia and sub-Saharan Africa. Only one of them was done in a stable malaria setting (entomological inoculation rate >1). Within this study (Curtis 1998, Tanzania) the risk for children under six to get re-infected with malaria was reduced approximately by half

(protective efficacy: 54%), but no change in incidence or prevalence rates was seen, which was surprising. However, the participants got treated at the start of the study to clear all infections. This is usually not the case in interventions and normally a reservoir of infected people is always present. Hence, the generalization of the results to other malaria endemic areas is questionable.

Three trials were done in unstable malaria settings (entomological inoculation rate <1). Two of them in Asia (Misra 1999 (India); Rowland 2000 (Pakistan)) and one in Africa (Mnzava 2001 (South Africa)). Combining the trials from India and Pakistan showed that IRS significantly protects all age groups in these settings from malaria infections, irrespective of the type of infection. There was no difference seen in the effectiveness when comparing IRS with ITNs.

The results of this review do not reconcile well with the impressive historical reductions of malaria in many areas of the world following the introduction of IRS. Among these areas were also a number of high transmission areas of Africa (Kouznetsov 1977; Mabaso 2004). Hence, the lack of positive evidence from formal trials should not, in the case of IRS, be interpreted as a lack of effect of the intervention, but rather as the consequence of a lack of high-quality and long-duration trials. As a result, the main aim of the review (to quantify the health effects of IRS) could not be achieved and our major conclusion is that high-quality evidence from RCTs is still required. For obvious ethical reasons a control group without vector control intervention is not acceptable any more and such trials should therefore have at least two arms, an IRS arm and an ITN arm. Given the importance of also assessing the combined effect of IRS and ITNs a third arm with both interventions together would be highly desirable.

5.7 Authors' conclusions

Implications for practice

- Overall, good quality evidence on the impact of IRS is scarce.

- The current evidence is insufficient to quantify properly the effect of IRS in high transmission settings.

- At present, a quantitative epidemiological comparison between IRS and ITNs is not possible.

- Available good quality evidence confirms that IRS with pyrethroids works in reducing malaria in unstable malaria settings

- No trial investigated the effect of IRS in reducing (child) mortality.

- There is insufficient epidemiological evidence to assess the effect of other determinants of impact, such as the insecticide class used for IRS, the type of transmission, the dominant vector species, socio-cultural determinants.

Implications for research

- There is an urgent need for more RCTs comparing IRS with ITNs in a number of settings with different epidemiological and socio-cultural characteristics.

- Ideally, such RCTs should have a third arm with a combination of high coverage IRS with high coverage ITNs.

- Participants of all age-groups should be included in such trials.

5.8 Acknowledgements

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5.9 Contributions of authors

BP: reference identification, screening retrieved papers for eligibility, data extraction, analysis of data, main writing. FT: preliminary reference identification, screening retrieved papers for eligibility, contribution to writing. CL: initial concept, organization of reference searching, contribution to writing. BS: Initial concept, technical inputs in technical matters. Sadly, BS passed away during the initial phase of the review. His experience, drive and good nature are sorely missed.

Declarations of interest

None known.

Differences between protocol and review

We planned to convert the outcomes for anaemia presented as g/dL into packed cell volume with a standard factor of 1:3. But Carneiro 2007 showed that this conversion is not always accurate and since we only had one trial providing haemoglobin measures, we presented them as they were presented within the paper (in g/dL).

We did not find individually randomised RCTs in the frame of this review. However, the methods of extracting data and analysing such trials would follow the methods outlined in the protocol.

We did not do a sensitivity analysis due to the small number of trials. However, the methods published in the protocol will be followed, if appropriate, in future updates of this review.

We only found one eligible ITS for our review and presented its results as shown in the paper. If further trials will be included in future updates we will analyse them as published in the protocol.

Methological quality was assessed using the Cochrane Collaboration's risk of bias tool (Higgins 2008).

No summary of the major debates and findings of other reviews on DDT was included in the discussion as none of our included studies used DDT as insecticide.

There was a change in the authorship. BP replaced FT as first author because she took the lead in this work.

5.10 Characteristics of studies

5.10.1 Characteristics of included studies

Curtis 1998

Methods	Study design: cluster randomised controlled trial. Unit of allocation: village. Number of units: 4:4:4 Length of follow-up: prevalence surveys and passive surveillance: 15 months (3rd & 4th quarter 95 to 4th quarter 96). Incidence surveys: 20 months (April 95 to Dec 1996). Incidence of re-infection was monitored once before the interventions and four times after introduction of intervention (once in each quarter of 1996) by taking weekly blood slides. Cross-sectional surveys were carried out monthly from April 1995 to December 1996. In addition, a passive surveillance system was set up. People feeling sick with fever were encouraged to visit a local research assistant, who was taking a blood slide from them, which were collected weekly. Different children were used for each cohort. Confidence intervals were not adjusted for clustering. We could retrospectively adjust for the prevalence, but not for the incidence data.

Participants	Number of participants: Incidence: 60:60:60 (control:ITNs:IRS), prevalence: 104:93:86, passive surveillance: 500:357:795. Inclusion criteria: incidence: children aged 1 to 6 with cleared pre-existing parasitaemia; prevalence: children aged 1 to 6, passive surveillance: people of all ages feeling sick with fever. Exclusion criteria: incidence: children away from home, for having missed the blood slide for more than 1 week; prevalence: children already included in the incidence group, children which were selected in the previous month and children with parasitaemia >4000/µl. Passive surveillance: no specific exclusion criteria mentioned.
Interventions	IRS: Microencapsulated lambdacyhalothrin (ICON™) 10%; dosage: 30 mg/m². The wall and roof areas were sprayed with Hudson X-Pert spray pumps. Re-spraying in the villages was carried out seven to eight months after the initial spraying (July to August 1996). The spray coverage was not specifically mentioned but maximal coverage was aimed for. ITNs: Lambdacyhalothrin (ICON™); dosage: 10 mg/m² in 2 villages, and 20 mg/m² in the other 2 villages. Retreatment after seven months. The coverage rate was not specifically mentioned.
Outcomes	(1) Incidence of re-infection after parasitological clearance with antimalarials (2) Malaria prevalence (3) Haemoglobin levels
Notes	Study location: six villages near Muheza, Tanga Region and six villages near Hale, both in northeast Tanzania EIR: estimated to be above 300 Malaria endemicity: high endemicity with intense perennial transmission Transmission season: April to June Main vector: *Anopheles gambiae*, *Anopheles funestus* and *Anopheles arabiensis* Material of wall sprayed: Mud Insecticide resistance: None (bioassay test showed mortality of 80-100%)

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Unclear	Quote: "Random assignment of intervention" Comment: Insufficient information to permit a judgement
Allocation concealment?	Unclear	Quote: "Random assignment of intervention" Comment: Insufficient information to permit a judgement
Blinding?	No	
Incomplete outcome data addressed?	Unclear	The study did not address this outcome
Free of selective reporting?	Unclear	There is insufficient information to permit a judgement
Free of other bias?	Yes	

Misra 1999

Methods	Study design: cluster randomised controlled trial. *Baseline:* Unit of allocation: village. Number of units: 15:15:15. Length of follow-up: two weeks (16th to 30th September 1996). Mass surveys in September 1996 (peak transmission season) in 45 villages within the three ecological zones. *Intervention:* Unit of allocation: groups of three nearby villages (but at least one km apart) formed a cluster for random assignment of IRS, ITNs or control). The villages were not evenly distributed within the three ecological zones, 30 in coastal zone, 51 in the plains zone and 45 within the foothill eco zone. The distribution of the randomised villages was equal within the three groups. Number of units: 42:42:42. Length of follow-up: 18 months. Active case detection by home visits twice a week by collecting blood smears from all fever cases. On Sundays, treatment was provided to any sick person calling on the health worker. Monitoring from May 1997 until May 1999 (only data collected until 31.12.1998 were evaluated). Cross-sectional mass surveys once per year within the second half of September in 1997 and 1998. Drop out rates were 6.0% for IRS, 5.2% for ITN and 5.6% for the control group. Confidence intervals were adjusted for clustering for the incidence data when *P. falciparum* and *P. vivax* were combined, using a Poisson regression model with random effects. The retrospective adjustment of incidence confidence intervals for *P. falciparum* and *P. vivax* separately was not possible. Neither was the adjustment for prevalence data possible.
Participants	*Baseline mass survey:* Number of participants: 34,292. No explicit inclusion/exclusion criteria (all ages). *Intervention:* Number of participants: 93,210 (IRS: 30,989; ITNs: 31,168; control: 31,053). No explicit inclusion/exclusion criteria.
Interventions	The first intervention round took place from 26.5. to 14.6.1997; the second round from 23.5 to 14.6.1998. IRS: deltamethrin 2.5% WP; dosage 20 mg/m². Indoor surfaces of the walls, ceiling, back of cupboards, cots, eaves and cattle sheds were sprayed. Overall spray coverage was 92.2% in 1997 and 95.1% in 1998. In 1997 spray coverage was least in the irrigated plain eco zone (87%) and over 95% in the foothill and coastal eco zone. In 1998, coverage was 95% in all 3 zones. 1.36% and 19.3% of the houses which have received IRS were re-plastered after three and six months, respectively. ITNs: deltamethrin 2.5% SC; dosage 25 mg/m². Overall net coverage of the whole population was very high, with 99.3% in 1997 and 85.4% in 1998, respectively. Overall, 86.8% of the nets were retreated, with the retreatment taking place one year after the distribution (May 1998). In the irrigated plain eco zone, 88.3% were retreated, whereas 95.7% of the nets in the foothill eco zone were retreated. The

	coverage rate for the coastal area was not mentioned. Only 4% of the nets were washed after nine months of treatment.
Outcomes	(1) Malaria prevalence. (2) Malaria incidence.
Notes	Study location: Surat District in Gujarat State, India. 3.7 million population with 54% distributed in 1281 villages. The district is divided into three ecological zones: (1) Foothill: Eastern tract of hilly land, largely deforested, summer hot and dry, maximal rainfall (2) Irrigated plain: cultivation of paddy, sugar cane and plantains, dam which irrigates the entire plain area (3) Coastal: Western coast belt with sandy soil and heavy industries. EIR: <1. Malaria endemicity: coastal area: hypo-endemic; getting hyperendemic towards eastern hilly tracts. Transmission season: perennial transmission with peak from June to September. Main vector: *Anopheles culicifacies* (zoophilic and endophilic). Material of wall sprayed: mud or cement. Insecticide resistance: *A. culicifacies* is resistant to DDT and malathion, but highly susceptible to deltamethrin, the insecticide used within the study (mortality range for susceptibility tests: 74.4 to 96.5) Net ownership prior to distribution of nets for the trial was significant higher in the IRS group (16.5%) than in the other two groups (ITN:9.5%; control: 15.5%; χ^2 test=144.69, df=2, p=0.001)

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Yes	Quote:"Public drawing, witnessed by elected leaders, community members, and project government officials". Comment: Valid as randomisation procedure.
Allocation concealment?	Yes	Quote: "Public drawing, witnessed by elected leaders, community members, and project government officials". Comment: As there were many witnesses, the adherence to the randomised allocation is secured.
Blinding?	No	
Incomplete outcome data addressed?	Yes	Losses to follow-up: Control: 1750/31053 losses to follow-up (344/31053 due to death; 1215/31053 due to emigration and 191/31053 married and moved away). IRS: 1866/30989 losses to follow-up (319/30989 due to death; 1332/30989 due to emigration and 215/30989 married and moved away). ITN: 1611/31168 losses to follow-up (346/31168 due to death; 1034/31168 due to emigration and 231/31168 married and moved away). Additions to the study population: Control: 1035/31053 additions (603/31053 due to birth, 249/31053 due to marriage and 183/31053 due to immigration. IRS: 1250/30989 additions (713/30989 due to birth; 305/30989 due to marriage and 232/30989 due to immigration). ITNs: 1424/31168 additions (704/31168 due to birth; 320/31168 due to marriage and 400//31168 due to immigration).

Free of selective reporting?	Unclear	There is insufficient information for judgement
Free of other bias?	No	Quote: "a high incidence (24.3%) of plastering mud on the walls of houses..." (Bhatia *et al.* 2004) Comment: the re-plastering of the walls after spraying most likely reduced the effectiveness of IRS

Mnzava 2001

Methods	Study design: cluster paired randomised controlled trial. Unit of allocation: geographical blocks (seven pairs of blocks formed on the basis of their average malaria incidence rate (being as similar as possible within each pair); randomisation to ITNs or IRS within each pair. Number of units: 7:7 blocks. Length of follow-up: 24 months. Routine active case detection took place monthly by malaria control teams. Blood slides were taken from any member of a household. Passive case detection was done by clinic and hospital staff. Whenever such a case was detected, all household members from where the case came from and including all people living within a 40 km radius of the homestead where the index case occurred, were bled as well. This is the routine procedure for all zones under malaria control in the KwaZulu Natal Province. Monitoring from January 1997 to December 1998. Drop-outs were not taken into account. Confidence intervals were not adjusted for clustering. They could not be adjusted retrospectively (due to matching of the pairs).
Participants	IRS: 7649 ITNs: 5450 No inclusion/exclusion criteria mentioned (all ages).
Interventions	IRS: spraying with deltamethrin; dosage 20 mg/m², yearly from September to December, starting in 1996 (prior to the malaria season). The interior walls, ceilings and eaves of all homesteads were sprayed with Hudson X-Pert spray pumps. Spraying coverage is not explicitly mentioned. ITNs: distribution of permethrin treated nets in January 1997; target dose 200 mg/m². Annual retreatment in January, using deltamethrin (KO-Tab) in 1998 and permethrin in 1999. Over 90% of the nets were retreated. Usage of ITN in 1997 was 98% and 100% in 1998. Houses in bednet blocks had already been sprayed by the time the nets were distributed in 2007. There was immediately an effort by investigators to re-plaster these houses to cover the insecticide on the walls. In subsequent years, however, house spraying was deliberately withdrawn in blocks with bednets.
Outcomes	(1) Malaria incidence (cases per 1000 person-years)
Notes	Study location: homesteads within Ndumu and Makanis areas of Ingwavuma district in KwaZulu Natal Province. 14,000 inhabitants (predominantly Zulus) served by four clinics and one referral hospital. The area has a long history of IRS. Before 1995, it was sprayed with DDT, thereafter there was a switch to deltamethrin. EIR: not known but very low because of long-standing malaria control efforts (decades of IRS). Malaria endemicity: not known (annual malaria incidence of 5%). Main vector: *Anopheles arabiensis* (after elimination of *A. funestus* by IRS).

	Material of wall sprayed: mud and cement. Insecticide resistance: within KwaZulu Natal Province, *A. funestus* was shown to be resistant to deltamethrin (Hargreaves 2000). However, according to Mnzava 2001 there is no evidence that pyrethroid-resistant *A. funestus* occurred in the area during the period of the study. For *A. arabiensis* no resistance was detected in the area.

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Yes	Quote: "Tossing of a coin during community meeting" Comment: Valid as randomisation tool
Allocation concealment?	Yes	Quote: "Tossing of a coin during community meeting" Comment: As there were many witnesses, the adherence to the randomised allocation is secured
Blinding?	No	
Incomplete outcome data addressed?	Unclear	Quote: "Any population changes over the study period could not be taken into account"
Free of selective reporting?	Unclear	There is insufficient information to permit a judgement
Free of other bias?	No	Houses in bed net blocks had already been sprayed by the time the nets were distributed in 2007. There was immediately an effort by investigators to re-plaster these houses to cover the insecticide on the walls. In subsequent years, however, house spraying was deliberately withdrawn in blocks with bed nets.

Molineaux 1980

Methods	Study design: controlled before-and-after study Unit of allocation: village Number of units: 5:6 (control : IRS) Every 10 weeks, house-to-house visits were done and a thick film taken. In case of absence a second visit to the home was done. Length of follow-up: 36 months. Drop-out rate unknown, high levels of migration - 15% to 20% per year.
Participants	IRS: 2310 Control: 1861 No explicit inclusion/exclusion criteria mentioned (all ages).
Interventions	Propoxur 50% WP; dosage 2 g/m². Three rounds of spraying were applied in 1972, starting on 1 May, 5 July and 6 September, respectively. The intervals between successive rounds in the same village were 61 to 66 days. In 1973 spraying was applied in April, June and August and in the southern part in October. The intervals between successive rounds were 56 to 66 days. Spray coverage: 74% to 100% (99% on average).
Outcomes	(1) Prevalence rate. (2) Incidence rate. (3) Infant mortality rate
Notes	Study location: Garki District in Northern Nigeria.

	EIR at baseline in treated villages: wet season:18 to 132; dry season: 0 (except Sugungum:13). EIR at baseline in untreated villages: wet season:17 to 37; dry season: N/A. Malaria endemicity: stable, seasonal. Main vector: *Anopheles gambiae* and *Anopheles funestus.* Material of wall sprayed: clay. Insecticide resistance: None mentioned.

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Unclear	
Allocation concealment?	Unclear	
Blinding?	No	
Incomplete outcome data addressed?	Yes	
Free of selective reporting?	Unclear	There is insufficient information to permit a judgement
Free of other bias?	Yes	

Rowland 2000

Methods	Study design: cluster-randomised controlled trial. Unit of allocation: sectors; the study area comprising 60 villages was divided into nine sectors of approximately equal population size and surface area and then each sector was assigned at random to control, Wettable Powder, or SC formulation). Number of units: 3:3:3 sectors. During analysis the two insecticide groups (WP and SC) were merged into a single group because there was no evidence of difference between them. Length of follow-up: 10 month (one season). Active case detection by home visits every fortnight. Blood slides were taken from any member of a household reporting to having had fever during the previous three days. Monitoring from April 1997 to January 1998, covering the entire malaria transmission season. Two cross-sectional surveys were carried out in April-May and September 1997, i.e. before and after the spraying, which was done in June 1997 (one survey within and one survey outside the malaria season, which runs from June to November). To assess the prevalence rate, blood slides were taken from children of one or two schools selected from sentinel villages in each sector. Drop-out rates unknown. Confidence intervals were not adjusted for clustering by authors. We could adjust the incidence and prevalence data retrospectively. See Data collection and analysis for more details.The rate ratio (RR) of IRS vs no IRS was estimated by a generalized linear model with negative binomial mean and variance functions. This model sowed to best fit the observed cluster-level incidence rates (Generalized Pearson statistics=1.29).
Participants	(1) Active case detection: Number enrolled:18,000 (2000 in each of the 9 sectors). Inclusion criteria into active surveillance group: any member of a household who reported having had fever during the previous 3 days. Exclusion criteria: No explicit exclusion (all ages). (2) Cross-sectional surveys:

	Inclusion criteria: School children aged 5 to 15 years present in school on the day of the survey. Number enrolled: 200 to 300 children per sector. Exclusion criteria: none.
Interventions	Alpha-cypermethrin WP and SC; dosage 25 mg/m²; living quarters, storage rooms and animals shelters were sprayed with Hudson X-pert spray pumps Spray coverage: WP: 96%, SC: 97%.
Outcomes	(1) Malaria incidence through active case detection (*P. falciparum* and *P. vivax*). (2) Malaria prevalence through cross-sectional surveys (*P. falciparum* and *P. vivax*).
Notes	Study location: 3 Union Councils, covering 180 km² of Sheikhupura District, Punjab Province, approximately 60 km west of Lahore, Pakistan. EIR: < 1 Malaria endemicity: not known (annual incidence of 50 episodes per 1000 person years). Malaria season: June to November. Main vector: *Anopheles stephensi*. Material of wall sprayed: mud and brick. Insecticide resistance: no detected resistance, 100% mortality of laboratory-reared and wild-caught *A. stephensi*.

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Unclear	Quote: "..each sector was assigned at random to untreated, WP, or SC spraying..." Comment: Insufficient information for judgement
Allocation concealment?	Unclear	Insufficient information for judgement
Blinding?	No	
Incomplete outcome data addressed?	Unclear	This outcome was not addressed by the study
Free of selective reporting?	Unclear	There is insufficient information to permit a judgement
Free of other bias?	Yes	

Sharp 2007

Methods	Study design: interrupted time series. Length of follow-up before intervention: 3 years (1999 to 2001). Length of follow-up after intervention: 4 years (2002 to 2005). Cross-sectional studies were done once per year (in June) within 26 sentinel sites. From a random sample of individuals malaria infections were tested by Rapid Diagnostic Tests (RDTs).
Participants	First year (1999): all age groups included. In subsequent years (2000 to 2005) children between 2 to 14 years were included in surveys.
Interventions	Bendiocarb; 400 mg/m². Twice annual spraying was done using Hudson pumps. Spraying personnel were trained in spraying techniques, safety measures and received personal protection equipment.
Outcomes	(1) Malaria prevalence of *P. falciparum*
Notes	Study location: Maputo Province in Southern Mozambique. EIR: > 1 before control activities started.

	Malaria endemicity: stable malaria before control activities started. Main vector: *A. arabiensis; A. funestus*. Material of wall sprayed: Not known. Insecticide resistance: None.

Risk of bias table

Item	Judgement	Description
Adequate sequence generation?	Unclear	
Allocation concealment?	Unclear	
Blinding?	No	
Incomplete outcome data addressed?	Unclear	This was not addressed by the study.
Free of selective reporting?	Unclear	There is insufficient information to permit a judgement.
Free of other bias?	No	Quote 1: "IRS was interrupted from 2001 to 2002 because of resource constraints, but resumed in the second half of 2003". Comment: Due to the interruption of IRS, it is likely that the effect of the spraying will be underestimated. Quote 2: "All age categories were sampled in December 1999, and subsequent surveys were confined to children two to 14 years of age". Comment: In a malaria endemic area, the risk of infection is higher for children than for adults. Therefore the prevalence might be affected when comparing the year with all age groups compared to the years with children only. However, looking at the difference in the prevalence rates of the year with all age-groups surveyed versus subsequent years, this effect seems negligible.

5.10.2 Characteristics of excluded studies

Afifi 1959
Reason for exclusion Data collection for groups not contemporaneous.
Afridi 1947
Reason for exclusion Not enough units/arm.
Alves 1953
Reason for exclusion Usage of an insecticide (Benzene Hexachloride (BHC)) which is not recommended by WHO
Andrews 1951
Reason for exclusion Review, not enough data to analyse.
Ansari 1986
Reason for exclusion Inappropriate choice of control site (Hexachlorocyclohexane (HCH) spraying).
Ansari 1990
Reason for exclusion Inappropriate choice of control site (HCH spraying).
Ansari 2004
Reason for exclusion Inappropriate choice of control site (HCH spraying).

Ansari 2004a

Reason for exclusion Inappropriate choice of control site (Malathion spraying); not enough units/arm.

Arredondo-Jimenez 1993

Reason for exclusion Not enough units/arm.

Arredondojimenez 1993

Reason for exclusion Inappropriate choice of control site (DDT spraying).

Barai 1982

Reason for exclusion Review, not enough data to analyse.

Barutwanayo 1991

Reason for exclusion ITS with mix of interventions (IRS, ITN, drainage and improvement of health system).

Bhatnagar 1974

Reason for exclusion Dosage of insecticide application not concurrent with WHO recommendations.

Bradley 1991

Reason for exclusion Usage of an insecticide (Benzene Hexachloride (BHC)) which is not recommended by WHO

Brieger 1996

Reason for exclusion Not enough data to analyse (denominators missing); author was contacted but could not supply missing data.

Cai 1999

Reason for exclusion Not enough units/arm.

Cavalié, 1961

Reason for exclusion Not enough data for pre- and post-intervention assessment.

Cavalié, 1991

Reason for exclusion Not enough data for pre- and post-intervention assessment.

Charlwood 1995

Reason for exclusion Inappropriate choice of control site (lambdacyhalothrin spraying); not enough units/arm; not enough data for pre-intervention assessment.

Conteh 2004

Reason for exclusion Non-eligible outcomes measured.

Coosemans 1978

Reason for exclusion Review, not enough data to analyse.

Coosemans 1989

Reason for exclusion Not enough data for quality assessment and analyses of the data.

Coosemans 1991

Reason for exclusion ITS with mix of interventions (IRS, ITN, drainage and improvement of health system).

Coppen 1999

Reason for exclusion Not enough data, conference abstract only; author could not be contacted for additional information.

Cot 1999

Reason for exclusion Not enough data, conference abstract only; author could not be contacted for additional information.

Cot 2001

Reason for exclusion not enough units/arm.

Cot 2002

Reason for exclusion Non-eligible outcomes measured.

Curtis 1999

Reason for exclusion Not enough data, conference abstract only.

Curtis 2000

Reason for exclusion Review, not enough data to analyse.

Dapeng 1996

Reason for exclusion Interrupted times series with mixed malaria control interventions (IRS and ITN).

Das 1987

Reason for exclusion Inappropriate choice of control site (DDT spraying).

de Zulueta 1954

Reason for exclusion Not enough units/arm; collection of data in control and survey area at different time points.

de Zulueta 1961

Reason for exclusion Not enough data for pre-intervention assessment; ITS with mixed interventions (IRS and treatment).

Deane 1948

Reason for exclusion Not enough data for pre/post-intervention assessment.

Dhiman 2005

Reason for exclusion ITS with mixed interventions (IRS and treatment).

Dodge 1965

Reason for exclusion Not enough data for pre-intervention assessment.

Doke 2000

Reason for exclusion Not enough data for pre/post-intervention assessment.

Dowling 1950

Reason for exclusion Inappropriate choice of control site.

Dowling 1951

Reason for exclusion Inappropriate choice of control site.

Eddey 1944

Reason for exclusion Not enough data for post-intervention assessment.

Edeson 1957

Reason for exclusion Not enough units/arm.

Farid 1954

Reason for exclusion ITS with mixed interventions (IRS and larviciding); mix of refugee and general population.

Faye 1992

Reason for exclusion Not enough units/arm; wrong dosage of insecticide (1g/m2 fenitrothion).

Fontaine 1976

Reason for exclusion Not enough units/arm.

Fontaine 1978

Reason for exclusion Not enough units/arm.

Gandahusada 1984

Reason for exclusion Non randomised allocation of intervention.

Gill 1997

Reason for exclusion Not enough units/arm.

Gunasekaran 2005

Reason for exclusion IRS coverage below 60%.

Guyatt 2002

Reason for exclusion Inappropriate choice of control site.

Hamon 1954

Reason for exclusion ITS with mixed interventions (IRS, larviciding and drug distribution).

Hii 1993

Reason for exclusion Study sites not comparable.

Ismail 1974

Reason for exclusion Not enough data for post-intervention assessment.

Ismail 1975

Reason for exclusion High level of population movement.

Ismail 1978

Reason for exclusion Not enough data for pre- and post-intervention assessment.

Jaggi 1984

Reason for exclusion Not enough data for pre-intervention assessment.

Jambou 2001

Reason for exclusion Control group not comparable; not enough data for pre-intervention assessment.

Kamolratanakul 2001

Reason for exclusion Not enough data for quality assessment.

Kere 1992

Reason for exclusion Non-eligible outcomes measured.

Kleinschmidt 2006

Reason for exclusion Not enough data for pre- and post-intervention assessment.

Kleinschmidt 2007

Reason for exclusion Not enough data for pre-intervention assessment.

Lambrecht 1952

Reason for exclusion Not enough data for pre-intervention assessment.

Lantoarilala 1998

Reason for exclusion Not enough units/arm.

Lópes 1993

Reason for exclusion Mix of intervention (IRS and drug distribution).

Maharaj 2005

Reason for exclusion Not enough data for post-intervention assessment.

Mastbaum 1951

Reason for exclusion Not enough units/arm.

Matola 1981

Reason for exclusion Usage of an insecticide (Dieldrin) which is not recommended by WHO.

Metselaar 1954

Reason for exclusion Not enough units/arm.

Metselaar 1957

Reason for exclusion Not enough units/arm.

Metselaar 1960

Reason for exclusion Not enough data for pre-intervention assessment.

Metselaar 1961

Reason for exclusion ITS with mixed interventions (IRS and drug distribution).

Mnzava 1993

Reason for exclusion Not enough units/arm.

Najera 1965

Reason for exclusion Not enough units/arm; Inappropriate choice of control site.

Najera 1967

Reason for exclusion Not enough units/arm.

Najjar 1959

Reason for exclusion Not enough data for pre-intervention assessment.

Nalim 1997

Reason for exclusion Not enough units/arm.

Nasir 1982

Reason for exclusion Not enough data on parasitological assessment.

Nguyen 1996

Reason for exclusion Not enough units/arm.

Nyarango 2006

Reason for exclusion Application of mixed interventions (IRS, ITN, larviciding and malaria case management).

Onori 1975

Reason for exclusion Not enough units/arm; not enough data for pre and post-intervention assessment.

Over 2003

Reason for exclusion Non-expermimental approach to analyse the impact of malaria intervention.

Over 2004

Reason for exclusion Non-expermimental approach to analyse the impact of malaria intervention.

Pampana 1950

Reason for exclusion Review, not enough data to analyse.

Pardo 2006

Reason for exclusion Not enough data for pre-and post-intervention assessment.

Pattanayak 1980

Reason for exclusion Not enough units/arm.

Payne 1976

Reason for exclusion Non enough units/arm.

Pletsch 1954

Reason for exclusion Not enough units/arm.

Protopopoff 2008

Reason for exclusion Control site not comparable.

Pujara 1983

Reason for exclusion Inappropriate choice of control site and not enough units/arm.

Rachou 1966

Reason for exclusion Not enough units/arm.

Rafi 1954

Reason for exclusion Not enough data for pre-intervention assessment.

Rajendram 1951

Reason for exclusion Insecticide (BHC) not recommended by WHO.

Rajendram 1951a

Reason for exclusion Insecticide (BHC) not recommended by WHO.

Rakotomanana 2001

Reason for exclusion Non-eligible outcomes measured.

Reisen 1993

Reason for exclusion Not enough units/arm.

Rodriguez 1994

Reason for exclusion Non-eligible outcomes measured.

Romi 2002

Reason for exclusion Not enough data for pre- and post- intervention assessment.

Russel 1939

Reason for exclusion Not enough units/arm.

Russel 1942

Reason for exclusion Not enough units/arm.

Sahondra 2001
Reason for exclusion ITS with mixed interventions (IRS and drug distribution).
Sahu 1993
Reason for exclusion Not enough units/arm (only 1 control village).
Sahu 1995
Reason for exclusion Non-eligible outcomes measured.
Saliternik 1977
Reason for exclusion Review, not enough data to analyse.
Sastry 1961
Reason for exclusion Review, not enough data to analyse.
Sexton 1994
Reason for exclusion Not enough data, conference abstract only.
Sharma 1982
Reason for exclusion DDT spraying in area with DDT resistant A. culicifacies.
Sharma 1985
Reason for exclusion Not enough units/arm.
Sharma 1986
Reason for exclusion Inappropriate choice of control site.
Sharma 1996
Reason for exclusion Not enough units/arm; spray coverage too low for malathion spraying.
Sharma 2005
Reason for exclusion Inappropriate choice of control site (DDT-spraying).
Sharp 2002
Reason for exclusion Not enough data for post-intervention assessment.
Singh 2006
Reason for exclusion ITS with mixed interventions (IRS, early detection and treatment, larvivorous fishes).
Taylor 1986
Reason for exclusion Not enough units/arm.
Tewari 1990
Reason for exclusion ITS with mixed interventions (IRS, space-spraying and anti-larval measures).
Trapido 1946
Reason for exclusion Not enough units/arm.
van Thiel 1951
Reason for exclusion Not enough units/arm.
van Wyk 2002
Reason for exclusion Not enough data, conference abstract only.
Verdrager 1975
Reason for exclusion Not enough units/arm.
Viswanathan 1947
Reason for exclusion Inappropriate choice of control site.
Viswanathan 1950
Reason for exclusion Mix of dosages used for IRS (two of them not in line with WHO recommendations).
Wattal 1978
Reason for exclusion Dosage of insecticide application not in line with WHO recommendations
WHO 2007
Reason for exclusion ITS with mixed interventions (IRS and anti-larval measures).

Wilson 1954

Reason for exclusion Not enough units/arm.

Wu 1984

Reason for exclusion Not enough units/arm.

Wu 1993

Reason for exclusion Inappropriate choice of control site.

Xu 1998

Reason for exclusion Not enough units/arm.

Xu 2002

Reason for exclusion Inappropriate choice of control site.

Zaphiropoulos 1959

Reason for exclusion Not enough data for pre-intervention assessment.

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6 Discussion

This discussion is divided into three parts. Firstly, the methodological approaches will be reviewed and an overview over the projects will be given. This will be followed by the key contributions made by this work to malaria control. Finally the prospects for future research will be presented.

6.1 Methodological issues and overview

6.1.1 Investigation of the malaria situation with the Higaturu Oil Palms plantations

The aim of our study within the plantations of the Higaturu Oil Palms (HOP) company was to provide an overview of the malaria epidemiology on the south-east coast of PNG, and to quantify the benefits of malaria control to the company. We used two different observational approaches in order to achieve this goal, namely by collecting routine health statistics data from the company aid posts and by conducting a cross-sectional survey within the study area.

To the best of our knowledge, this is the first work describing the impact of malaria on an agro-industrial operation in PNG. In addition, we provide for the first time a snapshot on the malariological situation around Popondetta.

Malaria was shown to be a major problem within the Higaturu Oil Palm plantations, with one third of the people (33.5%) having a positive blood slide. This is similar to surveys done in other non agro-industrial sites in PNG: a prevalence rate of 60% was found in the Wosera (Genton *et al.* 1995) and 35% to 43% surrounding Madang (Cattani *et al.* 1986b). Children and adults were both affected, which suggests there is little acquired immunity. Nevertheless, the prevalence was highest in the age group 5-9 years (40.3%), which is characteristic for a highly endemic area in PNG (Cattani *et al.* 1986a; Genton *et al.* 1995; Muller *et al.* 2003). *P. falciparum* was found to be the dominant species, followed by *P. vivax,* which is generally observed throughout PNG (Cattani *et al.* 1986b; Schuurkamp 1992; Genton *et al.* 1995; Mueller *et al.* 2003).

Since the HOP health staff routinely collects health data on all patients, it was operationally easy to compile the incidence rates of malaria patients. For HOP employees and their dependants the examination and treatment is provided for free at the company aid posts. The clinics are close by and easily reachable by foot. Once a month, the health staff holds health awareness sessions about TB, Malaria and HIV for all residents in every HOP village.

We therefore expected that almost everybody feeling sick with malaria symptoms would visit one of the clinics. This was confirmed with 99.5% of the participants having reported to have visited a health centre if feeling sick with 'malaria' within the last two weeks. This is substantially higher compared to other non-industrial PNG settings which experience attendance rates of 55% to 87% (Mueller *et al.* 2006; Mueller *et al.* 2007). However, care has to be taken when relying on routinely collected health data to monitor diseases, as the data quality is often poor (de Savigny & Binka 2004; Chilundo *et al.* 2004). Before the start of our survey, malaria was diagnosed by symptoms only. Symptoms of malaria are non-specific and many other diseases can present with the same clinical picture. Thus, a purely symptomatic diagnosis of malaria can lead to a vast over-diagnosis of malaria cases (Font *et al.* 2001; Wang *et al.* 2006). We therefore introduced rapid diagnostic tests (RDTs) for malaria in all aid posts. RDTs have been shown to perform well in routine practice (McMorrow *et al.* 2008; Hopkins *et al.* 2008) and they are now recommended for wider use (World Health Organization 2006a). In our study, the RDT sensitivity compared to reference microscopy was found to be 89%, which is excellent. As one drawback, the HRP2 may persist for several weeks after the parasites have been eradicated, posing a risk of obtaining false-positive results (Tjitra *et al.* 2001; Kyabayinze *et al.* 2008).

In areas with a high level of endemicity, it is rather likely that people have still antigens in their blood due to frequent malaria infections. Within the study area, a third of the population was found to be infected in a cross-sectional assessment, which speaks for a high level of endemicity. If many individuals resort to self-treatment there is a real possibility of false-positive results with RDTs. Nevertheless, after the introduction of RDTs, the number of malaria cases dropped significantly, highlighting that over-diagnosis did indeed take place on the basis of clinical suspicion alone. The feedback from the HOP health staff was also very positive. On the other hand, they also reported of patients pushing for a malaria treatment despite a negative RDT result, which is also found in other parts of the world (Williams *et al.* 2008).

Microscopy is still the most commonly used method for the diagnosis of malaria parasites. Reading blood slides needs a lot of expertise and the quality can be poor in areas where training, equipment and reagents are substandard (Perkins & Bell 2008). The slides of this survey were read by experienced microscopists from the Institute of Medical Research (IMR) in PNG. The slides were read twice by different microscopists to check for any discrepancies. Overall, the consistency of the two datasets was only barely sufficient, with an agreement of 78% (Kappa index of 0.43). Over 65% of the discrepancies were found in slides with very low

densities. This showed clearly the difficulty of reading accurately blood slides, even for experienced microscopists.

To get an idea of risk factors associated with malaria infections, we applied a questionnaire and combined the answers with the results of the blood slides. Care was taken when interpreting the results of such cross-sectional studies since exposure and outcome were measured at the same time, which makes it difficult to assess causality. Employees and dependants had the same risk of getting infected with malaria. At times, people prefer sleeping outside on the veranda or on a shelter. Surprisingly, there was no difference seen between these people and people sleeping inside the house. Since houses are not mosquito proofed, and since *A. punctulatus* is endo – but also exophilic (Charlwood *et al.* 1986) it is likely that the mosquitoes enter the houses and bite the inside sleeping inhabitants.

The company sells mosquito nets for half their commercial price, resulting in 70% of the people sleeping under a bednet. This is a higher coverage compared to findings of other studies done in the lowlands of PNG with 44% (Genton *et al.* 1995) and 55% (Genton *et al.* 1994), or studies in the highlands with 25% (Mueller *et al.* 2006). Nevertheless, Cattani *et al.* (1986b) reported a higher usage of 82% around Madang. ITNs are well known to reduce morbidity in different transmission settings (Lengeler 2004). In the Wosera area with a similar prevalence level as in our study area, bednet use was shown to reduce the risk of getting malaria with a protective efficacy of 37% (Genton *et al.* 1994). We therefore expected people sleeping under a bed net to have a reduced risk of getting malaria. While this was true on village level, no difference was seen on individual level. This is likely to be the result of the homogeneously high level of net use within our study area.

So far only few studies assessed the impact of malaria in a commercial company (Hedman *et al.* 1979; Yadav *et al.* 1991; Some 1992; Utzinger *et al.* 2002; Mills *et al.* 2008). At HOP, malaria impaired the company's effectiveness in different ways. As direct impact, they lost their workforce on average for 1.8 days per malaria episode. This is consistent with a study in Kenya, where an average malaria episode of an employee resulted in 2.4 man-days lost (Some 1992). Yadav *et al.* (1991) reported a much higher number of man-days lost per episode, ranging from 3.9 days up to 7.6 days. However, this study was conducted in a mining company and malaria was mostly affecting labourers of a low socio-economic group, which were suspected to have a lower health status and not taking treatment in a timely manner (Yadav *et al.* 1991). In a low transmission setting the lack of immunity is also likely to lead to a more severe disease. These factors might have prolonged the duration of the malaria episodes. Besides lost working days, *P. falciparum* infections were associated with anaemia, as found in the Wosera area by Genton *et al.* (1995b). Anaemia is known to reduce

physical strength and this is clearly reducing the HOP business effectiveness, since over two - thirds of the employees are harvesting fruits, which is a physically strenuous work. Treatment costs are also directly reducing the company's turn-over. More data on the impact of malaria on the company would have been highly interesting. However, shortly after the start of the project, the company was sold and the new owners reoriented their priorities, leading to a termination of the project.

Generalizing the results of this work to the rest of the PNG population is difficult, since an oil palm plantation presents a special setting. Only employees and their dependants are allowed to live within the plantation, which resulted in 40% of the participants being aged between 20 and 39, which is clearly not representative for the general population. In comparison, 40% of the population in PNG is under the age of 15 years (Hanson *et al.* 2001). The study participants also live in better houses than traditionally built houses in PNG, which reduces their risk of getting a malaria infection (Lindsay *et al.* 2002; Ye *et al.* 2006). In addition, all of the study participants have easy and free access to health care. Furthermore, an oil palm plantation represents an artificial environment with its own ecosystem, which is likely to have a different transmission rate than a natural habitat. *Anopheles punctulatus* - a major vector in PNG (Benet *et al.* 2004) - was found to be the main vector in the area (RD Cooper *et al.*, in press).

Malaria epidemiology is complex and it is influenced by many factors, such as malaria seasonality and transmission intensity, vector behaviour, resistance to drugs or insecticides, and the behaviour of the people. Depending on the availability of resources and feasibility, different control measures need to be applied. This highlights the importance of good knowledge of the local epidemiology in order to be able to apply the most feasible and effective malaria control measures.

Businesses have good prerequisites for a successful malaria control. They have a broad knowledge in managing people as well as devising operations and they have the resources for financing programmes (Global Health Initiative 2006). As a company is a rather small setting, they have the further advantage of operational feasibility and better access to the people.

6.1.2 Cochrane review on indoor residual spraying for preventing malaria

In 2006, the Director of the WHO's Global Malaria Programme declared his support for indoor residual spraying and especially the use of DDT for vector control in regions where malaria is a major health problem (Kochi 2006; World Health Organization 2006b). This

brought IRS back on the scene for malaria control and renewed interest has risen in evaluating IRS and to compare it to ITNs. While there is no doubt that IRS is effective in reducing malaria, this effect has never been quantified. In order to synthesize all information on IRS in an optimal way we produced a Cochrane systematic review. Cochrane reviews use a most rigorous methodological approach in order to minimise flaws and maintain a high standard of evidence (Olsen *et al.* 2001; Delaney *et al.* 2007; Moher *et al.* 2007). Though we included also a well designed controlled before-and-after study and one interrupted time series, we emphasised data from RCTs.

Cluster-randomised controlled trials are commonly used for measuring the effects of intervention that require application at a high coverage level to be effective – which is the case with IRS (Hayes *et al.* 2000; Donner & Klar 2002). Therefore, it was not surprising to find that all the included RCTs were cluster-randomised. Members within the same group tend to be more similar than individuals across different groups and this has to be accounted for in the analysis (Hayes *et al.* 2000; Donner & Klar 2002; Kim *et al.* 2006). Since the first comprehensive text on the design and analysis of clustered RCTs appeared in 1998 only (Murray *et al.* 2004) and since many of our included RCTs were published between 1998 and 2001, only one study (Misra *et al.* 1999) took the ICC into account when analysing the data. For few outcomes we were able to adjust the data for clustering retrospectively, but this was not always possible.

Cochrane reviews of interventions are considered to be the gold standard for determining the effectiveness of interventions (www.cochrane.org). However, due to the strict requirements of the Cochrane collaboration, we had to exclude 126 out of 132 studies in our review and remained with only six studies. In this case, a Cochrane review was not the right tool to assess whether IRS works. When assessing interventions with trials done in a time where the methodological and statistical methods were not as advanced as today, the risk of loosing a lot of studies and hence information is high. If in addition heterogeneity between the trials is high, a meta-analysis is not possible. However, our review also made clearly the point that a quantification of the effect of IRS is not possible at present, and this provides a strong rationale for further high-quality RCTs.

The external validity of the results is limited. The effectiveness of IRS is dependent on several factors including for example the class of insecticide used, the vectors present, the types of walls which were sprayed, malaria endemicity etc. The combination of firstly having only few trials on IRS and secondly of not being able to account for the many factors

influencing the effectiveness of IRS makes it impossible to generalize the findings of the Cochrane review.

6.2 Contribution to the knowledge on malaria control

With the development of DDT in 1939 it was thought that malaria could be eradicated, and in the 1950s, the World Health Organisation lunched the global malaria eradication campaign. The campaign was initially very successful in countries such as Italy, Cyprus and Greece or the Soviet Union (Najera 2001). These first results of IRS were so convincing that no need was seen to conduct proper controlled trials, which is aptly stated by Professor A.E. Beljaev of the Martinowsky Institute in Moscow: "*When I started my postgraduate studies in tropical medicine in 1963, the heroic era of malaria eradication in USSR was more or less over. We were taught that IRS was instrumental in interrupting the malaria transmission. This seemed so crystal clear to us youngsters that nobody sought of challenging this truth and verifying old reports (that were not so old in that time). One may compare DDT to Penicillin that became available roughly at the same time. Its effect in saving lives was so evident that nobody insisted on controlled clinical trial with placebo etc*.".

Malaria eradication was unfortunately never achieved and in 1969 the malaria eradication strategy using IRS and chemoprophylaxis was officially stopped (Najera 2001). This timing is also reflected within our Cochrane review: about a third (28%, n= 32) of the studies were carried out between 1950 and 70s. All of the old trials had to be excluded due to their inadequate quality. With the exception of the Garki project (Molineaux 1980), all the included trials were done between 1998 and 2007.

This situation contrasts with the high-quality effort to generate evidence on ITNs (Lengeler 2004). Overall, 22 randomised controlled trials met the inclusion criteria for the Cochrane review on ITNs (compared to four within the Cochrane review on IRS). With the exception of three studies, the ITN trials were all conducted between 1992 and 2005 (Lengeler 2004). A new quality leap in the generation of public health evidence was taken in recent years with the foundation of the intermittent preventive treatment in infants (IPTi) consortium in 2003 (Schellenberg *et al.* 2006). In total 15 institutions around the world, including WHO and UNICEF, collaborated and coordinated approaches ranging from drug safety, to evaluating statistics, to generate evidence for policy recommendations in a timely and coordinated manner (Schellenberg *et al.* 2006).

The paradox of our review is that while there is not the least doubt that IRS works well in most settings (in the presence of susceptible mosquitoes), we have so far failed to quantify that effect in a way similar to ITNs. This, unfortunately, impedes rational decision making for policy makers and makes it impossible to make an informed choice between IRS and ITNs. Furthermore without knowing the effectiveness of IRS, evaluating its cost-effectiveness is hampered and often has either to rely on modelling-derived effectiveness data, or on assuming an effect similar to ITNs (Yukich *et al* 2008).

Within our Cochrane review, only one RCT was conducted in a stable endemicity area, while three were done unstable malaria settings. Geographically, the RCTs were distributed in Asia (Pakistan and India) and in Africa (Tanzania and South Africa). No trials were available from South America. The few high-quality trials furthermore included different participants within the studies. While two RCTs included all age groups, two RCTs only included specific groups aged 1 to 15 years. A further problem was the previous history of malaria control within three trials, which obviously changed the entomological baseline situation and certainly the likely impact of vector control interventions. In addition, different outcomes were measured, but no sound data on the impact of mortality was available. As a result of this we clearly failed to achieve the stated goal of the review, to provide a clear quantification of the health effects of IRS and to compare it to ITNs.

Interestingly, WHO suddenly changed its recommendation for IRS in 2006. With no additional evidence, it now recommends IRS for all types of malaria transmission settings, from unstable to stable-endemic transmission (World Health Organization 2006b). Also, the recent Global Malaria Action Plan (GMAP) recommends the application of either IRS or LLINs in high transmission settings (Roll Back Malaria 2008). Finally, the World Malaria Report 2008 also mentions that IRS and ITNs can be deployed together to further enhance their impact (WHO & UNICEF 2008). All these new recommendations are based on little evidence.

In addition to the one RCT from Tanzania (Curtis *et al.* 1998) which is included in our review, one study in Kisumu, Kenya was conducted but with only one unit per arm (Payne *et al.* 1976). They evaluated IRS using fenitrothion and found an impressive impact on the crude mortality as well as the malaria transmission rate with a reduction of 43% and 96%, respectively (Payne *et al.* 1976). The trial in Pare-Taveta, Tanzania on the other hand was using Dieldrin as insecticide – an insecticide no longer recommended by WHO due to its toxicity (Najera & Zaim 2003). Nevertheless comparing the mortality rate before the intervention and in the third year after the intervention, a dramatic fall in the infant mortality

rate was seen, from 165 per 1000 to 132 per 1000, as well as in the crude mortality rate from 24 per 1000 to 16 per 1000. Infant parasite rates fell from 80% to 90% in 1955 to approximately 7% to 10% in 1959 (Bradley 1991).

Recently, nationwide programme evaluations from Bioko Island, Mozambique (under the Lubombo Spatial Development Initiative (LSDI)) and the Democratic Republic of São Tomé and Príncipe (DRSTP) have been reported. On Bioko Island, the prevalence rate was reduced by 15% within one year with IRS using Deltamethrin and case management. A protective efficacy of 47% was reported and children less than 15 years have benefited from the intervention. However, prevalence was only reduced to a level of 31% (Kleinschmidt *et al.* 2006). A better result was found in Mozambique by Sharp *et al.* (2007) with a reduction in prevalence of 43% within one year of IRS using bendiocarb. However, after interruption of IRS, the prevalence immediately rebounded to 59% and then steadily decreased to 23% after resumption of IRS (Sharp *et al.* 2007). In DRSTP, IRS with alphacypermethrin was initiated in 2004. Within two years, malaria prevalence was nearly reduced to zero (0.7%), lowering the prevalence by 19.4% (Tseng *et al.* 2008). The deaths attributed to malaria were reduced from about 65% to 37% (Teklehaimanot *et al.* 2009). In 2005, in addition to IRS, free LLINs were distributed all over the island and ACTs were introduced. As a result of the combined interventions, mortality was reduced to 3 malaria-related deaths in 2007 compared to 400 malaria-related deaths before 2005 (Teklehaimanot *et al.* 2009). In Zanzibar in 2007, with the combination of LLINs and IRS using pyrethroids, incidence cases at health facilities for children less than two years of age were reduced from about 22% to approximately 0.5% (PMI 2009).

The results of DRSTP and Mozambique were undoubtedly impressive. However, DRSTP is relatively small with only 150,000 inhabitants. This clearly simplifies malaria control compared to other countries on the African mainland such as e.g. Kenya with more than 22 million people at risk of malaria (World Health Organization for Africa 2007). Furthermore, the risk of vector immigration will be very limited on the islands. Only one species of vector is present on DRSTP (Teklehaimanot *et al.* 2009), which is also simplifying vector control. Mozambique is an area with low transmission intensity (Thompson *et al.* 1997) and has a rather good infrastructure. Therefore, these findings are clearly not representative for other endemic settings. The results of Bioko Island, with a high transmission intensity (Kleinschmidt *et al.* 2006) on the other hand were not so impressive, as they were only able to reduce the prevalence to 31%, despite the advantage of being an island.

However, the effectiveness of IRS alone should not be the only deciding factor. The public health usefulness of IRS is influenced by many other factors as well, such as the operational demands of IRS (which require a well structured system in place (Lengeler & Sharp 2003)), vector behaviour and resistance status, acceptance by the population, the type of housing and the nature of the walls to be sprayed, Finally, there is also an element of cost which needs to be taken into consideration (Yukich *et al.* 2008). Both interventions were found to be highly cost-effective but long-lasting insecticidal nets were still 2 to 3 times cheaper per person protected than IRS, especially if ITN coverage can be achieved with demographic targeting (Yukich *et al.* 2008).

There is nowadays broad consensus, that there is no global solution for malaria control as it should always be adapted to the local circumstances: “The first axiom of malariology, that lessons learnt in one part of the world may not be applied to other parts of the world, without local verification, is as true as ever; it is unfortunately as often neglected as ever” (D. Bagster) (Cambridge University Press 2009). This recommendation is particularly true for a country such as PNG, with several important vectors and malaria levels ranging from no malaria at all to high endemicity levels. In an oil palm plantation such as HOP, different control approaches are feasible and required compared to other parts of PNG.

6.3 Prospects for future research

More RCTs assessing the impact of IRS are urgently required. From a methodological point of view, the RCTs would ideally have three arms: one with IRS only, a second arm with ITNs, and a third arm in which IRS as well as ITN are implemented. In all arms, ITNs and IRS should be implemented at high coverage. Clearly, a control group not receiving any intervention would not be ethical since vector control was evidently shown to prevent malaria mortality and morbidity. An arm combining both interventions would be highly desirable because the recent goal of malaria elimination and ultimately eradication calls for a much higher level of transmission control (Roll Back Malaria 2008). This should be seen as a great priority at present. The trials should be conducted in a range of settings ranging from low levels of transmission to holoendemic levels. In countries with a national policy of using ITNs for malaria control, it will be difficult to find an area in which IRS alone can be implemented. In such settings the trials are likely to have only two arms (ITNs and the combination ITNs plus IRS). Hence these trials should ideally be carried out in countries in which malaria control policy entails IRS only. The trials should aim to measure different outcomes, ranging from malaria morbidity including anaemia to all-cause mortality.

Besides quantifying the effectiveness of existing insecticides, there is also an urgent need for new insecticides for vector control. Insecticide resistance is emerging, reducing the effectiveness of vector control (N'Guessan *et al.* 2007). The most recent insecticide made available for vector use is etofenprox, which was commercialized more than 20 years ago (in 1986). Up to now, no insecticides have been specifically produced for public health purposes. All were produced by large agro-chemical companies for an agricultural purpose (Tren *et al.* 2008). However, insecticides for public health use have different demands in comparison to the agricultural requirements: whereas public health insecticides should have a residual action and a broad spectrum of chemical activity, agricultural insecticides should be short acting and encompass a narrow activity spectrum (Tren *et al.* 2008). Unfortunately, the public health market for insecticides comprises only about 1.3% of the total pesticide market (Tren *et al.* 2008), and hence does not represent a big attraction for profit-making companies. The situation seems to be similar to the one for malaria drugs ten years ago. This could be impressively changed with the foundation of Medicine for Malaria Venture (MMV) (Medicine for Malaria Venture 2009), a not-for-profit public-private partnership (PPP) and such a collaboration of the public and private sectors should clearly also be considered for the research and development of new insecticides.

Conclusions

- Malaria poses a serious health problem for the Higaturu Oil Palms Company, but good conditions for control exist, provided the company management decides to act.

- Good quality evidence on the impact of IRS is scarce and the current evidence is insufficient to quantify properly the health effects of IRS even in the most general manner; specific recommendations for particular epidemiological situation are even more difficult to make. However, this should not imply that IRS lacks impact since the latter is amply demonstrated by historical evaluations.

- Available good quality evidence confirms that IRS with pyrethroids works in reducing malaria in unstable malaria settings but there is only very limited quality evidence on the effectiveness of IRS to reduce ill-health in stable malaria settings.

- There is no evidence of the effectiveness of IRS in reducing mortality, neither in stable nor unstable malaria settings.

- In absence of a robust quantification of the health effects of IRS, a meaningful comparison between IRS and ITNs is not possible.

- There is only very limited evidence to assess the effect of other determinants of impact, such as the insecticide class used for IRS, the type of transmission, the dominant vector species, and socio-cultural determinants.

- There is an urgent need for more RCTs comparing IRS and ITNs in a number of settings with different epidemiological and socio-cultural characteristics; if possible, child mortality should be measured as primary outcome. Ideally, such RCTs should have a third arm in which a combination of high coverage IRS with high coverage ITNs is implemented.

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