Journal of the American Mosquito Control Association, 22(3):501–506, 2006

INSECTICIDE-TREATED BED-NETS FOR MALARIA MOSQUITO CONTROL

C. F. CURTIS,[1] C. A. MAXWELL,[1,2] S. M. MAGESA,[2] R. T. RWEGOSHORA[2] AND T. J. WILKES[1]

ABSTRACT. Pyrethroid-treated bed-nets act against late-night biting mosquitoes, like traps baited by the body odor of the occupant. The personal protective effect of treated nets is considerable, even if they are torn. However, some biting of the occupants does occur, as shown by matching microsatellite alleles in mosquito blood meals to those of net occupants. When whole communities were provided with treated nets, ovarian age grading showed that mosquito survival was reduced, and so was the number of sporozoite-positive mosquitoes in malarious communities. Thus, a high percentage of coverage of all members of malaria-endemic communities is considered to be the most effective way of providing protection for highly malaria-vulnerable children and pregnant women. Teams distributing nets or retreating them free of charge show high productivity, and we consider this the most cost-effective way to proceed. There is evidence for reduced anti-malaria antibody levels in children in communities where treated nets have long been used. However, overall benefits in reduced anemia and mortality are sustained. A high frequency of the *kdr* resistance gene has not prevented pyrethroid-treated nets from functioning, but it is important to develop alternative fabric treatments in case stronger forms of resistance emerge.

KEY WORDS Bed-nets, insecticides, mosquito control, malaria

PERSONAL PROTECTION DUE TO PYRETHROID-TREATED NETS

The biting by most of the world's malaria vectors peaks in the middle of the night (Pates and Curtis 2005). It has been suggested that the biting habits of the dangerous older mosquitoes, which have survived long enough for *Plasmodium* sporozoites to have reached maturity, may not be the same as habits of the rest of the population. However, Maxwell et al. (1998) could find no significant difference with Tanzanian *Anopheles gambiae* s.s. and *An. funestus*, and their data indicate that 30 of 34 (88%) of sporozoite-positive bites occurred between 2200 and 0500 h. Ross (1910) perceived that biting of malaria vectors in the middle of the night should make bed-nets an effective protection against malaria but, if nets are torn, they provide no protection (Lines et al. 1987, Curtis et al. 1996, Mwangi et al. 2003). It was initially for this reason that the idea of adding a chemical barrier to the imperfect physical barrier was conceived, by treating nets with a pyrethroid that is safe for close contact with humans.

Treating a family's nets needs only about 1/6 as much insecticide as indoor residual spraying of their house (Curtis et al. 1998). In Tanzanian villages, nets are usually washed about 3–5 times a year, and bioassays confirm that retreatment once a year is sufficient to ensure continuous high insecticidal activity (Maxwell et al. 2003). In Vietnam, the nets of 10 million people are retreated annually by government teams (Tran Duc Hinh, unpublished data; reviewed by Curtis et al. 2004). There are now manufactured nets on the market in which the insecticide is incorporated in the netting fiber or attached to it with a resin. These deposits are more wash-resistant than those on conventionally dipped nets (Tami et al. 2004, Graham et al. 2005), and it is claimed that they remain insecticidal for the physical "life" of the net (approx. 3–8 years).

Counting of bloodfed mosquitoes resting in houses, and those driven out into exit traps on windows by the excitorepellent effect of pyrethroids, shows that treated nets greatly reduce biting, even if they are torn. Thus, a net provides considerable personal protection to the sleeper inside it if the net is intact, pyrethroid treated, or both (Curtis et al. 1996, Maxwell et al. 2003). It seemed possible that the few bloodfed mosquitoes found in rooms (and traps on windows of the rooms) with treated nets might have fed outside and entered already fed. However, Soremekun et al. (2004) matched the microsatellites in mosquito blood meals with those in the blood of people who slept under treated nets in the rooms where the mosquitoes were caught. The matches and mismatches were unequivocal; only 15% of the few bloodfed *An. gambiae* and *An. funestus* could be explained by entry of already fed mosquitoes, and the remainder had fed on the people who had slept under the nets. It is not known whether these feeds were by biting through the treated nets (despite their excitorepellent effect) or by waiting until the net occupant left the net during the night. However, the proof that these feeds occur allows us to conclude that although treated nets give good personal protection of the net occupant, the protection is not perfect (Table 1, columns 2 and 3).

[1] London School of Hygiene & Tropical Medicine, London WC1E 7HT United Kingdom.

[2] Ubwari Research Centre, Muheza, Tanga, Tanzania.

Table 1. Entomological data showing the relative importance of the personal and community-wide effects of treated nets.

	Personal protection of treated nets in rooms (corrected for mosquitoes entering after feeding elsewhere) (Soremekun et al. 2004)		Effect of community-wide use of treated nets (Maxwell et al. 2003)	
Treated nets/no nets in rooms and villages	% bloodfed of mosquitoes in rooms and exit traps	Feeding rate with nets as % of that without nets	EIR based on sporozoite +ve mosquitoes outside nets	EIR with treated nets as % of that without nets
Annually retreated nets	22.6	30.9	37.5	25.2
No nets	73.0		148.6	

COMMUNITY-WIDE EFFECTS OF TREATED NETS

Many dead mosquitoes can be collected in the morning from the floor of huts in which treated nets have been used the previous night. To properly assess this mortality in huts in tropical villages, ant traps are needed to prevent access by scavenging ants. One also needs to take into account mortality of mosquitoes that exited during the night but had already picked up a lethal dose of insecticide (Curtis et al. 1996). Among wild *An. gambiae* and *An. funestus* that enter huts in search of blood meals, mortalities of 25–75% have generally been observed with various different pyrethroids on nets that had been freshly treated or washed 5 times (Curtis et al. 1996, Maxwell et al. 1999).

As pointed out by Macdonald (1957), when residual insecticides are applied indoors, endophagic mosquitoes take a risk every time that they enter a house in search of a blood meal, which they do every 2–3 days at tropical temperatures. Thus, where there is high percentage of coverage of the houses in a village, the mean age of the village mosquito population is expected to be reduced, and very few mosquitoes will survive the approximately 12 days required for sporozoite maturation. Polovodova (1949) developed a method for assessing mosquito age from dilatations on the ovarioles (Detinova 1962). T. J. Wilkes applied this method to *An. gambiae* in Tanzanian villages before and after provision of treated nets or DDT spraying for all houses. The mosquitoes were also dissected and examined for sporozoites or tested for their presence by enzyme-linked immunosorbent assay (Burkot et al. 1984). As shown in Table 2, before these interventions, mean mosquito age was longer in traditional villages with thatched roofed houses than in a more modern village with iron roofs that become very hot at mid-day. When insecticidal nets or house spraying were provided, the mean age of the mosquitoes declined. In conformity with the observations about mean age, the sporozoite rates were less in the iron-roofed village and after each of the insecticidal interventions (Magesa et al. 1991).

Using light traps in bedrooms, we have repeatedly assessed the biting of anopheline mosquitoes outside nets in villages provided with treated nets in all the rooms except the "sentinel" rooms in which the light traps were run. As would be expected, because of the considerable mortality in populations, the biting densities were reduced, and multiplying these densities by the sporozoite rates led to estimated reductions in the rates of sporozoite-infective biting per night (entomological inoculation rate [EIR]) to about 25% of those in villages without nets (Table 1, columns 4 and 5).

Table 2. Data of T. J. Wilkes for *An. gambiae* on ovarian age grade (maximum number of dilatations per ovariole in each individual, averaged over all individuals) and percentage of mosquitoes positive for sporozoites. Data shown for 2 traditional and 1 more modern Tanzanian village, before and after provision of permethrin-treated bed-nets or DDT indoor residual spraying. Sample sizes on which age grading data were based were 95–1,533 and for sporozoite rates were 201–2,181 (Magesa et al. 1991).

Village (type of roof on houses)	Preintervention			Postintervention	
	Mean age grade	% sporozoite +ve	Intervention	Mean age grade	% sporozoite +ve
Mng'aza (thatch)	1.255	6.1	Pyrethroid treated nets	0.792	2.3
Mindu (thatch)	1.229	7.9	DDT indoor spraying	0.400	2.5
Mlingano (iron)	0.764	4.9	Pyrethroid treated nets	0.518	0.6

Comparison of the personal protection and community-wide "bonus" effects of treated nets (Table 1, columns 2 and 3 versus 4 and 5) shows that each reduces risk to 25–30% of what it would be without nets in rooms or villages. Thus, the overall reduction is to about 25% × 30%, i.e., to about 7.5%, or a reduction of 92.5%, due to the combination of both forms of protection. There are no exact data on the reduction of EIR at different percentages of coverage of all the beds in a village with treated nets, but it is a reasonable assumption that the higher the percentage of coverage, the greater the mosquito mortality and thus the greater the reduction in EIR.

From a large Centers for Disease Control and Prevention (CDC)/Kenya Medical Research Institute trial in Western Kenya, Hawley et al. (2003) showed that impact of treated nets on child mortality and morbidity extended out from areas with the nets to neighboring areas without them, because of the impact of the insecticidal nets on the EIR of the local vector populations (Gimnig et al. 2003).

In highly malaria endemic areas, such as lowland tropical Africa, children and pregnant women are the most vulnerable to malaria because they have lower levels of antimalaria immunity than the rest of the population. Schemes are being developed or in existence for targeting free or subsidized treated nets to the vulnerable children and pregnant women. The Abuja Declaration by African Heads of State set 60% provision of treated nets for children and pregnant women as a target. Achieving this goal should be given every encouragement, but the data in Table 1 strongly suggest that considerably better protection for the vulnerable categories would be achieved if insecticidal nets were provided for everybody in highly malaria-endemic rural communities, so that many mosquitoes are killed on the nets of the not-so-vulnerable before these mosquitoes can attack the very vulnerable.

COST OF UNIVERSAL PROVISION

The productivity is very high of teams who visit a village (by prior arrangement with the inhabitants) to check the numbers and approximate sizes of each bed or sleeping mat in each house and provide treated nets for each, against householders' signatures acknowledging the donation of the nets for that family's protection. That the teams are not required to try to collect money or to judge who deserves a free or subsidized net greatly adds to the speed at which the work can be done. Some feel that nets provided free will not be valued and will be sold or misused. However, that is not our experience in work in Tanzania over the past 18 years, and checks by Guyatt et al. (2002) showed that nets targeted by a UNICEF project for pregnant women in Kenya almost all reached them and were used by them. Similar high productivity and coverage have been found with net retreatment provided free of charge on an appointed day each year by a visiting supervisor and a team of village health workers. This system works far better than expecting very poor people to take the initiative and go to a shop to buy sachets of insecticide.

Table 3. Estimates of annual costs for free provision to a million people—700,000 nets to be replaced every 4th year and retreated every year (labor costs based on observed high productivity of Kenyan and Tanzanian teams not encumbered by having to collect money).

175,000 nets/year @ $2.50	$437,500
700,000 doses of alphacypermethrin	$280,000
2 teams of 5 net distributors + administrators: wages + transport:	$171,000
11 retreatment supervisors + 33 village workers: wages + transport:	$135,000
Total/million people/year:	$1,023,500[1]

[1] Therefore total costs estimated as ca. $358 million/year for scaling up to provide for 350 million p eople in rural lowland tropical Africa (excluding towns where many people buy nets against *Culex* nuisance, not malaria mosquitoes). Alternatively, if Olyset nets @ $5.50 last 8 years and do not require retreatment, total cost would be ca. $198 million/year.

Table 3 shows estimates of materials and labor for providing replacement nets every 4 years for a million people and for retreatment of these nets every year. The estimates assume nets at a bulk purchase price of $2.50 each, and labor and transport are as observed in free distribution operations in Kenya and Tanzania. The costs per million people may be multiplied by the total rural population of tropical Africa for a continent-wide estimate. This estimate assumes targeting at everyone in the highly malarious lowland, rural areas of Africa. There are already many nets in use in African towns because people have more access to cash there than do subsistence farmers and because the nuisance from *Culex* is generally worse in towns with poor sanitation than in rural areas, which have cleaner surface water and therefore far more *Anopheles* and malaria cases.

The costs shown in Table 3 may possibly be reduced if "long-lasting nets" live up to expectations and never require retreatment and are more physically robust than cheaper nets and thus require replacement less often. The costs listed in Table 3 may be compared with the money spent on controlling another blood-sucking insect—the cat flea, on the control of which Rust (2005) reported that $2.1 billion is spent annually in Europe and the USA.

Lower distribution costs than those shown in Table 3 have been reported by programs that couple free net distribution to child measles vaccination campaigns (Grabowsky et al. 2005a, 2005b). Such campaigns occur on a very large scale in Africa (Henao-Restrepo et al. 2003) and are very well attended by even the poorest people.

Table 4. Investigation of whether loss of immunity causes a "rebound" on the anemia in older children when treated nets are used for several years.

	% reduction in anemia		Antibody titers (in children >2 years old)	
Previous use of treated nets	<2 years old	2–5 years old	Anti-CSP[1]	Anti-VSA[2]
Newly introduced nets[3]	58.9*	58.5*	0.327	1,183
3–4 years previous use[4]	53.2*	30.1 ns[5]	0.161	964

[1] Circumsporozoite protein, difference significant at $P < 0.05$, Metzger et al. (1998).
[2] Variant surface antigen, difference significant at $P < 0.01$, Askjaer et al. (2001).
[3] Maxwell et al. (2003).
[4] Maxwell et al. (2002).
[5] ns, apparent difference not significant.
* difference significant at $P < 0.001$.

These campaigns provide an opportunity for a major and remarkably equitable scaling up of the coverage of children with treated nets throughout rural Africa. However, so far, these distributions have targeted children and not whole malaria-endemic populations, which, as suggested above, would give optimum protection of the vulnerable children.

Those who support the social marketing of nets would accept that taxpayers in developed countries are almost incomparably richer than African subsistence farmers. However, they take the view that, although there is considerable concern about malaria control expressed at present among donor governments, this concern probably will not last, and that it is better that the poorest people on earth get used to the idea that they must find the money that will be needed to protect their children from malaria. This is not our view nor that of the UN Millennium Project (2005) report, which unequivocally urges that a major requirement for an effective attack on highly endemic African malaria is organized free provision of insecticidal nets.

SUSTAINABILITY OF HEALTH BENEFITS FROM REDUCING MALARIA TRANSMISSION

There is much evidence from short-term trials that community-wide use of treated nets has major benefits in reducing malaria morbidity and mortality (Phillips-Howard et al. 2003a, 2003b; ter Kuile et al. 2003; Lengeler 2005). However, because of the importance of acquired immunity to malaria, it has been argued (with regard both to residual house spraying as envisaged in the 1950s and insecticide-treated nets in recent years) that major reductions in numbers of infective bites received by children may cause reduced buildup of antimalaria immunity. Hence, these reductions might fail to achieve an overall reduction of malaria morbidity and mortality, but only its postponement to later in childhood. If, at that somewhat later age, the children were more susceptible to cerebral malaria, the eventual effect of well-meaning attempts to reduce transmission could even be to make the burden of malaria worse (Trape and Rogier 1996).

Table 4 shows some data indicating reduced levels of two anti-malaria antibodies in children in villages where treated nets have been used for several years (Metzger et al. 1998, Askjaer et al. 2001, Kariuki et al. 2003). However, among these children, there was less anemia, which is the main route by which malaria kills in highly endemic areas. The continued reduction in anemia was highly significant in young children. In older children, there was not significantly less anemia in netted villages, but there was certainly not significantly more, i.e., no indication that the older children were "paying for" the benefit that they received when they were younger (Maxwell et al. 2002).

Similarly, 4- to 6-year follow-up of the large CDC-supported treated net project in Kenya showed that mortality of infants under 1 year continued to be reduced in the netted villages, and, at later ages, mortality was neither reduced nor increased (Lindblade et al. 2004, Eisele et al. 2005).

The best impact so far reported with a malaria vaccine (Alonso et al. 2004) was not as great as that due to treated nets. In view of the evidence in Table 4 that prolonged use of nets lowers antibody levels, one might consider a vaccine as a way of improving the long-term performance of treated nets by restoring antibodies to the high levels that are normal in areas highly endemic for malaria.

INSECTICIDE RESISTANCE

In our project area in Tanzania, we have tested for, but not found, pyrethroid resistance in *An. gambiae* and *An. funestus*. However, pyrethroid resistance does exist in the same area in *Culex quinquefasciatus*. Very low mortality of this species is observed in our experimental hut studies (Curtis et al. 1996), and, in contrast to anophelines, no effect of community-wide use of treated nets on *Culex* populations has been observed (Maxwell et al. 1999).

Village populations of the tropical bedbug *Cimex lectularis* were eradicated when treated nets were first introduced. However, after about 6 years of use of treated nets, villagers noticed the return of bedbugs. Susceptibility tests showed that in 5 of these villages there was detectable resistance, in contrast to 5 villages without nets (Myamba et al. 2002). It is important to find an alternative way of controlling these bedbugs, because the high level of acceptance of the use of treated nets and need to bring them for retreatment may have been partly because of the good effect of the nets on nuisance bedbugs.

In Côte d'Ivoire a high frequency of the *kdr* pyrethroid resistance gene has been found in *An. gambiae*, but high mortality of these mosquitoes has been observed in experimental huts with pyrethroid-treated nets (Darriet et al. 2000, Asidi et al. 2004), and there has been good control of malaria with these nets in villages (Henry et al. 2005). It is suggested that because this gene not only reduces susceptibility to being killed but also to excitorepellency, the mosquitoes continue to rest on treated nets until a lethal dose is absorbed. Other more powerful forms of pyrethroid resistance exist elsewhere in Africa (Hargreaves et al. 2000). Thus, it is important to develop alternative fabric treatments with little likelihood that cross-resistance to pyrethroids would occur. Entomopathogenic fungal spores, which kill adult *Anopheles* (Scholte et al. 2004), seem to be a possibility that is worth exploring.

REFERENCES CITED

Alonso P, Sacarlal J, Aponte JJ, Leach A, Macete E, Milman J, Mandomando I, Spiessens B, Guinovart C, Espasa M, Bassat Q, Aide P, Ofori-Aniyam O, Navia MM, Coracham M, Dubois MC, Demontie MA, Dubovsky F, Menendez C, Tornieporth N, Ballou WR, Thompson R, Cohen J. 2004. Efficacy of the RTS,S/AS02A vaccine against *Plasmodium falciparum* infection and disease in young African children: randomised controlled trial. *Lancet* 364: 1411–1420.

Asidi AN, N'Guessan RN, Hutchinson RA, Traoré-Lamizana M, Carnevale P, Curtis CF. 2004. Experimental hut comparisons of nets treated with carbamate or pyrethroid insecticides, washed or unwashed, against pyrethroid resistant mosquitoes. *Med Vet Entomol* 18:134–140.

Askjaer N, Maxwell C, Chambo W, Staalsoe T, Nielsen M, Hviid L, Curtis C, Theander T. 2001. Insecticide-treated bed nets reduce antibody levels and limit the repertoire of antibodies to *Plasmodium falciparum* variant surface antigens. *Clin Diagn Lab Immunol* 8:584–586.

Burkot TR, Williams JL, Schneider L. 1984. Identification of *Plasmodium falciparum* infected mosquitoes by a double antibody enzyme linked immunosorbent assay. *Am J Trop Med Hyg* 33:783–788.

Curtis CF, Jana-Kara B, Maxwell CA. 2004. Insecticide treated nets: impact on vector populations and relevance of initial intensity of transmission. *J Vector Borne Dis* 40:1–8.

Curtis CF, Maxwell CA, Maxwell CA, Finch RJ, Njunwa KJ. 1998. A comparison of use of a pyrethroid either for house spraying or for bednet treatment against malaria vectors. *Trop Med Int Health* 3:619–631.

Curtis CF, Myamba J, Wilkes TJ. 1996. Comparison of different insecticides and fabrics for anti-mosquito bednets and curtains. *Med Vet Entomol* 10:1–14.

Darriet F, N'Guessan R, Koffi A, Konan LY, Doannio IMC, Chandre F, Carnevale P. 2000. Impact de la résistance aux pyréthrinoides sur l'efficacité des moustiquaires impregnées dans la prevention du paludisme: résultats des essais en cases expérimentales avec déltaméthrine SC. *Bull Soc Pathol Exot* 95:131–134.

Detinova TS. 1962. *Age grouping methods in Diptera of medical importance.* WHO Monograph series No. 47. Geneva, Switzerland: World Health Organization.

Eisele TP, Lindblade KA, Wannemuehler KA, Gimnig JE, Odhiambo F, Hawley WA, ter Kuile FO, Philips-Howard PA, Rosen DA, Nahlen BL, Vulule JM, Slutsker L. 2005. Effect of sustained insecticide-treated bednet use on all-cause child mortality in an area of intense perennial malaria transmission in western Kenya. *Am J Trop Med Hyg* 70:149–156.

Gimnig JE, Vulule JM, Lo TQ, Kamau L, Kolczak MS, Phillips-Howard PA, Mathenge EM, ter Kuile FO, Nahlen BL, Hightower AW, Hawley WA. 2003. Impact of permethrin-treated bednets on entomologic indices in an area of intense year-round malaria transmission. *Am J Trop Med Hyg* 68(Suppl 4):16–22.

Grabowsky M, Farrell N, Hawley W, Chimumbwa J, Hoyer S, Wolkon A, Selaniko J. 2005a. Integrating insecticide-treated bednets into measles vaccination campaign achieves high, rapid and equitable coverage with direct and voucher-based methods. *Trop Med Int Health* 10:1151–1160.

Grabowsky M, Nobiya T, Ahun M, Donna R, Lengor M, Zimmerman D, Ladd H, Hoekstra E, Bello A, Baffoe-Wilmot A, Amofah G. 2005b. Distributing insecticide-treated bednets during measles vaccination: low-cost means of achieving high and equitable coverage. *Bull World Health Organ* 83:195–201.

Graham K, Kayedi MH, Maxwell C, Kaur H, Rehman H, Malima R, Curtis CF, Lines JD, Rowland MW. 2005. Multi-country field trials comparing wash-resistance of Permanet and conventional insecticide-treated nets against anopheline and culicine mosquitoes. *Med Vet Entomol* 19:72–83.

Guyatt HL, Gotink MH, Ochola S, Snow RW. 2002. Free bednets to pregnant women through antenatal clinics in Kenya: a cheap, simple and equitable approach to delivery. *Trop Med Int Health* 7: 409–420.

Hargreaves K, Koekemoer LL, Brooke BD, Hunt RH, Mthembu J, Coetzee M. 2000. *Anopheles funestus* resistant to pyrethroid insecticides in South Africa. *Med Vet Entomol* 14:181–189.

Hawley WA, Phillips-Howard PA, ter Kuile FO, Terlouw DJ, Vulule JM, Ombok M, Nahlen BL, Gimnig JE, Kariuki SK, Kolczak MS, Hightower AW. 2003. Community-wide effects of permethrin-treated bed nets on child mortality and morbidity in Western Kenya. *Am J Trop Med Hyg* 68(Suppl 4):121–127.

Henao-Restrepo AM, Strebel P, John Hoekstra E, Birmingham M, Bilous J. 2003. Experience in global measles control, 1990-2001. *J Infect Dis* 187(Suppl 1):S15–S21.

Henry M-C, Assi S-B, Rogier C, Dossou-Yovo J, Chandre F, Guillet P, Carnevale P. 2005. Protective efficacy of lambdacyhalothrin treated nets in *Anopheles gambiae* pyrethroid resistant areas of Côte d'Ivoire. *Am J Trop Med Hyg* 73:859–864.

Kariuki SK, Lal AA, Terlouw DJ, ter Kuile FO, Ong'echa JM, Phillips-Howard PA, Orago AAS, Kolczak MS, Hawley WA, Nahlen BL, Shi YP. 2003. Effects if permethrin-treated bed nets on immunity to malaria in Western Kenya II. Antibody responses in young children in an area of intense malaria transmission. *Am J Trop Med Hyg* 68(Suppl 4):108–114.

Lengeler C. 2005. Insecticide *treated bednets and curtains for malaria control: a Cochrane review*. The Cochrane Library, Issue 3. Oxford, United Kingdom: Oxford Update Software Ltd.

Lindblade KA, Eisele TP, Gimnig JE, Alaii JA, Odhiambo F, ter Kuile FO, Hawley WA, Wannemuehler KA, Philips-Howard PA, Rosen DA, Nahlen BL, Terlouw DJ, Adam K, Vulule JM, Slutsker L. 2004. Sustainability of reductions in malaria transmission and infant mortality in western Kenya with use of insecticide treated bednets. *J Am Med Assoc* 291:2571–2580.

Lines JD, Myamba J, Curtis CF. 1987. Experimental hut trials of permethrin-impregnated mosquito nets and eave curtains against malaria vectors in Tanzania. *Med Vet Entomol* 1:37–51.

Macdonald G. 1957. *The epidemiology and control of malaria*. London, United Kingdom: Oxford Univ. Press.

Magesa SM, Wilkes TJ, Mnzava AEP, Njunwa KJ, Myamba J, Kivuyo MDP, Hill N, Lines JD, Curtis CF. 1991. Trial of pyrethroid impregnated bednets in an area of Tanzania holoendemic for malaria. Part 2: effects on the malaria vector population. *Acta Trop* 49:97–108.

Maxwell CA, Wakibara J, Tho S, Curtis CF. 1998. Malaria-infective biting at different hours of the night. *Med Vet Entomol* 12:325–327.

Maxwell CA, Myamba J, Njunwa KJ, Greenwood BM, Curtis CF. 1999. Comparisons of bednets impregnated with different pyrethroids for their impact on mosquitoes and on re-infection with malaria after clearance of existing infections with chlorprogaunaildapsone. *Trans R Soc Trop Med Hyg* 93:4–11.

Maxwell CA, Msuya E, Sudi M, Njunwa KJ, Carneiro IA, Curtis CF. 2002. Effect on malaria morbidity of community-wide use in Tanzania of insecticide treated nets for 3-4 years. *Trop Med Int Health* 7:1003–1008.

Maxwell CA, Chambo W, Mwaimu M, Magogo F, Carneiro IA, Curtis CF. 2003. Variations in malaria transmission and morbidity with altitude in Tanzania and with introduction of alphacypermethrin treated nets. *Malar J* 2:28.

Metzger WG, Maxwell CA, Curtis CF. 1998. Anti-sporozoite immunity and impregnated bednets in Tanzanian villages. *Ann Trop Med Parasitol* 92: 727–729.

Mwangi TW, Ross A, Marsh K, Snow RW. 2003. The effects of untreated bednets on malaria infection and morbidity on the Kenyan coast. *Trans R Soc Trop Med Hyg* 97:369–372.

Myamba J, Maxwell CA, Asidi A, Curtis CF. 2002. Pyrethroid resistance in tropical bedbugs, *Cimex hemipterus*, associated with use of treated bednets. *Med Vet Entomol* 16:448–451.

Pates HV, Curtis CF. 2005. Mosquito behavior and vector control. *Annu Rev Entomol* 50:53–70.

Phillips-Howard PA, Nahlen BL, Kolcak MS, Hightower AW, ter Kuile FO, Alaii JA, Gimnig JE, Arudo J, Vulule JM, Odhacha A, Kachur AP, Schoute E, Rosen DH, Sexton JD, Oloo AJ, Hawley WA. 2003a. Efficacy of permethrin-treated bed nets in the prevention of mortality in young children in an area of high malaria transmission in Western Kenya. *Am J Trop Med Hyg* 68(Suppl 4):23–29.

Phillips-Howard PA, Nahlen BL, Wannemuehler KA, Kolczak MS, ter Kuile FO, Gimnig JE, Olson K, Alaii JA, Odhacha A, Vulule JM, Hawley WA. 2003b. Impact of permethrin-treated bed nets on the incidence of sick child visits to peripheral health facilities. *Am J Trop Med Hyg* 68(Suppl 4):38–43.

Polovodova VP. 1949. The determination of the physiological age of female *Anopheles* by the number of gonotrophic cycles completed. *Medit Parazitol Parazit Bolez* 18:352–355. [In Russian].

Ross R. 1910. *The prevention of malaria*. London, United Kingdom: Murray.

Rust MK. 2005. Advances in the control of *Ctenocephalides felis* (cat flea) on cats and dogs. *Trends Parasitol* 21:232–236.

Scholte E-J, Knols BGJ, Takken W. 2004. Autodissemination of the entomopathogenic fungus *Metarhizium anisopliae* amongst adults of the malaria vector *Anopheles gambiae s.s. Malar J* 3:45.

Soremekun S, Maxwell CA, Zuwakuo M, Chen C, Michael E, Curtis C. 2004. Measuring the efficacy of insecticide treated bednets: the use of DNA fingerprinting to increase the accuracy of personal protection estimates in Tanzania. *Trop Med Int Health* 9:664–672.

Tami A, Mubyazi G, Talbert A, Mshinda H, Duchon S, Lengeler C. 2004. Evaluation of Olyset insecticide-treated nets distributed seven years previously in Tanzania. *Malar J* 3:19.

ter Kuile FO, Terlouw DJ, Kariuki SK, Phillips-Howard PA, Mirel LB, Hawley WA, Friedman JF, Shi YP, Kolczak MS, Lal AA, Vulule JM, Nahlen BL. 2003. Impact of permethrin-treated bed nets on malaria, anemia and growth in an area of intense perennial malaria transmission. *Am J Trop Med Hyg* 68(Suppl 4):68–77.

Trape J-F, Rogier C. 1996. Combating malaria morbidity and mortality by reducing transmission. *Parasitol Today* 12:236–240.

UN Millennium Project. 2005. *Coming to grips with malaria in the new millennium*. United Nations Development Programme, London, United Kingdom: Earthscan.