Stochastic models for the consequences of *Plasmodium falciparum* infections in the human host: malaria morbidity, mortality and infectivity

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Prof. Dr. H.-P. Hauri
Dekan

".. if the consequences of alternative available strategies cannot be predicted to the point where we can effectively discriminate between them, there is no rational basis for any kind of choice."

N. Bailey, 1982

Summary

The consequences of *Plasmodium falciparum* infections for humans range from self-limiting asymptomatic parasitaemia to rapid death. Annually, *P. falciparum* malaria is estimated to cause 0.5 billion acute febrile episodes, 2-3 million severe episodes warranting hospital admission and one million deaths. This enormous burden demands effective control strategies. A number of different interventions are available, but policy-makers need a rational basis for discriminating between them.

The likely consequences of each intervention, or combination of interventions, must be considered. Trials can provide estimates of the impact of interventions on acute episodes over a short timespan. Predictions are required where field data are not available: over longer time periods, for severe outcomes, for many combinations of interventions, or for interventions which do not yet exist. A model intended for making quantitative predictions of the impact of interventions must relate transmission and infection to the key outcomes used by health-planners such as morbidity, mortality and cost-effectiveness. It must also allow for dynamic effects on transmission and acquired immunity, and incorporate the effect of the health system. Until recently, there was no such model. In the past, emphasis had lain with the transmission cycle. In addition, the most practical models would be individual-based with high computational demands.

In response to this need, a stochastic individual-based integrated model has been developed at the Swiss Tropical Institute. The core of the integrated model is a description of asexual parasite densities, providing a basis for the effects of acquired immunity on reducing densities and for the density-based consequences of infection. This thesis contributes those elements that consider the immediate consequences of human infection: morbidity, mortality and transmission to the vector. The integrated model is then applied to questions concerning a new intervention, intermittent preventive treatment in infants (IPTi).

A framework for morbidity and mortality is proposed. The probability of an acute febrile malaria episode is related to parasite densities via individual- and time-specific pyrogenic thresholds that respond dynamically to recent parasite load. Severe episodes result either from overwhelming parasitaemia, or from acute episodes in conjunction with a co-morbidity which acts to weaken the host. Both direct and indirect mortality were considered. Age-dependent case fatality rates estimated from field data were used to quantify the probability of direct mortality. Indirect deaths occur following an acute episode with subsequent co-morbidity after the parasites have cleared, or within the neonatal period as a consequence of maternal infection. Co-morbidity is assumed to be age-dependent. The model components are fitted to field data or to summaries of field data, and can simultaneously account for the observed age- and exposure-specific patterns of paediatric malaria and malaria-associated mortality.

The model component for infectivity relates asexual parasite densities to the probability of infecting a feeding mosquito, taking into account the delay resulting from the

timecourse of gametocytaemia and the need for both male and female gametocytes in the blood meal. This component is fitted to data from malariatherapy patients and can account for observed patterns of human infectivity. The integrated model is validated against published estimates of the contribution of different age groups to the infectious reservoir.

The integrated model, in conjunction with an added component for the action of sulphadoxine-pyrimethamine (SP) and site-specific inputs, reproduced the pattern of results of the IPTi trials reasonably well. The model was modified to represent different hypotheses for the mechanism of IPTi. These hypotheses concerned the duration of action of SP, the empirical timing of episodes caused by individual infections, potential benefits of avoiding episodes on immunity and the effect of sub-therapeutic levels of SP on parasite dynamics. None of the modified versions improved the fit between the model predictions and observed data, suggesting that known features of malaria epidemiology together with site-specific inputs can account for the pattern of trial results. Predictions using the integrated model suggest that IPTi using SP is effective over a wide range of transmission intensities at reducing morbidity and mortality in infants. The predicted cumulative benefits were proportionately greater for mortality and severe episodes than for acute episodes, due to the age-dependent co-morbidity functions in the model. IPTi was predicted to avert a greater number of episodes where IPTi coverage was higher, the health system treatment coverage lower, and for drugs which were more efficacious and had longer prophylactic periods. Additionally, IPTi was predicted to have little impact on transmisison intensity.

This is the first major attempt to model the dynamic effects of malaria transmsission, parasitological status, morbidity, mortality and cost-effectiveness using model components which were fitted to field data. The model can be extended to predict the dynamic effects of different interventions, and combinations of interventions. The ability to compare the likely impact of different interventions on the same platform will be a valuable resource for rational decisions about strategies to control the intolerable burden of malaria.

Zusammenfassung

Die Folgen einer Infektion mit *Plasmodium falciparum* beim Menschen reichen von einem asymptomatischen Verlauf der Krankheit bis zu einem schnellen Tod. Jedes Jahr, so schätzt man, versursacht *P. falciparum* Malaria ungefähr eine halbe Milliarde akuter Fieberepisoden, zwei bis drei Millionen schwere, eine Hospitalisierung nach sich ziehende Krankheitsfälle, sowie eine Million Todesfälle. Diese enorme Krankheitslast verlangt nach wirkungsvollen Interventionsstrategien. Wohl steht eine Anzahl verschiedener Bekämpfungsmethoden zur Verfügung, jedoch benötigen die politischen Entscheidungsträger eine rationale Grundlage, um Wirkungsvolle Strategien zu entwickeln.

Die zu erwartenden Folgen einer Intervention oder Kombination von Interventionen müssen genau in Betracht gezogen werden. Feldversuche sind in der Lage, die Auswirkungen von bestimmten Massnahmen auf die Anzahl akuter Krankheitsfälle über kurze Zeiträume abzuschätzen. Modellbasierte Voraussagen sind jedoch immer dort vonnöten, wo Daten aus dem Feld nicht erhältlich sind: Um Aussagen über längere Zeiträume, schwere Krankheitsverläufe, Kombinationen verschiedener Massnahmen, oder Massnahmen zu machen, welche sich erst in der Entwicklung befinden. Ein Modell zur quantitativen Vorhersage des Effektes von verschiedenen Interventionen muss eine Beziehung zwischen Übertragung und Infektion sowie denjenigen möglichen Folgen herstellen, welche für Gesundheitsplaner von Interesse sind. Dazu gehören Morbidität, Mortalität und Kosteneffizienz. Zudem muss es auch in der Lage sein, dynamische Rückkopplungseffekte auf Übertragung und Immunität miteinzubeziehen und sollte dabei auch den Einfluss der Gesundheitssysteme berücksichtigen. Bis vor kurzem gab es jedoch kein Modell, welches genannte Anforderungen erfüllt hätte.

Aus diesem Grund wurde am Schweizerischen Tropeninstitut ein individuen-basiertes, integriertes Modell entwickelt. Herzstück dieser Computersimulation ist eine mathematische Beschreibung der Parasitendichte im Blut, welche als Ausgangspunkt für die Wirkungen erworbener Immunität sowie für die klinischen Folgen einer Infektion dient. Diese Dissertation trägt jene Teile zu genanntem Modell bei, welche die unmittelbaren Folgen einer Infektion im Menschen betrachten: Morbidität, Mortalität, und Übertragung auf den Vektor. Das ganze Modell wird am Schluss angewandt auf eine neue Intervention, genannt „Intermittent Preventive Treatment in Infants“ (IPTi), bei welcher Säuglinge wiederholt präventiv behandelt werden.

Es wird ein Konzept zur Modellierung von Morbidität und Mortalität unterbreitet. Die Wahrscheinlichkeit einer akuten fiebrigen Malariaepisode hängt dabei über individuen- und zeitspezifische Fieber-Schwellenwerte von der Parasitendichte ab. Diese Schwellenwerte selber wiederum verändern sich dynamisch in Abhängigkeit von der Parasitendichte. Schwere Episoden entstehen entweder als Folge einer ungewöhnlich hohen Parasitendichte, oder durch Zusammentreffen einer akuten Episode mit einer Komorbidität, welche den Patienten zusätzlich schwächt. Sowohl

direkte wie auch indirekte Mortalität wurden in Betracht gezogen. Altersspezifische Todesraten aus Felddaten wurden benutzt, um die Wahrscheinlichkeiten direkter Mortalität zu beziffern. Indirekt verursachte Todesfälle treten im Modell entweder nach einer akuten Episode mit darauffolgender Komorbidität auf, oder innerhalb des neonatalen Zeitraumes als Folge einer Infektion der Mutter. Der altersabhängigen Verteilung von Komorbiditäten wird dabei Rechnung getragen. Die einzelnen Modellparameter wurden mit Hilfe von Felddaten geschätzt, sodass das Modell nun die beobachteten alterspezifischen Muster pädiatrischer Malaria und malaria-assoziierter Mortaliät wiedergeben kann.

Die Modellkomponente für Infektivität des Menschen setzt die Dichte asexueller Parasiten im Blut des Menschen in Beziehung zur Wahrscheinlichkeit, eine stechende Mücke zu infizieren. Dabei wird sowohl der Verzögerung, welche sich aus dem Zeitverlauf der Gametozyten-Entwicklung ergibt, als auch der Notwendigkeit, dass männliche und weibliche Gametozyten sich in ein und derselben Blutmahlzeit befinden müssen, Rechnung getragen. Die Parameter dieser Komponente wurden aus Daten von Malariatherapie-Patienten geschätzt, und das Modell ist in der Lage, die beobachteten Muster menschlicher Infektivität wiederzugeben. Das integrierte Modell wurde mit Hilfe publizierter Schätzungen des Beitrags verschiedener Altersgruppen zum Infektionsreservoir validiert.

Das Modell, zusammen mit einer hinzugefügten Komponente für die Wirkung von Sulphadoxine-Pyrimethamine (SP) und lokalitätsspezifischen Eingabedaten, reproduzierte die Resultate der bis heute durchgeführten IPTi-Feldversuche ziemlich gut. Das Modell wurde modifiziert, um verschiedene Hypothesen über den Wirkungsmechanismus von IPTi wiederzugeben. Diese Hypothesen betrafen die Zeitdauer einer Wirkung von SP, das zeitliche Auftreten von Episoden im Verlauf einer bestimmten Infektion, den möglichen Nutzen der Vermeidung einer Episode in Bezug auf Immunität, und den Effekt von subtherapeutischen SP-Konzentrationen auf die Parasitendynamik. Keine der modifizierten Versionen erklärte die Daten besser als das ursprüngliche Modell. Die Vorhersagen des integrierten Modells legen nahe, dass IPTi mit SP über eine breite Spannweite von Transmissionsintensitäten Morbidität und Mortalität bei Kleinkindern wirkungsvoll reduzieren kann. Der vorhergesagte Nutzen war im Vergleich grösser in Bezug auf Mortalität und schwere Episoden als für akute Episoden, aufgrund der altersabhängigen Komorbiditäts-Funktionen im Modell. Die zu erwartende Reduktion in der Anzahl Episoden war umso grösser, je besser die Flächendeckung von IPTi, je geringer die Effizienz der örtlichen Gesundheitsversorgung, je effektiver und je länger prophylaktisch wirksam die verwendete Substanz war. Zudem wurde vorausgesagt, dass die Anwendung von IPTi nur einen geringen Einfluss auf die lokale Übertragungsintensität hat.

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Abbreviations

ACPR	adequate clinical and parasitological cure rate
ACT	artemisinin-based combination therapy
AQ	amodiaquine
CQ	chloroquine
DDT	dichloro-diphenyl-trichloroethane
dhfr	dihydrofolate reductase
EIR	entomological incoulation rate
GSK	GlaxoSmithKline
HIV	human immunodeficiency virus
IPTi	intermittent preventive treatment in infants
IPTp	intermittent preventive treatment in pregnancy
ITN	insecticide-treated net
LBW	low birth weight
MCMC	Markov chain Monte Carlo
MMV	Medicines for Malaria Venture
MVI	Malaria Vaccine Initiative
PE	protective efficacy
RBC	red blood cell
SES	socio-economic status
SP	sulphadoxine-pyrimethamine
WSS	weighted sum of squares

Chapter 1. Introduction

1.1 The consequences of *Plasmodium falciparum* infection in humans

Plasmodium falciparum is a protozoan parasite, one of four species that account for nearly all human malaria infections.[1] Of the four, *P falciparum* causes the majority of infections in Africa and is responsible for most severe disease and mortality. Approximately 2.2 billion people globally are exposed to the risk of infection.[2]

P. falciparum has a complicated life cycle undergoing several transformations and involving both human and mosquito hosts (Figure 1.1). Human infection occurs when an infectious female *Anopheles* mosquito feeds on a human, acquiring blood that is needed for eggs to develop. Parasites in the salivary glands are transmitted as the mosquito injects saliva that acts to inhibit the blood from coagulating.

The possible consequences of infection are manifold. For the parasite, infection of a human host potentially provides a nuturing environment, providing food and enabling development and multiplication, and allows onward transmission to mosquitoes. For the human host, the clinical consequences of infection range from self-limiting parasitaemia to rapid death. Symptoms common in uncomplicated malaria episodes are fever, chills, nausea and flu-like illness. Severe episodes may involve coma or impaired consciousness, seizures, respiratory distress and severe anaemia. The reasons why infections can lead to different outcomes are not well known, but human genetics (for example, the sickle cell trait) and maternal immunity and immunity acquired through repeated exposure play a role. Estimates suggest that, annually, *P. falciparum* malaria directly causes a million deaths, 2-3 million severe clinical attacks, and about 0.5 billion acute attacks.[1,2] Additionally, infection during pregnancy can lead to adverse effects both for the mother and fetus. In areas of stable transmission, maternal anaemia and fetal growth restriction may occur. In areas of unstable transmission, the effects on both mother and infant tend to be more severe.[3] Beyond the clinical consequences, malaria has been estimated to cost Africa about 12 billion US dollars every year due to loss of productivity and tourism.[1] The social consequences are less well characterised, but it is known that the burden falls disproportionately on the poor and vulnerable.[1] The enormous burden of morbidity and mortality demands effective malaria control strategies.

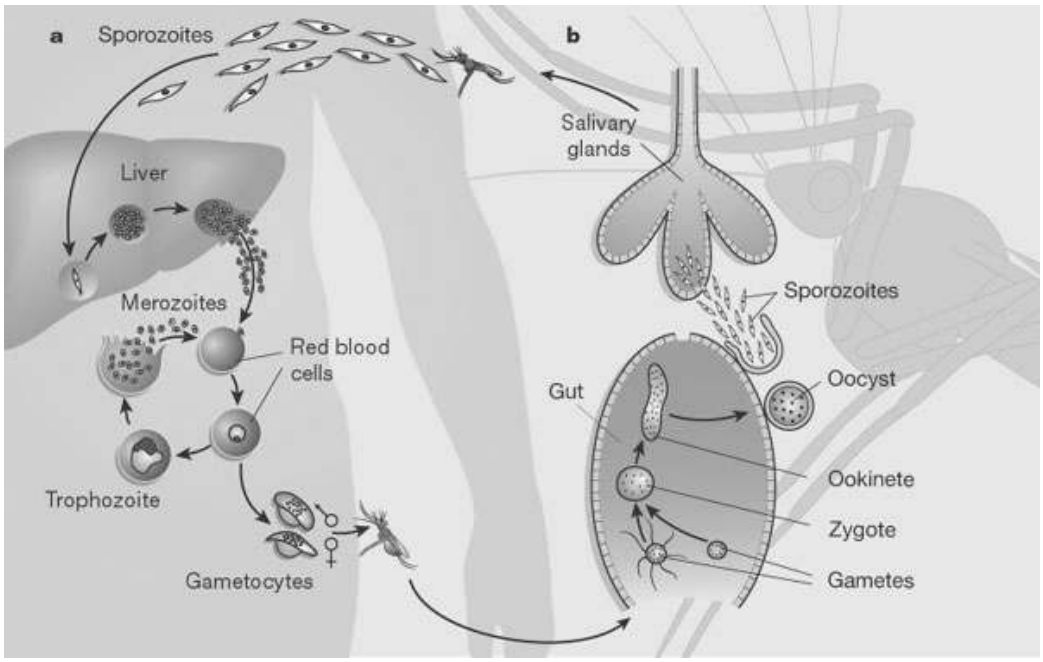


Figure 1.1 Life cycle of the parasite *Plasmodium falciparum.* Source: Wirth, 2002[4]

a. When an infectious mosquito feeds on a human, it injects the parasites in their sporozoite form. The parasites ride the flume of the circulatory system reaching the liver within a few minutes. Each *Plasmodium* invades a different liver cell. Inside the hepatocyte, the parasite digests the cell contents and undergoes development, multiplying into tens of thousands of merozoites. The cell ruptures after 5-6 days, releasing the merozoites into the bloodstream where they quickly invade red blood cells (RBC) and multiply via the trophozoite stage. When the RBC bursts, 8-32 merozoites are released each invading another RBC and the sequence of reproduction and release continues. Infected RBCs bind to endothelium or placenta, the adhesion prevents them from passing into the spleen where they would be killed. Clinical features of malaria, including fever and chills, anaemia and cerebral malaria are associated with infected RBC. A small proportion of merozoites develop into immature male and female gametocytes. **b.** A feeding mosquito takes up blood containing male and female gametocytes which develop into reproductive cells (gametes) inside the mosquito's stomach. A male gamete fuses with a female gamete to produce a zygote. The zygote in turn develops into the ookinete, which crosses the wall of the gut and forms a sporozoite-filled oocyst. When the oocyst bursts, the sporozoites migrate to the mosquito's salivary glands, and the process begins again.

1.2 A choice of malaria control strategies

There is an array of possible malaria control interventions targeting different stages in the complex life cycle of *P. falciparum*.[5] A number have proved efficacious such as insecticide-treated nets (ITN), indoor residual spraying (IRS) and artemisinin-based combination therapy (ACT). Recent developments suggest promising new interventions such as pre-erythrocytic vaccines and intermittent preventive treatment in infants and children. Funding for large-scale implementation has been limited, however in the past few years the problem of malaria has captured the attention of donors such as The Bill and Melinda Gates Foundation and the US President's Initiative and consequently funding has increased substantially. Thus, decision-makers have genuine choices and are faced with the problem of choosing between strategies, and of avoiding wrong priorities. They need to know which intervention, or combination of interventions, is likely to be the most cost-effective both for the rational use of currently-available tools and for investing in new tools for the future.

Information about the likely consequences of an intervention can come from a number of different sources. Randomised controlled trials can provide an estimate of the impact of interventions over a short timeframe, with a well-controlled delivery system, for interventions that already exist.[5,6] Malaria intervention trials frequently use acute episodes as the primary endpoint, yet the relatively rare outcomes of severe episodes and mortality are of greater concern. Monitoring of ongoing programmes takes into account the reality of health systems, but may be difficult to interpret if biases cannot be ruled out. In addition, it is difficult to field-test a large number of combinations of interventions.[7] Where data do not exist, predictions are needed in order to make comparisons between strategies. The inherent non-linearities mean that predictions are hard to make without a formal structure.[8] In these circumstances, mathematical models are one of the few tools available to decision-makers.

1.3 The need for a comprehensive model of malaria epidemiology

Malaria has been modelled for over a hundred years. For a large proportion of that time, models, including the landmark works of Ross,[9] Macdonald[10] and Dietz *et al*,[11] focused on transmission dynamics. They were concerned with the connection between entomological circumstances and the parasitological status of a population. They did not include the determinants of entomology, the consequences of infections, or the social, economic and behavioural factors that modify the parasitology.[12] They were created to be used for specific questions, such as predicting whether elimination might result from given interventions. As hopes for global eradication subsided, public health priorities shifted from eradication to control and the reduction of the burden of disease.[13] Very recently, there has been interest in elimination, however the reduction of morbidity and mortality is likely to remain a priority. New models capable of making predictions of the impact of a control strategy on morbidity, mortality and cost-effectiveness are required.

A model of the impact of interventions on malaria morbidity and mortality must include not only the direct short-term effects, but also the longer-term dynamic effects due to changes in the immune status of the population through reduced exposure and benefits from lowered transmission intensity.[7,14] The effects of a health system on the gains expected from an intervention must also be taken into account.

> "*No sensible decision can be made any longer without taking into account not only the world as it is, but the world as it will be*" Isaac Asimov

Calls have previously been made for an integrated model.[6,8,15,16] Twenty-five years ago Bailey suggested a multidisciplinary approach, linking a biomathematical model of malaria to an econometric model.[15] He observed that a model combining these elements would allow clinical and epidemiological knowledge to be translated more effectively into the achievement of social goals.[15] Others have proposed a focus on interventions and decision-making aids.[6,8] Although there are many models of transmission or of specific aspects of malaria, until recently there has been no model that simultaneously captures the dynamics of infection, acquired immunity, parasite densities, the consequences of infections (morbidity, mortality and infectivity to mosquitoes), the health system and economics.

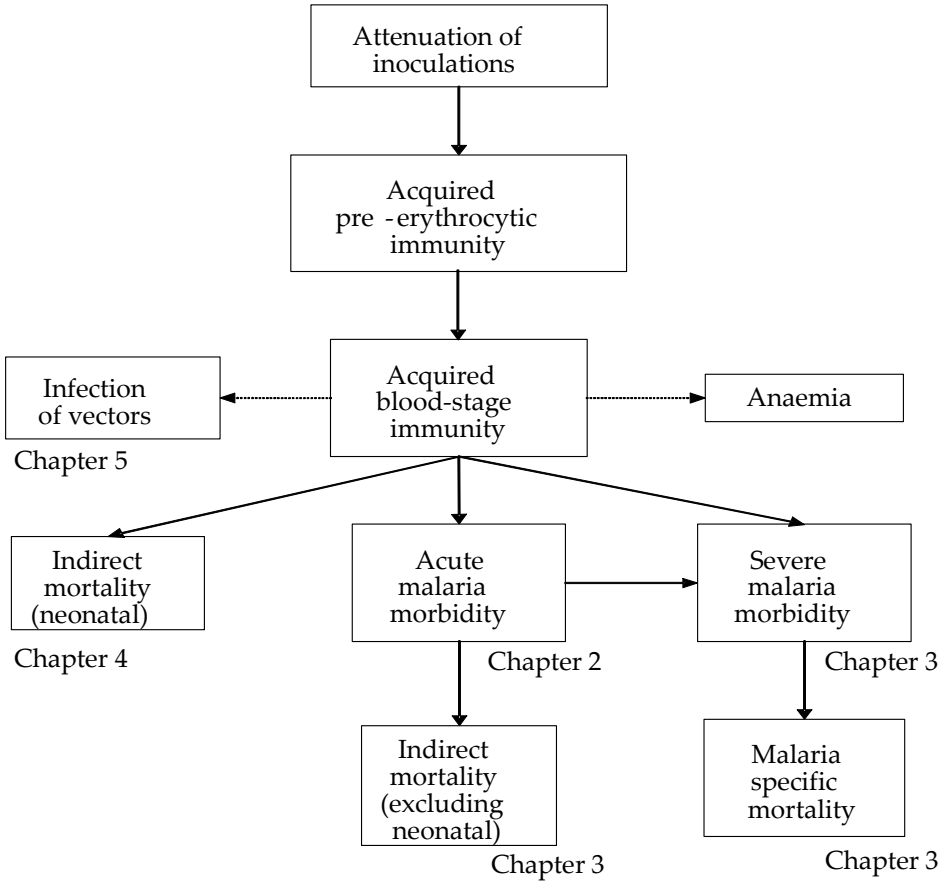
In response to the need for an integrated model, the malaria modelling group at the Swiss Tropical Institute has developed a set of stochastic simulations that captures all of these elements. The details of how the infection process has been modelled are reported elsewhere.[17,18] This thesis describes those elements of the integrated model that consider the immediate consequences of human infection: acute morbidity, severe morbidity, mortality and transmission to the vector.

1.4 The strategy for a new integrated model

The development of the model followed a strategy. The modular structure allowed the concurrent development of components and ensured that the fitting of the model to data was feasible in terms of the computational demands and complexity of fitting.

The core of the model is the specification of the course of infections: a description of the asexual blood-stage parasite densities (Figure 1.2). The densities provide a basis for the effects of naturally acquired immunity, for accommodating superinfection, and for model components for the consequences of infection. Naturally acquired immunity acts to reduce density, rather than the duration of infection.[7,19,20] High parasite densities are a trigger for clinical malaria, and the probability that a mosquito is infected when feeding on an infected human depends on the gametocyte density.

Figure 1.2 Integrated model components



Adapted from Smith *et al*.[5]

The model is individual-based in that individual humans and individual infections are simulated, although individual mosquitoes are not. The model must accurately describe immunity to malaria which is partial and gradually acquired[21] and which provides a basis to incorporate the density-based consequences of infection (clinical outcomes and infectivity to mosquitoes). An individual-based approach was chosen because it can represent both partial immunity and parasite densities extremely easily. Classical mathematical forms would struggle to encompass the required range of details.[16] However, compartmental models that can incorporate partial immunity have been developed. They bridge the gap between Susceptible-Infected-Susceptible and Susceptible-Infected-Recovered models either by continuous immunity[22] or in discrete steps.[23] Individual-based models have previously been used to describe malaria transmission dynamics.[24,25]

The model is stochastic. An individual malaria infection can last many months[26], during which densities of both asexual parasites and gametocytes vary irregularly as consequences mainly of the developmental cycle of the parasite, of immunity, and of antigenic variation. These processes are not well understood and the strategy was to avoid predicting intermediate variables whose quantitative relationship with epidemiological outcomes are uncertain[5]. Since the course of each infection is different,

and the average behaviour is of less importance than the extent of variation, the irregularities in parasite densities can be captures as statistical fluctuations. Convincing models for individual infections (such as [27,28]) have adopted stochastic simulation approaches, rather than treating the development of an individual malaria infection as a deterministic process.

"*Life is a stochastic process.*" Anon

With an individual-based stochastic model, discrete time is the easiest option. Five days was a pragmatic choice for the time interval, as a reasonable common denominator for important time periods such as the hepatic stage (five days) and pre-patent blood stage (ten days), and also for computational demands.

The overall strategy sets some general criteria for the model components for the consequences of infection. They must use available interconnecting variables as their starting point, be individual-based, five-day timestep models, use biologically-plausible mechanisms, reproduce age-patterns in available data, be fitted to data and allow modification for the effect of interventions.

1.5 Applying the model: intermittent preventive treatment in infants

Intermittent preventive treatment in infants (IPTi) is being considered as a new intervention with the potential to reduce the burden of malaria in infants via a simple delivery system at low cost. The strategy of IPTi is to give antimalarial drugs during the first year of life at the time of routine immunizations, irrespective of whether the infants are known to have malaria infections.[29] The limited number of doses is intended to retain the benefits of weekly or fortnightly chemoprophylaxis whilst avoiding the disadvantages: thus reducing malaria morbidity and mortality without incurring difficulties in sustainability, accelerating drug resistance or impairing the development of natural immunity.

Six trials of IPTi with sulphadoxine-pyrimethamine (SP) have been completed to date. Questions have arisen. The reasons for the variability in the trial estimates of efficacy are not known, nor the process by which IPTi might work, nor the likely impact of IPTi in different epidemiological settings. This thesis describes the application of the integrated model to these questions. The impact of IPTi on drug resistance has been considered elsewhere.[30,31]

1.6 Objectives of the thesis

- Model components for the consequences of infections in the human host which can be integrated into a comprehensive model of malaria epidemiology. These comprise acute morbidity (Chapter 2), severe morbidity (Chapter 3), and malaria mortality (including via maternal infection) (Chapters 3 and 4) and infectivity to mosquitoes (Chapter 5).

- The application of the model, with additional components for the effects of SP, to simulate the IPTi trials, investigate hypotheses for the mechanism of IPTi and make predictions of the impact of IPTi in different settings (Chapter 6).

1.A References

1. Greenwood B, Bojang K, Whitty C, Targett G. Malaria. *Lancet* 2005, 365: 1487-1498

2. Snow RW, Guerra CA, Noor AM, Myint HY, Hay SI. The global distribution of clinical episodes of *Plasmodium falciparum* malaria. *Nature* 2005, 434: 214-217

3. Desai M, ter Kuile F, Nosten F, McGready R, Asamoa K, Brabin B, Newman R. Epidemiology and burden of malaria in pregnancy. *Lancet Inf Dis* 2007, 7: 93-104

4. Wirth D. The parasite genome: biological revelations. *Nature* 2002, 419: 495-496

5. Smith T, Killeen GF, Maire N, Ross A, Molineaux L, Tediosi F, Hutton G, Utzinger J, Dietz K, Tanner M. Mathematical modeling of the impact of malaria vaccines on the clinical epidemiology and natural history of *Plasmodium falciparum* malaria: Overview. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 1-10

6. McKenzie FE, Samba EM. The role of mathematical modeling in evidence-based malaria control. *Am J Trop Med Hyg* 2004, 71 Suppl 2: 94-96

7. Smith T, Maire N, Ross A, Penny M, Chitnis N, Schapira A, Studer A, Genton B, Lengeler C, Tediosi F, de Savigny D, Tanner M. Towards a comprehensive simulation model of malaria epidemiology and control. *Parasitol* 2008, in press.

8. Koella JC. On the use of mathematical models of malaria transmission. *Acta Trop* 1991, 49: 1-25

9. Ross R. The prevention of malaria. 2nd ed. London: Murray, 1911

10. Macdonald G. The epidemiology and control of malaria. London: Oxford University Press, 1957

11. Dietz K, Molineaux L, Thomas A. A malaria model tested in the African savannah. *Bull WHO* 1974, 50: 347-357

12. Molineaux L. The pros and cons of modelling malaria transmission. *Trans R Soc Trop Med Hyg* 1985, 79: 743-747

13. Litsios S. The tomorrow of malaria. Wellington: Pacific Press, 1996

14. Struchiner CJ, Halloran ME, Spielman A. Modeling malaria vaccines. I: New uses for old ideas. *Math Biosci* 1989, 94: 87-113

15. Bailey N. The biomathematics of malaria. London: Charles Griffin and Co Ltd, 1982

16. McKenzie FE. Why model malaria? *Parasitol Today* 2000, 16: 511-516

17. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 19-31

18. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationship between the entomologic inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 11-18

19. Sama W, Killeen G, Smith T. Estimating the duration of *Plasmodium falciparum* infection from trials of indoor residual spraying. *Am J Trop Med Hyg* 2004, 70: 625-634

20. Sama W, Owusu-Agyei S, Felger I, Dietz K, Smith T. Age and seasonal variation in the transition rates and detectability of *Plasmodium falciparum* malaria. *Parasitol* 2006, 132: 13-21

21. Anderson RM, May RM. Infectious diseases of humans: dynamics and control. Oxford: Oxford University Press, 1991

22. White L, Medley G. Microparasite population dynamics and and continuous immunity. *Proc Biol Sci* 1998, 265: 1977

23. Gomes M, White L, Medley G. Infection, reinfection, and vaccination under suboptimal immune protection: epidemiological perspectives. *J Theor Biol* 2004, 228: 539

24. Gu W, Killeen G, Mbogo CM, Regens JL, Githure JI, Beier JC. An individual-based model of *Plasmodium falciparum* malaria transmission on the coast of Kenya. *Trans R Soc Trop Med Hyg* 2003, 97: 43-50

25. McKenzie FE, Bossert WH. An integrated model of *Plasmodium falciparum* dynamics. *J Theor Biol* 2005, 232: 411-426

26. Collins WE, Jeffery GM. A retrospective examination of sporozoite- and trophozoite-induced infections with *Plasmodium falciparum*: development of parasitologic and clinical immunity during primary infection. *Am J Trop Med Hyg* 1999, 61: 4-19

27. Molineaux L, Diebner HH, Eichner M, Collins WE, Jeffery GM, Dietz K. *Plasmodium falciparum* parasitaemia described by a new mathematical model. *Parasitol* 2001, 122: 379-391

28. Gatton ML, Cheng Q. Modeling the development of acquired clinical immunity to *Plasmodium falciparum* malaria. *Infect Immun* 2004, 72: 6538-6545

29. IPTi Consortium. www.ipti-malaria.org. 2008.

30. Alexander N, Sutherland C, Roper C, Cissé B, Schellenberg D. Modelling the impact of intermittent preventive treatment for malaria on selection pressure for drug resistance. *Malar J* 2007, 6: 9

31. Prudhomme O'Meara W, Smith D, Mckenzie F. Potential impact of intermittent preventive treatment (IPT) on spread of drug-resistant malaria. *PLoS Med* 2006, 3: 141

Chapter 2

An epidemiological model of the incidence of acute illness in *Plasmodium falciparum* malaria

Thomas Smith, Amanda Ross, Nicolas Maire, Christophe Rogier,
Jean-François Trape and Louis Molineaux

Swiss Tropical Institute, Basel, Switzerland
Institut de Médicine Tropicale du Service de Santé, France
Institut de Recherche pour le Développement, Dakar, Senegal
retired, formerly WHO, Geneva, Switzerland

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2.1 Summary

We propose a stochastic model for simulating malaria tolerance. The model relates the probability of a clinical attack of malaria to the peripheral parasite densities via a pyrogenic threshold that itself responds dynamically to the parasite load. The parameters of the model have been estimated by fitting it to the relationship between incidence of clinical episodes and the entomological inoculation rate, using age-specific incidence data from two villages in Senegal and one village in Tanzania. The model reproduces the shifts in age distribution of clinical episodes associated with variation in transmission intensity, and in keeping with the data, predicts a slightly higher lifetime number of episodes in the mesoendemic village of Ndiop than in the holoendemic village of Dielmo. This model provides a parsimonious explanation of counter-intuitive relationships between the overall incidence of clinical malaria and transmission intensity. In contrast to the theory of endemic stability, recently proposed to apply to *P. falciparum*, it does not assume any intrinsic age dependence in the outcome of infection. This model can be used to explore the consequences for predictions of the effects of different anti-malarial interventions on the incidence of clinical malaria.

2.2 Introduction

The clinical outcome of *Plasmodium falciparum* malaria infection can range from an absence of detectable morbidity to rapid death.[1] In naive hosts, symptoms occur before the first peak of parasitaemia, but untreated infections can persist for many months with intermittent periods of acute illness. In malaria-endemic areas of sub-Saharan Africa, exposed people are subjected to frequent superinfections, and develop partial immunity that leads to control both of parasite densities and to reduction in the frequency of clinical episodes. Malaria morbidity is shifted into older ages as transmission intensity is reduced. This has been studied intensively in two villages in Senegal.[2,3] In Dielmo, where the annual entomological inoculation rate (EIR) is estimated to be approximately 200,[3,4] almost all episodes are concentrated in the first years of life. In Ndiop, with an annual EIR of 20,[5] there is a substantial peak shift, with a high incidence in adolescents and adults. In Ndiop, the EIR was detectable only during the short rainy season, whereas in Dielmo it was detectable throughout the year. The published data from Ndiop and Dielmo do not provide a breakdown of the age-pattern in the first year of life. In Idete in Tanzania, where transmission intensity is similar to Dielmo,[3,6] the incidence of clinical attacks in the first three months of life is very low, but increased strongly with age.[6] A higher number of lifetime episodes occurred in the lower transmission setting of Ndiop compared to Dielmo (even assuming the same life expectancy), a pattern seen elsewhere.[7] To predict the potential impact of interventions that affect parasitaemia, mathematical models are needed that predict not only the likely incidence of infections but also how frequently these will result in clinical episodes of malaria.

There is abundant evidence that most clinical episodes are caused by newly inoculated genetically distinct parasites.[8,9] One proposed model is that parasite populations are structured into a limited number of strains, each stimulating long-term clinical immunity.[10,11] However most analyses of the population biology of *P. falciparum* have concluded that there is frequent genetic exchange,[12-15] many malaria antigens are extremely polymorphic,[16-18] cross-protection is clearly important, and natural immunity to the immunodominant epitopes is not necessarily lifelong.[19-21]

The adequate modelling of all these complex immunological phenomena represents a major challenge. However epidemiological analyses of the tolerance of parasites can be used to predict the likelihood of clinical episodes as a function of densities of peripheral parasitaemia without explicitly considering how those densities occur.[22-25] In a study carried out in Dielmo, where parasitaemia was assessed twice weekly Rogier and others estimated well-defined pyrogenic thresholds for different ages of human host.[24] We have now further analysed this data to derive predictions of the thresholds as functions of recent levels of parasitaemia, rather than of the age of the host. We have linked these predictions to a stochastic model that predicts parasite densities in endemic areas as a function of the pattern of transmission[26,27] and fitted the model for the incidence of clinical episodes to field data from different epidemiological settings in Ndiop, Dielmo and Idete.

The resulting model enables us to predict, for a wide range of malaria transmission settings, the occurrence of clinical episodes and to assess the likely effects of interventions on the incidence of clinical attacks.

2.3 Methods

2.3.1 Model for parasite densities

The starting point for our model for the incidence of clinical malaria is an individual-based stochastic simulation model for *P. falciparum* parasitology.[26,27] This model makes predictions of the parasite density for each member of the simulated population using a five-day time step, with the seasonal pattern of the EIR as input. The parasite densities are sampled from log normal distributions. We compared the observed parasitological data to the predictions of this model for the Ndiop and Dielmo transmission patterns,[3,5] to evaluate its appropriateness as a basis for the predicting clinical episodes in this setting.

2.3.2 Model for clinical malaria episodes

The parasitological simulation includes stochastic variation between individual humans in average parasite densities and also stochastic variation around that average.[26] We model clinical immunity as a function of these stochastically varying

parasite densities, and of a set of five parameters that are independent of the individual and of the transmission setting.

To predict the clinical outcome, for each five-day time step we draw five independent samples from the simulated parasite density distribution for each concurrent infection (to simulate potential daily changes in morbidity status) and consider only the maximum, $Y_{\max}(i,t)$, of the simulated densities to determine whether a clinical episode occurred. When the host is infected by several concurrent infections it is likely that one of these contributes the bulk of the parasite load, so it is logical to define $Y_{\max}(i,t)$ as the maximum over all infections.

A simple model is to assume that for each host there is a specific parasite density, or pyrogenic threshold, at which symptoms (e.g. fever) are triggered. Rogier and others[24] considered a cohort of the inhabitants of the holoendemic village of Dielmo, Senegal and fitted a step function to the probability of fever as a function of parasite density. The parasite density at which the step occurs corresponds to the pyrogenic threshold, which was shown to vary with age.

In general, it is not realistic to assume that all individuals of the same age will have exactly the same pyrogenic threshold,[28-31] so it is more reasonable to expect a sigmoidal relationship between the risk of fever and the parasite density than a step function. We therefore propose a model in which the probability that an episode occurs in individual *i*, at time *t*, is related to the parasite density via a function of the following form

$$P_m(i,t) = \frac{Y_{\max}(i,t)}{Y^*(i,t) + Y_{\max}(i,t)} \qquad (2.1)$$

where $Y^*(i,t)$, the pyrogenic threshold for individual *i* at time *t*, is defined as the parasite density at which the probability of a clinical episode reaches 0.5, and $Y_{\max}(i,t)$ is the maximum density during the time interval *t* (note that we present only the formulae for our final choice of models).

The age pattern in the pyrogenic threshold in Dielmo, together with data derived from other study sites,[29,32,33] supports the idea that the density of parasites required to stimulate acute pathology is higher in individuals who have been recently exposed to high parasite densities. This may be a result of stimulation of immune responses to toxins released at schizogony, and very likely involves physiological tolerance of cytokines.[34] The mechanism must be consistent with both rapid acquisition and rapid loss of tolerance and cannot be a simple function of antibody against toxin, which have a completely different age-pattern from that of the pyrogenic threshold.[35]

We model the dynamics of the pyrogenic threshold with a function of the form

$$\frac{dY^*(i,t)}{dt} = f_1\left(Y(i,t)\right) f_2\left(Y^*(i,t)\right) - \varpi Y^*(i,t) \qquad (2.2)$$

where $f_1\left(Y(i,t)\right)$ is a function describing the relationship between accrual of tolerance and the parasite density $Y(i,t)$; $f_2\left(Y^*(i,t)\right)$ describes saturation of this accrual process at high values of $Y^*(i,t)$, and the term $\varpi Y^*(i,t)$ leads to decay of the threshold with first order kinetics. The decay ensures that the model conforms to the epidemiological evidence suggesting that parasite tolerance is short lived.

We define the function $f_1\left(Y(i,t)\right)$ in such a way as to ensure that the stimulus is not directly proportional to $Y(i,t)$ but rather that it asymptotically reaches a maximum at high values of $Y(i,t)$, using

$$f_1\left(Y(i,t)\right) = \frac{\alpha Y(i,t)}{Y_1^* + Y(i,t)}. \qquad (2.3)$$

To ensure saturation of the accrual process, we require that at high values of $Y^*(i,t)$, a higher parasite load is required to achieve the same increase by defining

$$f_2\left(Y^*(i,t)\right) = \frac{1}{Y_2^* + Y^*(i,t)} \qquad (2.4)$$

Overall therefore we propose the following dynamics for $Y^*(i,t)$

$$\frac{dY^*(i,t)}{dt} = \frac{\alpha Y(i,t)}{\left(Y_1^* + Y(i,t)\right)\left(Y_2^* + Y^*(i,t)\right)} - \varpi Y^*(i,t) \qquad (2.5)$$

where α, Y_1^*, and Y_2^* are constants to be estimated. To complete the specification of the model, we set the initial conditions to be $Y^*(i,0) = Y_0^*$ at the birth of the host, thus defining a further parameter Y_0^*.

2.3.3 Data sources

We fitted the model for acute episodes to two distinct datasets. The first was published data on the age pattern of clinical episodes in the villages of Ndiop and Dielmo in Senegal.[2] The village populations were visited daily to detect and treat any clinical malaria attacks (with quinine). Hence, effectively all acute episodes were thought to be treated in these villages. In the simulations of Dielmo and Ndiop we assumed that there had been no treatment of clinical malaria prior to the start of the follow-up period. To ensure that the analysis remains tractable, we approximate the patterns of transmission with recurring annual cycles (although there was variation between years in the predominant vectors and seasonality of transmission).

We also compared the predicted patterns from the simulation model for *P. falciparum* parasitology with those from parasitological surveys in these two villages to evaluate its appropriateness as a basis for predicting clinical episodes in this setting. In Dielmo, two thick blood smears were prepared per week in each individual from 29 May to 30 September 1990. In Ndiop, one thick blood smear was prepared per week in each individual from 15 July 1993 to 15 January 1994 and one per month from 1 February 1994 to 15 July 1995. Slides were only declared negative after 200 high-power fields had been scanned for parasites. Parasite densities were originally expressed as the parasite:leucocyte ratio. To adjust these densities to the same scale as that used in fitting the simulation model to other datasets, the parasite:leucocyte ratios were then multiplied by a factor of 1,416 to give a notional density in parasites/microliter of blood.[26]

The model was fitted to a second dataset of age-incidence rates for clinical malaria in infants less than one year of age recorded at the health centre in the village of Idete, Tanzania, from June 1993 to October 1994.[6,25] These data were included to estimate the initial conditions (the value of Y_0^*) and to ensure that the model predicts the age pattern of acute episodes that is actually observed in infants. For the Idete data we used the case definitions and age groups in the paper by Vounatsou and others[25] and the annual pattern of inoculations reported by Charlwood and others[36] as input. We assume a common value of Y_0^* across all sites and therefore require data for infants from only a single transmission setting.

2.3.4 Implementation and fitting of the simulation model

To obtain estimates of the five parameters α, ϖ, Y_0^*, Y_1^*, and Y_2^* we fitted the model to the age-pattern of clinical malaria in all three villages (i.e. Ndiop and Dielmo in Senegal, and Idete in Tanzania) and simultaneously to the pyrogenic thresholds for Dielmo estimated by Rogier and others[24] (Table 2.1).

For Dielmo and Ndiop we further predicted parasite densities for a sample of 10,000 individuals over a 10 year period, drawn from the age-groups of interest. For Idete, where we were concerned only with infants less than one year of age, we used a sample size of 2,000. In each village we assumed a typical sub-Saharan African age-distribution taken from the demographic surveillance area that includes Idete.[37]

Simulated clinical episodes of malaria occurred with probability $P_m(i,t)$, which was dependent on both the simulated maximum density and the current value of $Y^*(i,t)$ for each individual and each five-day time point in the 10-year follow-up period. In the simulations of Ndiop and Dielmo we simulated effective treatment of all clinical episodes within the five-day period in which they occurred. In the simulation of Idete we assumed that some proportion, P_t, of the episodes were effectively treated (i.e the parasites were cleared within the course of one time interval), and that this proportion

corresponded to the proportion of episodes reported to the village dispensary. In Idete village, simulated episodes occurring within 30 days of a preceding episode were not counted (these have been registered in the surveillance system as recrudescence, rather than new episodes). In Ndiop and Dielmo this restriction did not hold.[38]

For each simulated individual in each village the model thus predicted the incidence of clinical malaria, as a stochastic function of the inoculation rate. These incidences were summarised over age groups and compared with the published values.[2,25] Similarly, the model predicted the pyrogenic threshold, $Y^*(i,t)$, at each time point for each individual. The geometric mean of these values was calculated for each age group in the simulation of Dielmo village, and the logarithms of these values compared with the logarithms of the age-specific pyrogenic thresholds estimated by Rogier and others.[24] Simulated annealing[39,40] was used to identify the parameter values that minimised the residual sum of squares summed over all three villages and both outcomes for Dielmo. The Fisher information estimated from a least squares quadratic fit to the residual sum of squares was used to give approximate confidence intervals.

2.4 Results

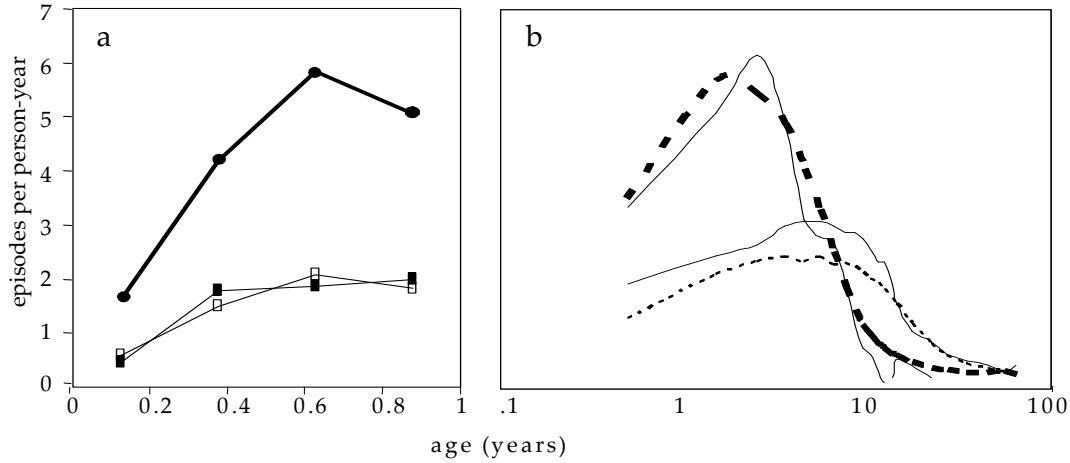
The parameter estimates are given in Table 2.1. Our model was able to reproduce the age incidence patterns very well considering that only five parameters were fitted across three datasets (Figure 2.1).

Table 2.1 Parameter estimates from the best fitting model*

Para-meter	Meaning of parameter	Estimate	95% Confidence Intervals
α	Factor determining increase in $Y^*(i,t)$	143,000 parasites^{2}µl^{-2}day^{-1}	103,000-197,000
ϖ	Decay rate of pyrogenic threshold	2.5 year^{-1}	2.1-3.0
Y_0^*	Pyrogenic threshold at birth	296.3 parasites/µl	3-30,000
Y_1^*	Critical value of parasite density in determining increase in $Y^*(i,t)$	0.60 parasites/µl	0.17-2.13
Y_2^*	Critical value of $Y^*(i,t)$ in determining increase in $Y^*(i,t)$	6.5x10^3 parasites/µl	5.2x10^3 - 8.2x10^3
P_t	Compliance in Idete (proportion of episodes detected and treated)	0.36	0.27-0.48

*The residual sums of squares for the three datasets were 0.2 (Idete), 4.4 (Ndiop) and 3.4 (Dielmo), computed from 4, 22 and 22 distinct age groups, respectively (corresponding to a total of 43 residual degrees of freedom. The residual sum of squares for the pyrogenic threshold for Dielmo was 3.3

Figure 2.1 Age incidence curves of clinical malaria

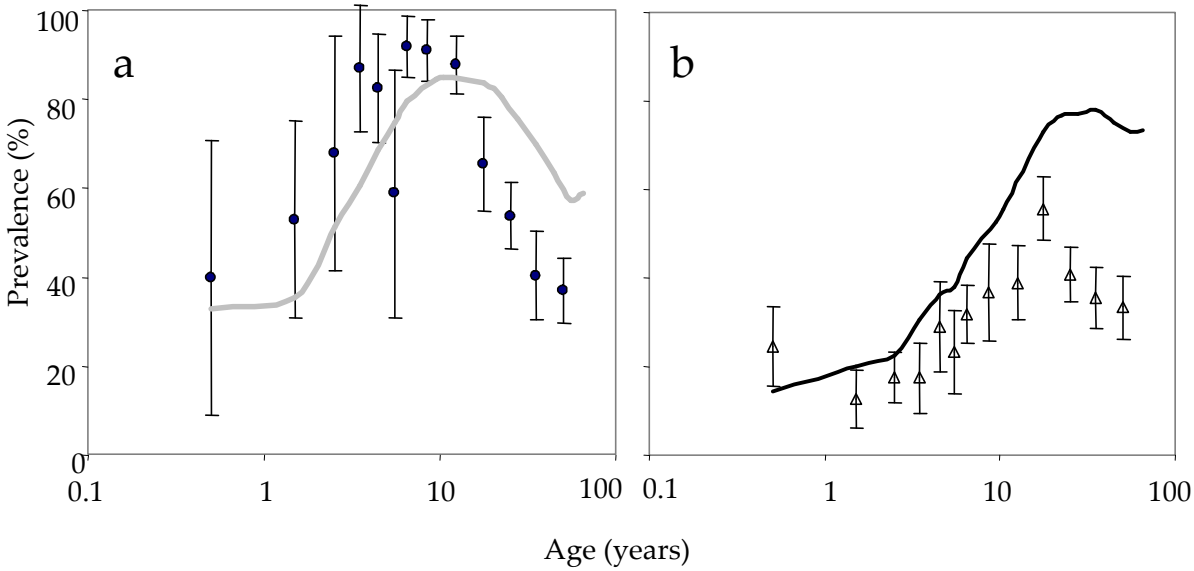


a. Idete (Tanzania). Filled squares = measured incidence of clinical malaria at health centre; open squares = model prediction for incidence of clinical malaria at health centre; thick line = model prediction for overall incidence of clinical malaria**. b. Ndiop and Dielmo (Senegal).** Thick dotted line = model prediction of incidence of clinical malaria in Dielmo; thin dotted line = model prediction of incidence of clinical malaria in Ndiop; Thin lines = observed incidence of clinical malaria in Ndiop and Dielmo

The value of ϖ, estimated as 2.5/year, implied that in the absence of stimulation, the pyrogenic threshold decays with a half life of 0.28 years. The predicted total numbers of episodes up to age sixty were 56 for Ndiop (EIR=20) and 53 for Dielmo (EIR=200), compared with the published overall incidence of clinical malaria cumulative numbers of episodes up to the age of sixty of 62 and 43, respectively.[2] In the simulations of both villages the age of peak incidence was a little younger than in the data. The predicted incidence in the youngest individuals was higher in Dielmo, but lower in Ndiop, in comparison with the observed values. The extent of the peak shift was similar in the model to the data.

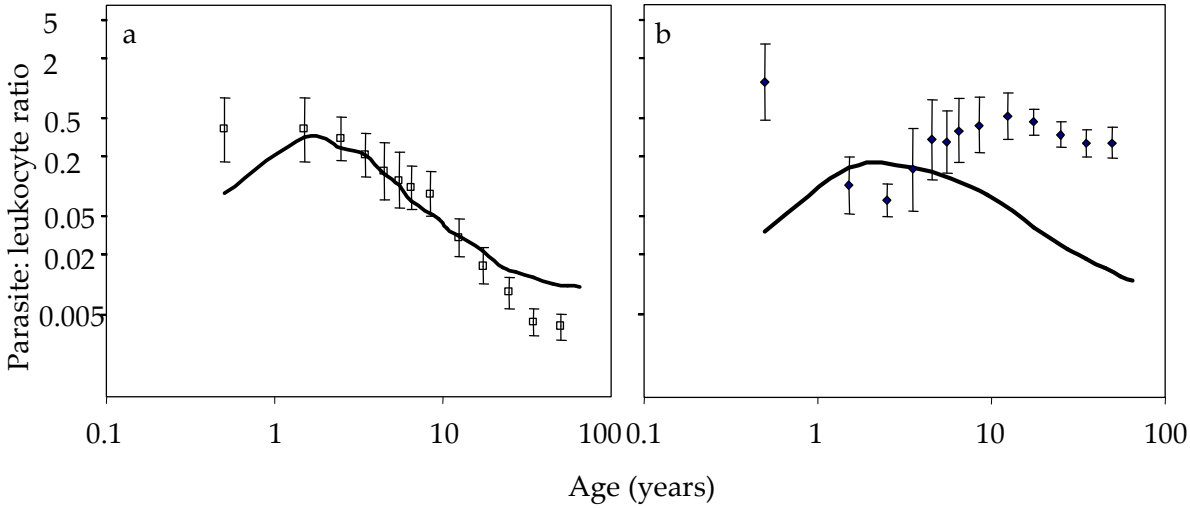
Although the model was not fitted to the patterns of age prevalence and of age density in Dielmo or Ndiop, it does make predictions of these quantities which we could therefore compare with the observed curves. The predicted age-prevalence curve for Dielmo was very similar to that observed (Figure 2.2.a), as were the predicted geometric mean densities in children in that village (Figure 2.3a). In adults the model predicted rather higher densities than those observed in Dielmo, while for adults in Ndiop the model predicted higher prevalence in adults (Figure 2.2b) but lower densities (Figure 2.3b) than those observed. This would be expected if the burden of malaria is concentrated in a smaller proportion of individuals in Ndiop than in the dataset to which the parasitological model was fitted. A reasonably good fit was obtained for the average pyrogenic threshold, but the model did not give a very good fit to the age-trend in $Y^*(i,t)$, predicting that the peak was at a greater age than the estimates of Rogier and others[24] (Figure 2.4).

Figure 2.2 Parasite prevalence by age



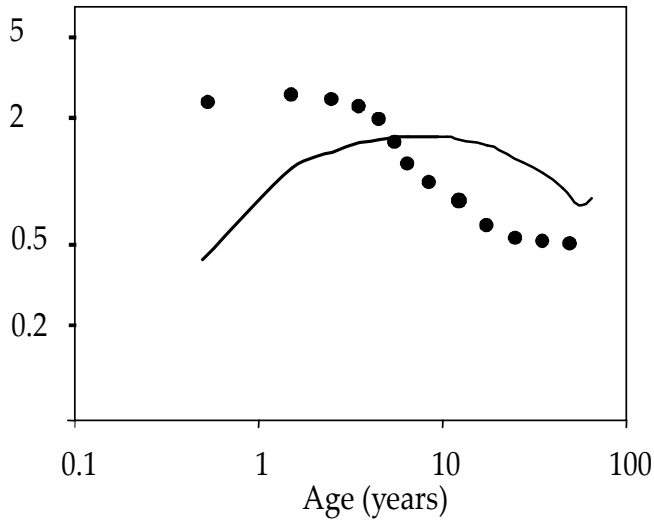
a. Dielmo. Points and error bars show prevalence of patent parasitaemia and 95% confidence intervals determined in surveys from 1990 to 1994. Continuous line= model predictions. **b. Ndiop.** Points and error bars: prevalence of patent parasitaemia and 95% confidence intervals determined in surveys from 1990 to 1994. Continuous line= model predictions. Prevalence is assessed as the proportion of individuals with parasite density (simulated or observed) above the actual level of detection used in the field study.

Figure 2.3 Geometric mean parasite densities by age



a. Dielmo. Points and error bars show geometric mean and 95% confidence intervals of densities of patent parasitaemia determined in surveys from 1990 to 1994. Continuous line= model predictions of the geometric means. **b. Ndiop**. Points and error bars show geometric mean and 95% confidence intervals of densities of patent parasitaemia determined in surveys from 1990 to1994. Continuous line= model predictions of the geometric means.

Figure 2.4 Pyrogenic threshold in the village of Dielmo, Senegal



Filled circles =pyrogenic threshold by age in Dielmo (results of Rogier and others [24]). Continuous line= model prediction

2.5 Discussion

Our model can reproduce the patterns of the age-specific incidence of acute episodes from the three transmission settings. In particular, we were able to reproduce both the shape of the age-specific incidence curves and the total lifetime incidence of acute episodes for sites with very different transmission intensities with a model with only five parameters. Within this model, the higher incidence of clinical attacks in older individuals in Ndiop than in Dielmo arises both because of lower immunological control of asexual blood stages and less clinical tolerance.

This good fit was obtained despite the use of a parasitological model that only crudely reproduces within-host parasite dynamics, since we fitted it to cross-sectional data.[26] Day-to-day variation in parasite densities may be critical in determining levels of tolerance, and our model, based on five-day time steps, did not aim to simulate this accurately. This may explain why the density of patent parasitaemia did not appear to be very important, and may also be the explanation of why we could not obtain a better fit for the age-pattern of the pyrogenic threshold. It is possible that the important variations in density and levels of tolerance are much more rapid than our model could capture, especially if they involve physiological tolerance of cytokines.[34]

The relatively poor prediction of parasite prevalence and density in adults in Ndiop is possibly because the model assumes the degree of within-village heterogeneity in transmission to be the same in each village. Focality of transmission leads to lower prevalence but higher densities in those who are infected because of increased levels of super-infection. Based on these criteria, transmission in Ndiop appears to be more focal than that in the villages to which we fitted the parasitological model.[26] Within our model effects of focal transmission on incidence of clinical episodes should be only of secondary importance because there is little interaction between concurrent co-infections. Thus, at any given level of immunity the incidence of clinical episodes depends primarily on the overall force of infection and not on how the infections are distributed between individuals.

It was not possible to obtain a better fit for infants in Ndiop because the number of infection events predicted for this age group by our model of infection[27] is less than the number of clinical episodes. We have assumed all episodes are immediately treated so that no more than one episode can occur for any one infection event, but this was not necessarily always the case. We assumed mosquito biting to be proportional to body surface area, using Tanzanian anthropometric data to estimate age-specific surface areas.[41] Different patterns of human growth or mosquito behavior may account for some of the discrepancies. Selection effects that might arise because of differential mortality of susceptible individuals are a further factor that we did not take into account.

Our model assumes particular functional forms for the relationships between the pyrogenic threshold and the risk of clinical episodes, and the pyrogenic threshold and

the parasite density itself. Exploratory analyses indicated that the fitted age-incidence relationships are not very sensitive to the exact functional forms used for these relationships. Empirical relationships between parasite density and risk of illness depend on how the cases are detected. In the studies in Senegal, parasitaemia and fever were monitored daily so episodes were generally detected early and this may account for the abrupt pyrogenic thresholds reported by Rogier and others [24]. More usually, fever cases are detected when they report to a health facility, as in the study in Idete.[6] The arrival of the cases at the health facility is at varying intervals after the beginning of the episode and this tends to blur the relationship between fever risk and parasite density. If fever episodes are detected at household visits, which are carried out at intervals of more than a few days at times that are unrelated to the onset of disease, then the relationship between parasite densities and fever risk is weaker (e.g.[22,42]).

In areas endemic for *P. falciparum* malaria, the incidence of clinical attacks is highly age dependent, with the peak incidence occurring at younger ages the higher the transmission. Such peak shifts are not only characteristic for malaria but also for many other infectious diseases.[43] A superficially similar shift is also seen in patterns of age prevalence for *P. falciparum,*[44] but the peak in prevalence is generally reached at an older age than that of acute morbidity. Unlike the pattern for clinical episodes, reduction in transmission is associated with reduction in infection prevalence over almost all of the age range, (although in some age groups there may be a small increase). The peak shift in clinical attacks is more pronounced than that in prevalence and the incidence of acute malaria attacks in older children and adults can be substantially greater at low transmission levels than at high ones (Figure 2.1).

The observation that reduction in transmission may lead to an increased incidence of disease in *P. falciparum* has been attributed to the phenomenon of endemic stability observed with many veterinary pathogens.[45] For endemic stability to occur there must be at least two processes accounting for the age-incidence curves, one leading to an increase in incidence with age in the youngest age groups, and the other to a decreasing incidence in older individuals. Coleman and others[45] suggest that the first of these conditions must be satisfied by a worsening of the outcome of infection with age over at least part of the age range. Our model demonstrates that this assumption is not necessary, for we explain the initial increase in morbidity with age as a consequence of increase in exposure to mosquitoes as the host grows in body surface area.[27] Idete, Dielmo and Ndiop are all villages with stable endemic *P. falciparum.* While Ndiop is an example of mesoendemicity, contrasting with the holoendemic transmission in Idete and Dielmo, it still experiences a much higher EIR than areas of unstable transmission. The theory of endemic stability therefore needs adapting for the analysis of the case of endemic malaria.

We propose to use the model component described by equations 2.1 to 2.5 as part of a comprehensive model for examining the likely consequences of a wide range of interventions, including vaccination. The incidence of acute illness is only one of these consequences, which can include severe life-threatening disease, chronic anaemia, and

indirect mortality. Even an intervention that leads to an increase in the incidence of uncomplicated illness in some age groups might lead to a reduction in mortality or severe disease. We know that parasite tolerance and anti-parasitic immunity have different dynamics, and may conjecture that they make differential contributions to uncomplicated and severe disease respectively.

2.A References

1. Breman JG. The ears of the hippopotamus: manifestations, determinants, and estimates of the malaria burden. *Am J Trop Med Hyg* 2001, 64: 1-11

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

3. Trape JF, Rogier C, Konate L, Diagne N, Bouganali H, Canque B, Legros F, Badji A, Ndiaye G, Ndiaye P. The Dielmo project: a longitudinal study of natural malaria infection and the mechanisms of protective immunity in a community living in a holoendemic area of Senegal. *Am J Trop Med Hyg* 1994, 51: 123-137

4. Konate L, Diagne N, Brahimi K, Faye O, Legros F, Rogier C, Petrarca V, Trape JF. [Biology of the vectors and transmission of *Plasmodium falciparum*, *P. malariae* and *P. ovale* in a village in the savanna of west Africa (Dielmo, Senegal)]. *Parasite* 1994, 1: 325-333

5. Fontenille D, Lochouarn L, Diatta M, Sokhna C, Dia I, Diagne N, Lemasson JJ, Ba K, Tall A, Rogier C, Trape JF. Four years' entomological study of the transmission of seasonal malaria in Senegal and the bionomics of *Anopheles gambiae* and *A. arabiensis*. *Trans R Soc Trop Med Hyg* 1997, 91: 647-652

6. Kitua A, Smith T, Alonso PL, Masanja H, Urassa H, Menendez C, Kimario J, Tanner M. *Plasmodium falciparum* malaria in the first year of life in an area of intense and perennial transmission. *Trop Med Int Health* 1996, 1: 475-484

7. Mwangi TW, Ross A, Snow RW, Marsh K. Case definitions of clinical malaria under different transmission conditions in Kilifi district, Kenya. *J Infect Dis* 2005, 191: 1932-1939

8. Lines J, Armstrong-Schellenberg JR. For a few parasites more: inoculum size, vector control and strain-specific immunity to malaria. *Parasitol Today* 1992, 8: 381-382

9. Contamin H, Fandeur T, Rogier C, Bonnefoy S, Konate L, Trape JF, Mercereau-Puijalon O. Different genetic characteristics of *Plasmodium falciparum* isolates collected during successive clinical malaria episodes in Senegalese children. *Am J Trop Med Hyg* 1996, 54: 632-643

10. Gupta S, Day KP. A theoretical framework for the immunoepidemiology of *Plasmodium falciparum* malaria. *Parasite Immunol* 1994, 16: 361-370

11. Gupta S, Trenholme K, Anderson RM, Day KP. Antigenic diversity and the transmission dynamics of *Plasmodium falciparum. Science* 1994, 263: 961-963

12. Babiker H, Ranford-Cartwright L, Walliker D. Genetic structure and dynamics of Plasmodium falciparum infections in the Kilombero region of Tanzania. *Trans R Soc Trop Med Hyg* 1999, 93 Suppl 1: 11-14

13. Ranford-Cartwright L, Balfe P, Carter R, Walliker D. Frequency of cross-fertilization in the human malaria parasite *Plasmodium falciparum*. *Parasitol* 1993, 107: 11-18

14. Walliker D. Genetic recombination in malaria parasites. *Exp Parasitol* 1989, 69: 303-309

15. Conway D, Roper C, Oduola AM, Arnot D, Kremsner PG, Grobusch MP, Curtis CF, Greenwood BM. High recombination rate in natural populations of *Plasmodium falciparum*. *Proc Natl Acad Sci USA* 1999, 96: 4506-4511

16. Felger I, Beck H-P. Genotyping of *Plasmodium falciparum*. PCR-RFLP analysis. *Methods Mol Med* 2002, 72: 117-129

17. Alloueche A, Silveira H, Conway D, Bojang K, Doherty T, Cohen J, Pinder M, Greenwood BM. High-throughput sequence typing of T-cell epitope polymorphisms in *Plasmodium falciparum* circumsporozoite protein. *Mol Biochem Parasitol* 2000, 106: 273-282

18. Bull P, Marsh K. The role of antibodies to *Plasmodium falciparum*-infected-erythrocyte surface antigens in naturally acquired immunity to malaria. *Trends Microbiol* 2002, 10: 55-58

19. Giha H, Staalsoe T, Dodoo D, Elhassan IM, Roper C, Satti G, Arnot D, Theander TG, Hviid L. Nine-year longitudinal study of antibodies to variant antigens on the surface of *Plasmodium falciparum*-infected erythrocytes. *Infect Immun* 1999, 67: 4092-4098

20. Recker M, Nee S, Bull P, Kinyanjui S, Marsh K, Newbold C, Gupta S. Transient cross-reactive immune responses can orchestrate antigenic variation in malaria. *Nature* 2004, 429: 555-558

21. Saul A. Transmission dynamics of *Plasmodium falciparum*. *Parasitol Today* 1996, 12:74-79

22. Smith T, Hurt N, Teuscher T, Tanner M. Is fever a good sign for clinical malaria in surveys of endemic communities? *Am J Trop Med Hyg* 1995, 52: 306-310

23. McGuinness D, Koram K, Bennett S, Wagner G, Nkrumah F, Riley E. Clinical case definitions for malaria: clinical malaria associated with very low parasite densities in African infants. *Trans R Soc Trop Med Hyg* 1998, 92: 527-531

24. Rogier C, Commenges D, Trape JF. Evidence for an age-dependent pyrogenic threshold of *Plasmodium falciparum* parasitemia in highly endemic populations. *Am J Trop Med Hyg* 1996, 54: 613-619

25. Vounatsou P, Smith T, Kitua A, Alonso PL, Tanner M. Apparent tolerance of *Plasmodium falciparum* in infants in a highly endemic area. *Parasitol* 2000, 120: 1-9

26. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 19-31

27. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationship between the entomologic inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 11-18

28. Jeffery GM, Young MD, Burgess RW, Eyles DE. Early activity in sporozoite-induced *Plasmodium falciparum* infections. *Ann Trop Med Parasitol* 1959, 53: 51-58

29. Smith T, Genton B, Baea K, Gibson N, Taime J, Narara A, Al-Yaman F, Beck H-P, Hii J, Alpers MP. Relationships between *Plasmodium falciparum* infection and morbidity in a highly endemic area. *Parasitol* 1994, 109: 539-549

30. Boisier P, Jambou R, Raharimalala L, Roux J. Relationship between parasite density and fever risk in a community exposed to a low level of malaria transmission in Madagascar highlands. *Am J Trop Med Hyg* 2002, 67: 137-140

31. Molineaux L, Trauble M, Collins WE, Jeffery GM, Dietz K. Malaria therapy reinoculation data suggest individual variation of an innate immune response and independent acquisition of antiparasitic and antitoxic immunities. *Trans R Soc Trop Med Hyg* 2002, 96: 205-209

32. Miller MJ. Observations on the natural history of malaria in the semi-resistant West African. *Trans R Soc Trop Med Hyg* 1958, 52: 152-168

33. Trape JF, Peelman P, Morault-Peelman B. Criteria for diagnosing clinical malaria among a semi-immune population exposed to intense and perennial transmission. *Trans R Soc Trop Med Hyg* 1985, 79: 435-442

34. Clark IA, Al-Yaman F, Cowden WB, Rockett KA. Does malarial tolerance, through nitric oxide, explain the low incidence of autoimmune disease in tropical Africa? *Lancet* 1996, 348: 1492-1494

35. Boutlis CS, Gowda DC, Naik RS, Maguire GP, Mgone CS, Bockarie M, Lagog M, Ibam E, Lorry K, Anstey NM. Antibodies to *Plasmodium falciparum* glycosylphosphatidylinositols: inverse association with tolerance of parasitemia in Papua New Guinean children and adults. *Infect Immun* 2002, 70: 5052-5057

36. Charlwood JD, Smith T, Lyimo E, Kitua A, Masanja H, Booth M, Alonso PL, Tanner M. Incidence of *Plasmodium falciparum* infection in infants in relation to exposure to sporozoite-infected anophelines. *Am J Trop Med Hyg* 1998; 59: 243-251

37. INDEPTH Network. Population, Health and Survival at INDEPTH Sites. Ottawa: IDRC, 2002.

38. Rogier C, Trape JF. [Study of premunition development in holo- and meso-endemic malaria areas in Dielmo and Ndiop (Senegal): preliminary results, 1990-1994]. *Med Trop (Mars)* 1995, 55: 71-76

39. Kirkpatrick S, Gelatt CDJr, Vecchi MP. Optimization by simulated annealing. *Science* 1983, 220: 671-680

40. Press WH, Flannery BP, Teukolsky SA, Vetterling WT. Numerical recipes in C: the art of scientific computing. Cambridge: Cambridge University Press, 1988

41. Smith T, Killeen G, Lengeler C, Tanner M. Relationships between the outcome of *Plasmodium falciparum* infection and the intensity of transmission in Africa. *Am J Trop Med Hyg* 2004, 71 (2 Suppl): 80-86

42. Cox MJ, Kum DE, Tavul L, Narara A, Raiko A, Baisor M, Alpers MP, Medley GF, Day KP. Dynamics of malaria parasitaemia associated with febrile illness in children from a rural area of Madang, Papua New Guinea. *Trans R Soc Trop Med Hyg* 1994, 88: 191-197

43. Woolhouse ME. Patterns in parasite epidemiology: The peak shift. *Parasitol Today* 1998, 14: 428-434

44. Smith T, Hii J, Genton B, Muller I, Booth M, Gibson N, Narara A, Alpers MP. Associations of peak shifts in age-prevalence for human malarias with bed net coverage. *Trans R Soc Trop Med Hyg* 2001, 95: 7-13

45. Coleman PG, Perry BD, Woolhouse ME. Endemic stability--a veterinary idea applied to human public health. *Lancet* 2001; 357: 1284-1286.

Chapter 3

An epidemiological model of severe morbidity and mortality caused by *Plasmodium falciparum*

Amanda Ross, Nicolas Maire, Louis Molineaux and Thomas Smith

Swiss Tropical Institute, Basel, Switzerland
retired, formerly WHO, Geneva, Switzerland

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3.1 Summary

The intensity of *Plasmodium falciparum* transmission has multifarious and sometimes counter-intuitive effects on age-specific rates of severe morbidity and mortality in endemic areas. This has led to conflicting speculations about the likely impact of malaria control interventions. We propose a quantitative framework to reconcile the various apparently contradictory observations relating morbidity and mortality rates to malaria transmission. Our model considers two sub-categories of severe malaria episodes. These comprise episodes with extremely high parasite densities in hosts with little previous exposure, and acute malaria episodes accompanied by co-morbidity or other risk factors enhancing susceptibility. In addition to direct malaria mortality from severe malaria episodes, the model also considers the enhanced risk of indirect mortality following acute episodes accompanied by co-morbidity after the parasites have been cleared. We fit this model to summaries of field data from endemic areas of Africa, and show that it can account for the observed age- and exposure-specific patterns of paediatric severe malaria and malaria-associated mortality in children. This model will allow us to make predictions of the long-term impact of potential malaria interventions. Predictions for children will be more reliable than those for older people because there is a paucity of epidemiological studies of adults and adolescents.

3.2 Introduction

The outcomes of *Plasmodium falciparum* infections range from self-limiting asymptomatic parasitaemia to rapid death. It is not well understood why some infections have much worse consequences than others[1] and this makes it difficult to predict the epidemiological effects of malaria interventions.

Different outcomes have different age patterns: the more severe the outcome, the younger the age group most affected. The age-pattern of each outcome also varies with the intensity of transmission.[2] In stable endemic areas, the incidence of clinical malaria episodes is highest at intermediate levels of transmission.[3] Hospital-diagnosed severe malaria in children also appears to be most frequent at intermediate transmission,[3] but in infants shows an increase with transmission intensity,[4] as does all cause mortality.[5] Hospital case fatality rates are age-dependent, with the highest rates in young infants and older children and a minimum in an intermediate age group.[6,7] Malaria-specific mortality rates might therefore be expected to show different relationships with age and transmission intensity than do morbidity rates. Community-based estimates of malaria-specific mortality rates have been estimated using verbal autopsies for a number of endemic areas but the relationships with transmission intensity are unclear. One reason may be that verbal autopsies have poor sensitivity and specificity for malaria.[8,9]

The risk of malaria-diagnosable morbidity and mortality is thought to depend on other risk factors such as malnutrition and co-infections. It has been suggested that approximately 60% of malaria mortality is attributable to low weight, vitamin A deficiency and/or zinc deficiency.[10] Eight percent of severe malaria cases in Kenya were found to be bacteraemic.[11] In addition to causing direct malaria mortality, *P. falciparum* is likely to be a contributory factor in many deaths that would not be diagnosed as malaria by a physician.[12-14] Many malaria control or local elimination programs decreased all-cause mortality by more than the initial estimates of malaria specific mortality.[15-19] The differential mortality required to explain frequencies of sickle cell hemoglobin (HbAS) is substantially greater than that generally attributed to malaria alone.[14,16,20] However, the relative contribution of this indirect mortality has been debated.[2,21]

We propose a model to explain these patterns as consequences of two processes with different relationships to host age and the level of malaria transmission. The first of these is the level of immunity to asexual blood stages of the malaria parasite. The second is the chance that the host defences are compromised by some co-morbidity or enhanced susceptibility around the time of the clinical malaria attack.

To predict the long-term impact of potential interventions on *P. falciparum* malaria, there is a need for dynamic models linking severe and fatal malaria to transmission.[14] We now incorporate our proposal for the causes of severe malaria and malaria attributable mortality into a simulation model of malaria transmission, parasitaemia and acute morbidity. We fit the model to published data and show that the apparently conflicting observations relating morbidity and mortality rates to malaria transmission can be reconciled within a coherent framework that corresponds to current knowledge of malaria biology.

3.3 Methods

3.3.1 Model

Severe malaria episodes We consider severe malaria episodes as those events that would have led to an admission diagnosis of severe malaria, had the patient presented to a health facility. The probability that a clinical malaria episode occurs in individual i at time t, $P_m(i,t)$, depends on both the simulated parasite density, $Y(i,t)$, and the modelled pyrogenic threshold $Y^*(i,t)$ (Chapter 2). These episodes (A, Figure 3.1) include a subset that are severe (B, Figure 3.1).

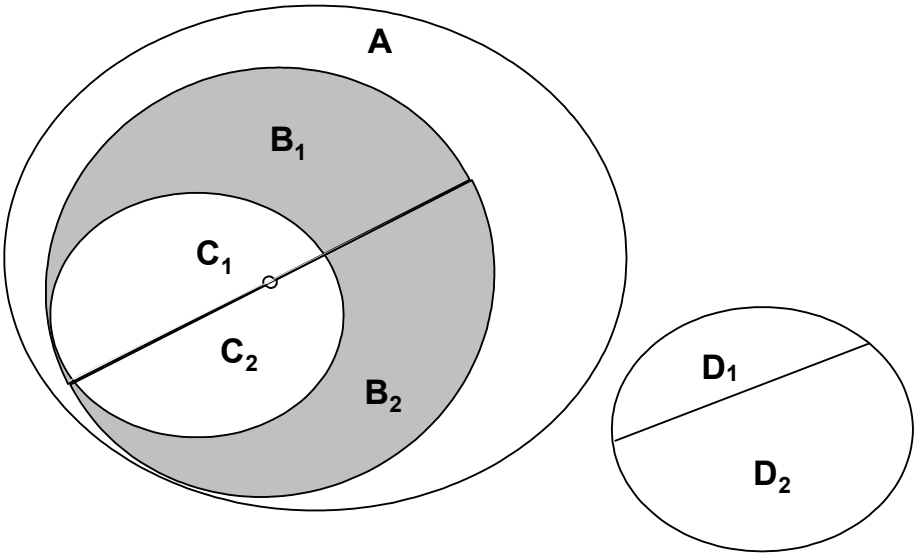


Figure 3.1 Classes of malaria morbidity and mortality

A: All clinical malaria episodes. **B**: Severe malaria. Episodes in class B_1 arise because of hyper-parasitaemia; those in class B_2 because of co-morbidity or enhanced susceptibility. **C**: Direct malaria mortality. Deaths in classes C_1 and C_2 arise from severe malaria episodes B_1 and B_2.

D: Indirect malaria mortality. These are deaths that would not be diagnosed as malaria deaths but would not have occurred without malaria exposure. D_1 represents deaths resulting from pre-natal exposure of the mother; D_2 represents subsequent deaths where an acute malaria episode is a contributing factor in conjunction with non-malaria morbidity

We propose that severe malaria episodes can occur as a result of one or other of two distinct processes (B_1 and B_2). These categories do not necessarily correspond to any of the specific syndromes of severe malaria.

One subset of the severe malaria episodes (B_1, Figure 3.1) comprises those that occur when the host experiences an overwhelming parasite density. We define $\mathrm{H}(i,t)$ to be the clinical status of individual i at time t, $P_{B_1}(i,t)$ to be the probability that a clinical malaria episode in individual i at time t is severe as a result of this process, and specify this probability using

$$P_{B_1}(i,t) = \Pr(\mathrm{H}(i,t) \in \mathrm{B}_1 | \mathrm{H}(i,t) \in \mathrm{A}) = \frac{Y_{max}(i,t)}{Y^*_{B_1}(i,t) + Y_{max}(i,t)} \qquad (3.1)$$

where $\in$ indicates membership of a set, P indicates probability, and $|$ is the symbol for conditional probability. $Y^*_{B_1}(i,t)$ is then a critical value and $Y_{max}(i,t)$ is the simulated maximum of daily parasite density measurements in individual i at 5-day time interval t. We evaluate the fit of two possible algorithms for $Y^*_{B_1}(i,t)$:

(i) We propose that the parasite density required to cause a severe episode is some multiple, α_s, of that required to cause an uncomplicated episode in the same individual at the same timepoint, i.e. $Y^*_{B_1}(i,t) = \alpha_s Y^*(i,t)$, where $Y^*(i,t)$ is the previously defined pyrogenic threshold for individual *i* at time *t* (Chapter 2).

(ii) Whereas the concept of a pyrogenic threshold for uncomplicated clinical episodes of malaria is widely accepted, the pathogenesis of severe malaria may differ from that of uncomplicated episodes, so it is not obvious that the critical level of parasitaemia for severe malaria is related to $Y^*(i,t)$. We therefore also considered a model in which severe episodes of class B_1 arise when a single host- and exposure-independent critical parasite density is exceeded, i.e. $Y^*_{B_1}$ is a constant over all individuals and time points.

The second, non-intersecting, subset of severe malaria episodes (B_2) occur when an otherwise uncomplicated malaria episode happens to coincide with some other insult (e.g. a bacterial infection, malnutrition or anaemia[2]), which occurs with risk $F(a(i,t))$, a function of the age $a(i,t)$ of individual *i* at time *t*. We considered three proposals for the age profile of these non-malaria insults (Appendix 3.A).

We assume that, conditional on the age of the host, the risk of an acute malaria attack is independent of the risk of such an insult, but that the risk of severe malaria does depend on $F(a(i,t))$. The probability that an episode belonging to class B_2 occurs at time *t*, conditional on there being a clinical episode at that time is $P_{B_2}(i,t)$ defined as

$$P_{B_2}(i,t) = \Pr(\mathrm{H}(i,t) \in \mathrm{B}_2 \mid \mathrm{H}(i,t) \in \mathrm{A}) \qquad (3.2)$$

and calculated as

$$P_{B_2}(i,t) = F(a(i,t)). \qquad (3.3)$$

The age and time specific risk of severe malaria morbidity conditional on a clinical episode is then given by

$$P_B(i,t) = P_{B_1}(i,t) + P_{B_2}(i,t) - P_{B_1}(i,t)P_{B_2}(i,t) \qquad (3.4)$$

The term $P_{B_1}(i,t)P_{B_2}(i,t)$ is subtracted to avoid double-counting of that small proportion of episodes that qualify as severe by both definitions. Thus the unconditional risk of a severe malaria episode is $P_B(i,t)P_m(i,t)$ where $P_m(i,t)$ is the probability of a clinical episode (Chapter 2).

Direct malaria mortality We refer to deaths resulting from episodes of either class B_1 or B_2 as direct malaria mortality (classes C_1 and C_2 in Figure 3.1). We assume that 48% of severe malaria episodes present to hospital (Appendix 3.B) and that this applies equally to both class B_1 and class B_2 episodes. Age-specific hospital case fatality rates were

taken from those reported from Tanzania.[7] We assume that these hospital case fatality rates remain the same, even if the case mix of type B_1 and B_2 severe malaria episodes varies between transmission settings.

The mortality risk for a severe episode at age a in the community $Q_c(a)$, is estimated with

$$Q_c(a) = \frac{Q_h(a)\varphi_1}{1 - Q_h(a) + Q_h(a)\varphi_1}, \qquad (3.5)$$

where $Q_h(a)$ is the reported hospital case fatality rate at age a, and φ_1 is the estimated odds ratio for death in the community compared to death in inpatients. Malaria mortality is then predicted by the sum of the hospital and community malaria deaths. We estimate φ_1 by fitting to malaria-specific mortality rates.

Indirect malaria mortality. In addition to the direct malaria mortality C_1 and C_2 we need to model additional, indirect, malaria deaths in order to replicate the association between all-cause mortality and the entomological inoculation rate (EIR), specifically in infants. We define as indirect malaria deaths those that would not have occurred in the absence of prior malaria exposure, but where the terminal illness would not have been diagnosed as malaria by a competent physician. We do not classify deaths in class C_2 as indirect mortality because we consider a death in the same five-day interval as a precipitating clinical malaria episode to be diagnosable as malaria.

In the cases of indirect deaths, we propose that malaria exposure acts to enfeeble the individual, leading to subsequent mortality. These deaths would be prevented if malaria was removed, and so should be included in predictions of the potential impact of malaria interventions.

We consider two distinct classes of indirect mortality (Figure 3.1). D_1 comprises neonatal mortality resulting from maternal infection during pregnancy. The model we use to predict the incidence of such deaths is considered in Chapter 4. D_2 comprises post-neonatal indirect mortality that is provoked by an acute attack of malaria, which together with other co-morbidity or enhanced susceptibility leads to subsequent death. The insults contributing to a death in class D_2 could be sequential or they could occur together. Since this makes little difference to the predicted incidence, for mathematical convenience we use a model analogous to that for severe malaria in class B_2. In this model an event in class D_2 is instigated at time *t*, conditional on there being a clinical episode at that time, with probability $P_{D_2}(i,t)$ defined as

$$P_{D_2}(i,t) = \Pr(\mathrm{H}(i,t) \in \mathrm{D}_2 \mid \mathrm{H}(i,t) \in \mathrm{A}) \qquad (3.6)$$

and calculated as

$$P_{D_2}(i,t) = \frac{Q_D}{1+\left(\frac{a(i,t)}{a_F^*}\right)} \qquad (3.7)$$

where Q_D is the limiting value of $P_{D_2}(i,t)$ at birth. The deaths in class D_2 are simulated as occurring 30 days after time t. This allows for the possibility that the host dies of an event in class C_1 or C_2 before the indirect death occurs.

3.3.2 Data and fitting of the model

Severe morbidity. Data on the relative incidence of severe malaria in children across different transmission intensities have been collated by Marsh and Snow.[6] They summarize the relationship between severe malaria hospital admission rates and *P. falciparum* prevalence in children less than nine years of age. To obtain a continuous function relating hospital incidence to prevalence, we linearly interpolated between data points. To convert the hospital incidence rates to community severe malaria incidence, we divided the hospital admission rates by the assumed proportion (48%) of severe episodes presenting to hospital (Appendix 3.B). To fit our model to this relationship we ran our simulation model of *P. falciparum* incidence, parasitology and clinical episodes, and assumed one of the models for severe malaria described earlier in this chapter, with the published transmission patterns for all the sites in Table 3.1 as input. We compared the predicted absolute incidence of severe malaria with the value on the interpolated curve corresponding to the predicted prevalence for the simulated site.

More detailed age-specific severe malaria hospital admission rates are published for five of these sites which have varying transmission intensities, together with the parasite prevalence in children aged 1-9 years.[4] We summarized the patterns of incidence by age in 1-4, and 5-9 year-old children, compared to 1-11 month-old infants by calculating the relative risks. To fit our model to these data, we chose sites to represent the transmission settings on the basis of their predicted prevalence. Four sites were chosen, a fifth could not be matched to the very low transmission setting with 2% prevalence (Bakau, The Gambia).

Table 3.1 Sites used for fitting the model for the incidence of severe malaria*

	Entomology reference	EIR data Year	EIR
Burkina Faso			
ITC control	[22,23]	1994-95	389
Karangasso	[24]	1985	244
Kongodjan	[25]	1984	133
Ziniare	[26]	1994-95	70
Burundi			
Gihanga	[27]	1983	205
Katumba	[27]	1982	13.6
Kenya			
Chonyi	[28,29]	1992-93	50
Kilifi North	[28,29]	1992-93	10.5
Kilifi Town	[28]	1990-91	2.8
Saradidi	[30]	1986-87	239
Senegal			
Bandafassi	[31,32]	1995-96	363
Mlomp	[31,32]	1995	30
Niakhar	[33]	1995	11.6
Tanzania			
Namawala	[34]	1990-91	329
Yombo	[35]	1992	234
The Gambia			
Areas I-V	[36,37]	1991	†
Farafenni	[36]	1987	8.9
Others			
Bo, Sierra Leone	[38]	1990-91	34.7
Ganvie, Benin	[39]	1993-95	11
Manhica,Mozambique	[40]	2001-02	38
Matsari, Nigeria	[20]	1971	68
Navrongo, Ghana	[41]	2001-02	418

*EIR = entomological inoculation rate; ITC = control group of randomized trial of insecticide-treated nets. †Five sites with annual EIR between 1 and 10.

For both sets of sites we simulated the incidence of severe malaria using a version of the model without effects of treatment of uncomplicated malaria episodes or any malaria mortality. The simulated population was stable in size, and the age-distribution was fixed to be approximately the same as Ifakara, Tanzania using the algorithm reported elsewhere.[40]

We simultaneously fitted our models to both the absolute incidence of severe malaria in children less than nine years of age and the age-specific relative risks by weighted least squares of the log-transformed rates, where the weights were chosen so that the two analyses were weighted approximately equally.

Simulated annealing[42,43] was used to identify the parameter values that minimized the weighted residual sum of squares. Approximate confidence intervals were obtained by estimating the Fisher information for the parameters from a least squares fit of local quadratic approximations to the (stochastic) log likelihood. In addition to considering the formal model fit, we also assessed the biological plausibility of the models and the predictions for the age groups for which we had no data.

Direct malaria mortality The odds ratio for death of a case in the community relative to that in hospital, φ_1, was estimated by fitting the malaria-specific mortality rates in children less than five years of age predicted by the severe malaria models above assuming the published hospital case fatality rate. The data were derived from verbal autopsy (VA) studies in sites with prospective demographic surveillance and were adjusted for the effect of malaria transmission intensity on the sensitivity and specificity of the cause of death determination.[44] Sites with both VA data and seasonal patterns of the EIR (Table 3.2) were used for estimating φ_1. The fitting algorithm was the same as for the severe malaria model and we assume that there was no effective treatment of uncomplicated malaria episodes.

Indirect malaria mortality The deaths in class C predicted by the model could not account for the relationship observed between EIR and infant mortality,[5] and we propose that the difference is due to deaths in class D_2. We assembled a library of sites for which entomological data was collected at least monthly and all-cause infant mortality rates (IMR) were available (Table 3.2). We use the entomological data as input and estimate Q_D and the infant mortality that is independent of malaria Q_n by the same fitting algorithm that was used for the severe malaria and direct mortality components. The model for indirect mortality is conditional on our models for severe malaria and direct malaria mortality, and assumes no effective treatment of uncomplicated malaria episodes.

Table 3.2. Sites used for fitting the model for direct and indirect malaria mortality

Site, Country	Estimated malaria mortality (< 5 year old)[44]*		All-cause IMR†		
	Year	Deaths/1000 person years	Ref for IMR	Year	IMR/1000 livebirths
Area V, The Gambia	-		[45]	1992	83
Farafenni, The Gambia	1988-90	9.4	-		
Kilifi North	1991-93	9.2	-		
Bo, Sierra Leone	1990	12.8	[46]	1990	74
Karangasso, Burkina Faso	-		[47]	1986-88	121
Yombo, Tanzania	1992-94	22.1	[35]	1992-94	131
Niakhar, Senegal	1990-95	10.9	[48,49]	1995-99	80
Manhica, Mozambique	-		[49]		78.5
Navrongo, Ghana	1990	9.3	[49]		111.9
Bandafassi, Senegal	1984-89	5.9	[49]		124.9
Mlomp, Senegal	-		[32]	1995	61
Saradidi, Kenya	1997	20.8	[50]	1981-82	109.4
Kongodjan, Burkina Faso	1982-86	2.2	-		
Namawala, Tanzania	-		[51]	1997	95.2

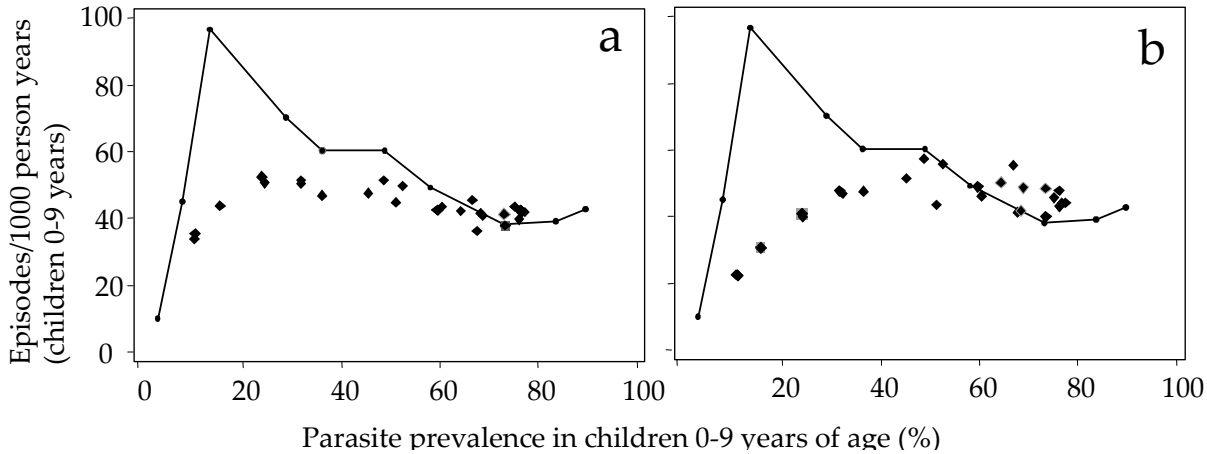
* Used for direct malaria mortality model. † Used for indirect malaria mortality model. IMR = all-cause infant mortality rate /1000 livebirths

Since a study found no clear relationship between all-cause mortality for children aged 1-4 years and transmission intensity,[5] we did not use data for children over one year of age to estimate the parameters of the model for indirect mortality.

3.4 Results

Severe malaria We compared the best-fitting models of the two forms proposed (Table 3.3). Both models produced similar predictions (Figure 3.2). Both gave a good fit to most of the data, and in particular they reproduced the decrease in incidence with transmission intensity in highly endemic areas.

Figure 3.2 Model predictions of the incidence of severe disease compared with observed data



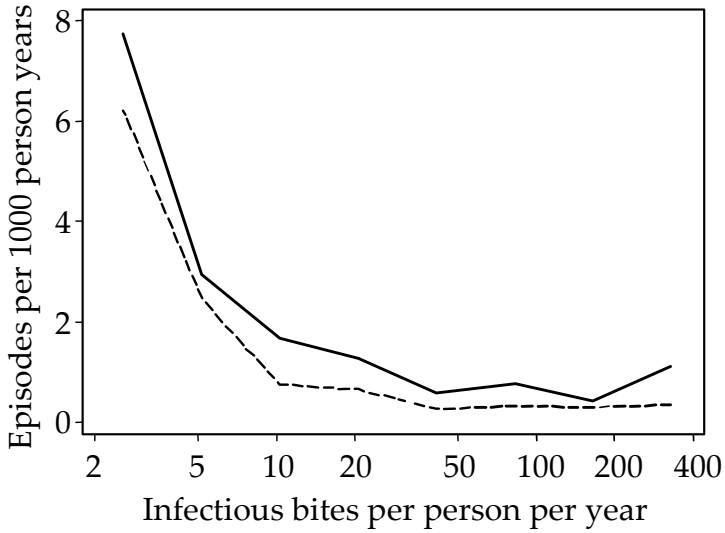
♦ a. Model 1; b Model 2. —●—: data reported by Marsh and Snow.[6] The hospital incidence rates have been divided by 0.48 to provide estimates of the incidence in the community.

Neither model reproduced the sharp peak in incidence associated with a prevalence of just under 20%, which is most pronounced in the rate reported from a hospital in Ethiopia. Model 1, with the severe malaria threshold as a multiple of the individual's pyrogenic threshold, had a better fit (weighted residual sum of squares 3.35 versus 8.97). However, it predicted a rather high incidence of severe episodes in adults (for whom few data are available[52]) (Figure 3.3) and this in turn lead to estimates of malaria mortality rates that exceed recorded all-cause mortality rates in some age groups.[49]

Table 3.3. Parameter estimates with 95% confidence intervals

		Model 1		Model 2
Severe malaria model parameters				
Threshold density multiplier (Model 1); Threshold (Model 2)	α_s	983 (518, 1869)	$Y^*_{B_1}$	784,000 (530,000, 1162,000)
Prevalence at birth of co-morbidity contributing to severe episodes	F_0	0.127 (0.060, 0.258)	F_0	0.092 (0.074, 0.127)
Critical age for co-morbidity (years)	a^*_F	0.078 (0.053, 0.113)	a^*_F	0.117 (0.086, 0.160)
Weighted residual sum of squares		3.35		8.97
Malaria mortality model parameters				
Odds ratio for case fatality in the community compared to in hospital	φ_1	2.00 (1.33, 5.26)	φ_1	2.09 (1.31, 7.63)
Weighted residual sum of squares		3.24		3.68
Indirect malaria mortality model parameters				
Non-malaria intercept for infant mortality rate	Q_n	58.2 (30.5, 111.0)	Q_n	49.5 (33.3, 73.8)
Prevalence at birth of co-morbidity contributing to indirect mortality	Q_D	0.018 (0.006, 0.047)	Q_D	0.019 (0.010, 0.037)
Weighted residual sum of squares		0.31		0.33

Figure 3.3 Predicted incidence of severe malaria in adults 20-39 years of age by transmission intensity

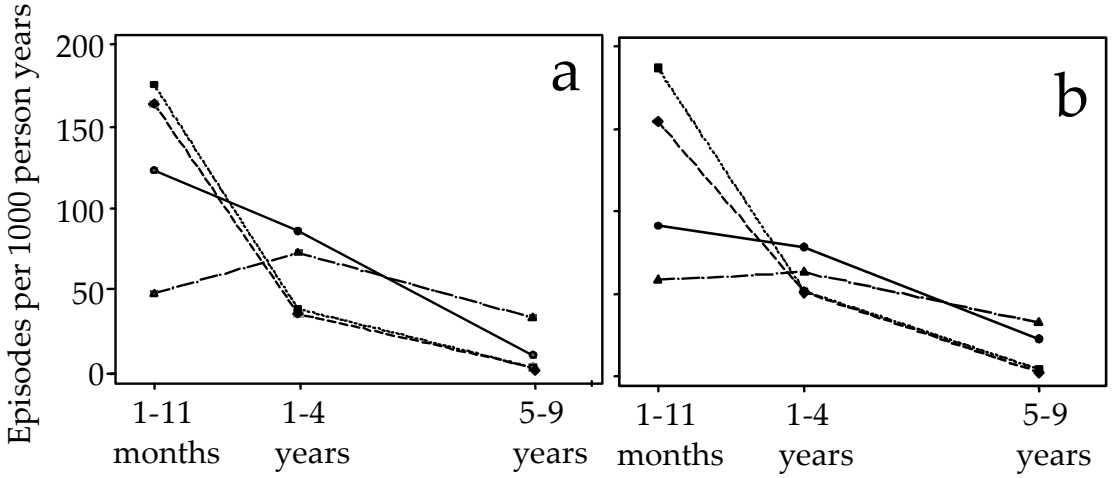


——: Model 1; ---- Model 2**;** The seasonality of transmission follows that of Namawala in Tanzania,[34] scaled to sum to different values of infectious bites per person per year.

Predicted mortality rates in older age-groups were lower with model 2. The assumption of a constant parasitaemia threshold for severe malaria in model 2 is also more attractive because of the evidence that total parasite biomass is critical in precipitating severe malaria episodes.[53] The estimate of this threshold of 784,000 parasites/μl is high, but within the range observed in severe malaria patients. We do not attach much credibility to the precise value of this threshold since our simulation model only reproduces distributions of parasite densities very approximately.[40]

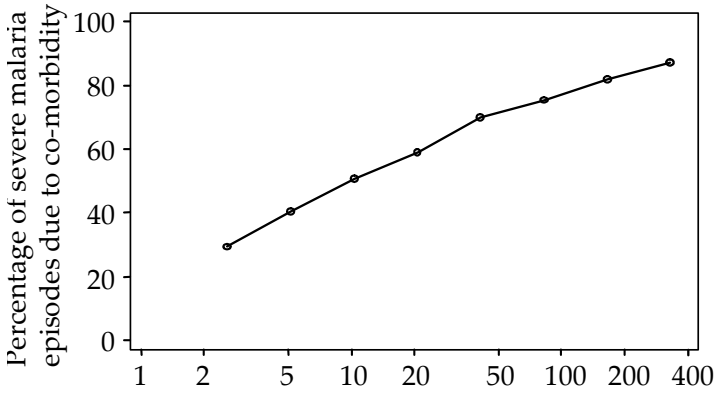
Model 2 reproduces the age-patterns from the four sites with different transmission intensities reasonably well (Figure 3.4). The proportion of predicted severe malaria cases that belong to class B_2 in this model increases with transmission intensity because the infections tend to occur at younger ages (Figure 3.5).

Figure 3.4 Age-specific incidence of severe malaria



a. Community incidence rates calculated from the hospital data reported by Snow and others[4] by dividing by the notional hospital attendance rate of 0.48. **b**. Predicted incidence rates from model 2 for the four scenarios chosen on the basis of similar parasite prevalence values (1-9 years) to the sites above.

Figure 3.5 Percentage of severe malaria episodes arising due to age-dependant cofactors (B_2) by transmission intensity



These predictions are from model 2 and include all age groups. The seasonal pattern of transmission intensity follows that of Namawala in Tanzania,[34] scaled to sum to eight different values of infectious bites per person per year.

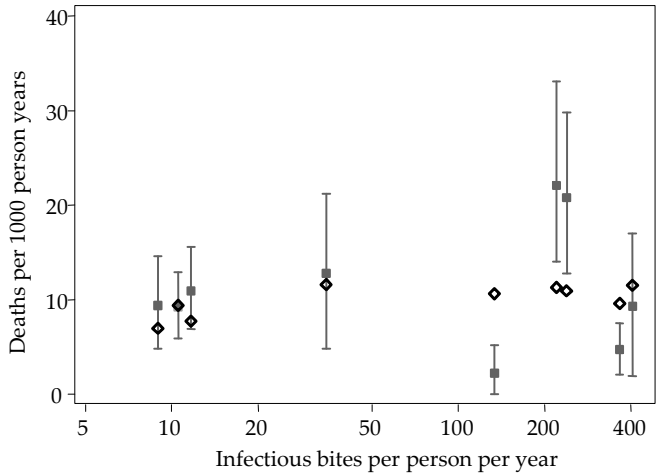
Direct malaria mortality To reconcile the field estimates of malaria-specific mortality rates with either model for severe malaria, odds ratios of approximately 2 were estimated for case fatality in the community compared with in hospital (Table 3.3). The predicted age-specific community case fatality is shown in Figure 3.6.

Figure 3.6. Case fatality rates by age

—●—: reported hospital case fatality rates[7] ------: case fatality in the community obtained using the estimate from model 2

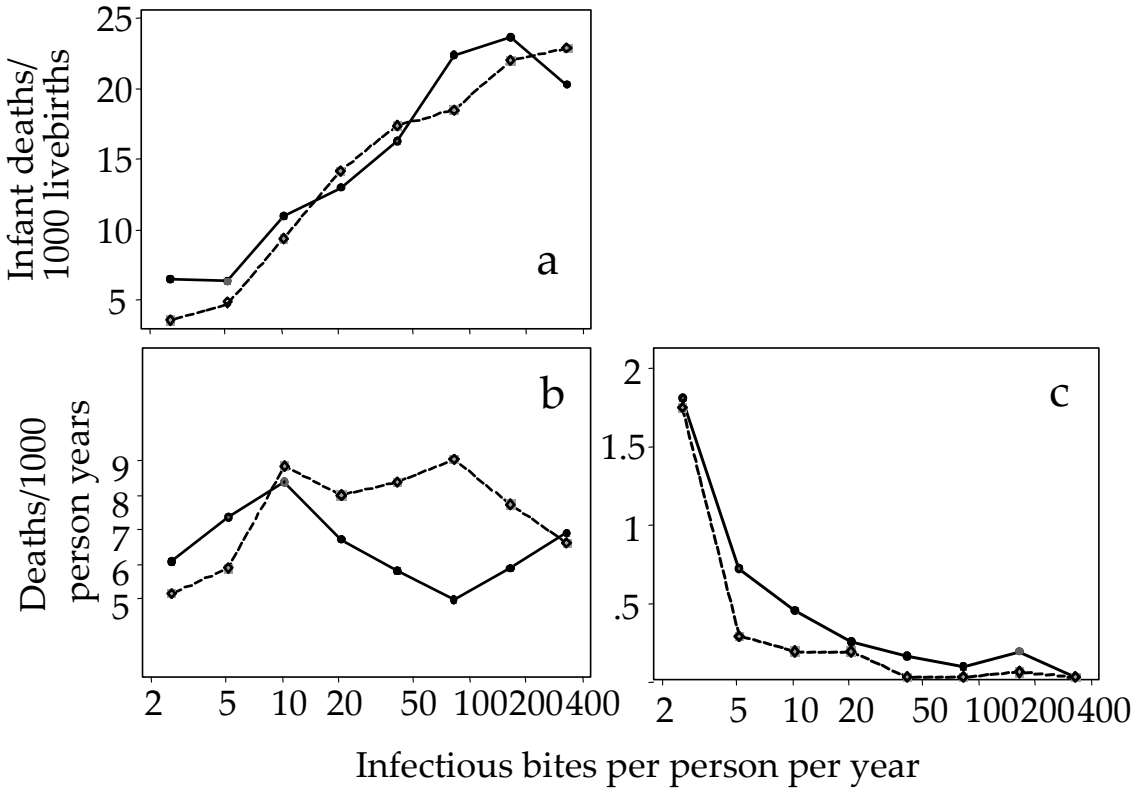
Empirical malaria mortality rates for children less than 5 years are shown in Figure 3.7, together with the predictions for the same sites using the severe malaria model that we have adopted (model 2). Both the observed data and predictions show no obvious trend with transmission intensity, and there is a large variation between the sites in the verbal autopsy-based rates. The predicted malaria mortality rates show a clear increase with transmission intensity in infants and no apparent trend for 1-4 year-old children for both models (Figure 3.8a and b). Using the severe malaria model with a multiplier for the pyrogenic threshold (model 1), adults 20-39 years of age had a rather high predicted malaria mortality rate (Figure 3.8c). This is a result of the high predictions for incidence of severe malaria with this model.

Figure 3.7. Direct malaria mortality in children under five years of age



■ Observed malaria-specific mortality in children under 5 years[44] (error bars are 95% confidence intervals); ◊ Model predictions. Predictions and observed data for the same sites are vertically aligned since they have the same transmission intensity.

Figure 3.8. Predicted malaria-specific mortality rates by transmission intensity

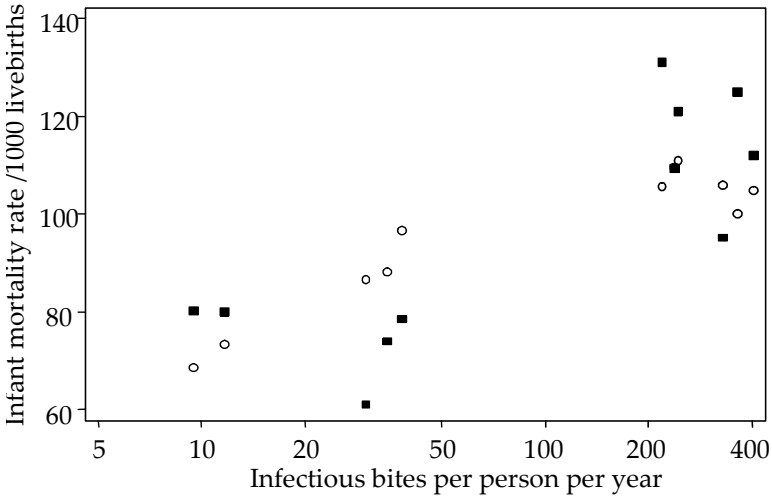


a. infants; b. children 1 to 4 years of age; c. adults 20-39 years of age.
──: model 1; ---- model 2. The seasonal pattern of transmission intensity follows that of Namawala in Tanzania,[34] scaled to sum to different values of infectious bites per person per year

Indirect malaria mortality We estimate that in the absence of *P. falciparum*, the IMR for the sites included in the analysis, Q_n, would average about 50 per 1000 live-births (Table 3.3) but this quantity was estimated very imprecisely, since the parameters Q_D and Q_n are highly correlated**.**

There was an association between the observed all-cause IMR and transmission intensity, as previously reported using broader inclusion criteria[5] (Figure 3.9). The predicted IMR for these sites using model 2 (incorporating the effects of severe malaria and malaria mortality models as above) reproduces this apparent trend.

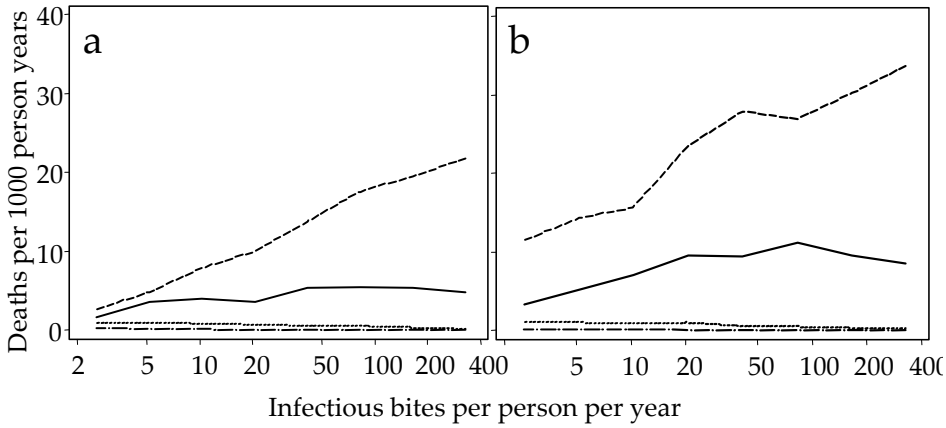
Figure 3.9. Observed and predicted infant mortality rates



■ Infant mortality rates from field data; ○ Predictions using model 2

Predictions of indirect malaria mortality for different age groups show similar patterns with transmission intensity to those of the direct malaria mortality (Figure 3.10). Although the deaths in infants tend to increase, this is not the case for either direct or indirect malaria mortality for older age groups. Taking all age groups together, the ratio of indirect: direct malaria deaths was 0.6 for an EIR of 5, this increased to 1.4 for an EIR of approximately 100 and did not increase further for higher transmission intensities.

Figure 3.10. Predicted mortality rates by transmission intensity



a. Direct malaria mortality b. Indirect malaria mortality. Age groups: ---- 0-1; ——1-4; ……5-20; — — 20-39 years. Predictions from model 2 using as input the seasonal pattern of inoculations for Namawala, Tanzania, scaled to different numbers of infectious bites per person per year.

3.5 Discussion

Our model replicates reasonably well the associations of severe malaria incidence and transmission intensity in sub-Saharan Africa. Severe episodes resulting simply from very high parasite densities (B_1 in Figure 3.1) most clearly represent malaria-specific morbidity. These are more frequent at moderate levels of transmission and account for the peak in the incidence of paediatric severe malaria at intermediate levels of transmission. Within our model, this is mainly because maternal immunity helps to control the first infections at very high levels of transmission, so that the initial infections are less well controlled if they occur later in life.[40]

The patterns of events in classes B_1 and B_2 with age and with transmission have similarities to those described for severe malaria anaemia, and for patterns for cerebral malaria[54] respectively. However, our simple structure for the different classes of events does not aim to map onto the pathophysiology of these syndromes. Recent work has suggested that the different syndromes of severe malaria are overlapping.[55] A major uncertainty lies in the choice between models 1 and 2 for the relationship between parasite density and severity of disease. This is likely to have an important effect on our predictions of the impact on interventions that affect blood stage densities, and points to a gap in our knowledge of pathogenesis. The fitting of these models suggests that a substantial proportion of severe malaria episodes involve age-dependent co-factors that are concentrated in the youngest children. This is consistent with the fact that these children have the least immunity to other infections and are also at high risk of nutritional problems.

We assumed the same age dependence in co-morbidity in estimating the contribution of malaria to indirect deaths (D_2 in Figure 3.1) and thus the effects of co-morbidity dominate those of high parasite densities in determining the impact of *P. falciparum* on all-cause mortality in the youngest children. The strong age dependence is supported by ecological comparisons of all-cause mortality rates and malaria transmission intensity, where there is no clear association after the first year of life.[5] It is also in agreement with analyses of HbAS frequencies that have suggested that indirect malaria mortality is likely to be concentrated in the youngest children.[20]

Clinical malaria episodes are also more concentrated in younger children as the transmission intensity increases (Chapter 2). Therefore, within our model, the probability that these risks coincide to cause either severe malaria episodes (B_2) or subsequent indirect mortality (D_2) increases with transmission level. We used clinical malaria episodes for the predisposing factor for indirect deaths, but it is also possible that symptomless parasitaemia plays this role.[21]

The model points to other important areas of uncertainty. Malaria in adults is an example of this. It is generally thought that severe malaria occurs only infrequently in adults in the stable endemic conditions prevailing in much of Africa[6,52] and although severe malaria is commonly diagnosed in African adults, many of these represent

misdiagnoses.[56,57] In a randomised trial, insecticide-impregnated nets did not reduce mortality in Ghanaian adults, suggesting that malaria is not a major cause of death in this age group.[58] However, immunologically naive adult visitors to endemic areas are highly susceptible and major epidemics with high case fatality may occur in areas of initially low transmission to which malaria returns after having been nearly eliminated.[59,60] A recent observational study in an endemic area of Papua New Guinea suggested that mosquito nets have a substantial effect in reducing all-cause mortality in adults in an area of moderate transmission.[61] These results suggest that malaria may be an important cause of adult mortality in areas of low endemicity.

We expect that severe malaria is infrequent in those adults with a substantial history of exposure to *P. falciparum* because they control parasite densities and thus rarely develop any acute clinical episodes. Major epidemics should not occur as a rebound if malaria control is abandoned in areas of very high previous exposure because of persistence of immunity against asexual stages of the parasite. In contrast, people who become infected with *P. falciparum* after spending most of their lives without being exposed are highly susceptible to severe episodes. Current efforts to control malaria may lead to sustained reductions in malaria transmission without eliminating the parasite, and this could place many older children and adults in this position. The shape of our function for co-morbidity is critical in our predictions of the public health burden that this implies. If co-morbidity follows the strong increase in infectious disease mortality with age that is observed in adults, then we would predict thath in low and unstable transmission settings where most adults never acquire much immunity *P. falciparum* may be an important cause of mortality in elderly people. There is a need to test whether this is the case.

There are many other factors influencing the risk and outcome of severe malaria that we have not been able to consider explicitly. These include effects of host genetic markers and of seasonality.[12] In addition, field estimates of malaria morbidity and mortality rates are unavoidably plagued by effects of attendance bias and diagnostic uncertainties. The empirical basis for estimating the effect of in-patient care on case fatality rates is particularly weak. There are estimates of four relevant quantities, the hospital case fatality rate,[6,56] the overall malaria mortality rate,[44] the proportion of malaria deaths that seek care in health facilities[62] and the per capita admission rates for severe malaria.[6] However, these do not provide a basis for convincing estimation of the case fatality rate in the community. This adds considerable uncertainty when our model is used to estimate the likely public health impact of improving curative services.[63]

In the context of recent developments in malaria control[64-66] there is a need for comparisons of the likely epidemiological impact of different intervention strategies. Randomized controlled trials provide a solid basis for predictions of the short-term impact but these cannot necessarily be extrapolated beyond the time horizon of the trial, which is rarely more than 1-2 years. Adverse consequences resulting from interference with the acquisition of natural immunity may take much longer than this

to become apparent, and the full impact of malaria interventions on human-vector transmission is also only likely to be seen over longer periods. The model we propose represents a first step towards making predictions of longer term effects that can allow for these factors.

Appendices

3.A Candidate functions for co-morbidity contributing to type B_2 severe malaria , $F(a(i,t))$

We considered three proposals for the age pattern of events that, when co-incident with a malaria attack lead to a severe episode (Figure 3.11). Infectious disease morbidity in rural African sites decreases strongly over the first few years of life so we require that, at least over this period, $F(a(i,t))$ should be a decreasing function of the age *a(i,t)* of individual *i* at time *t.*

(i) A simple proposal is an exponential decay with age

$F(a(i,t)) = \beta_1 \exp\left(-\beta_2 a(i,t)\right)$ where β_1 and β_2 are constants.

(ii) A second proposal is a hyperbolic curve

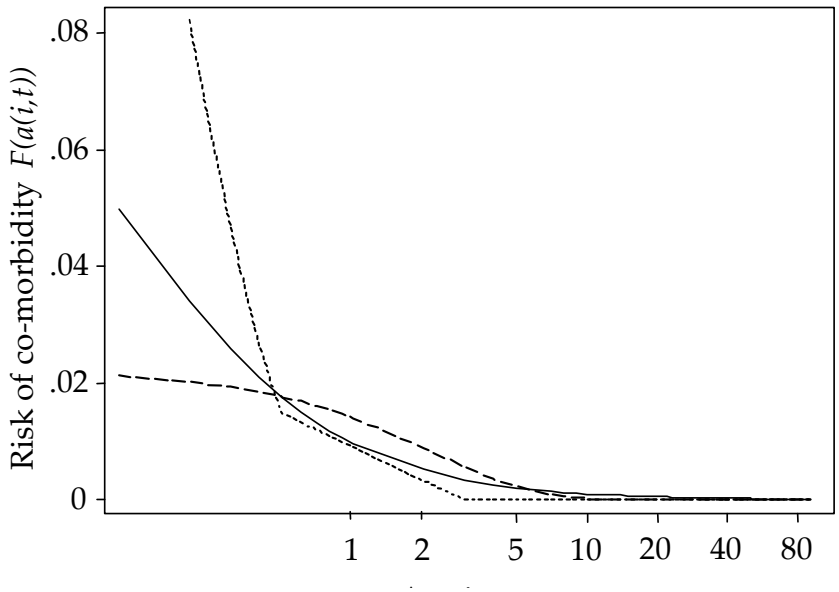
$$F(a(i,t)) = \frac{F_0}{1+\left(\frac{a(i,t)}{a_F^*}\right)} \qquad (3.8)$$

where a_F^* and F_0 are constants.

(iii) An alternative is to use an empirical function. We explored a function based on the first principal component of the life tables for demographic surveillance sites in predominantly rural communities in Africa.[49] This curve decreases with age in very young children but increases with age in adults. We expect this to represent mainly the age-pattern of infectious disease mortality (excluding that due to human immunodeficiency virus), but it is not necessarily an appropriate curve to represent the age-pattern of relevant co-morbidity. We scale the risk of an insult that would convert an uncomplicated episode to a severe attack by assuming in our model that

$$\text{logit}\left(P_{B_2}(i,t)\right) = F(a(i,t)) + \beta_3. \qquad (3.9)$$

Figure 3.11. Proposals for age-profile of co-morbidity risk for type B_2 severe malaria



– – : Negative exponential function (i)
——: Hyperbolic function (ii)
…….: Empirical mortality data (iii)

The age patterns were best reproduced using the hyperbolic curve (option ii), and thus this proposal is adopted as part of the model. The estimates for a_F^* and F_0 are given in Table 3.3. We assumed the same function for co-morbidity contributing to indirect deaths (equation 3.7). For this, we estimate the prevalence at birth of co-morbidity, Q_D, but the same value for a_F^* was used because we fit the indirect model only to infant data which does not give information about the decrease of the function with age (Table 3.3).

3.B The effect of the health system on the case fatality rate

The evidence base for estimating the effect of in-patient care on case fatality rates is weak, largely because the incidence of severe episodes and case fatality in the community are not known. Formulae for the community case fatality rates can be derived from the overall incidence of severe malaria, proportion of cases admitted to hospital and the hospital case fatality rates (Table 3.4). For simplicity, they ignore age and season dependence and consider very approximate average rates for children.

Table 3.4 Case fatality rates for severe malaria

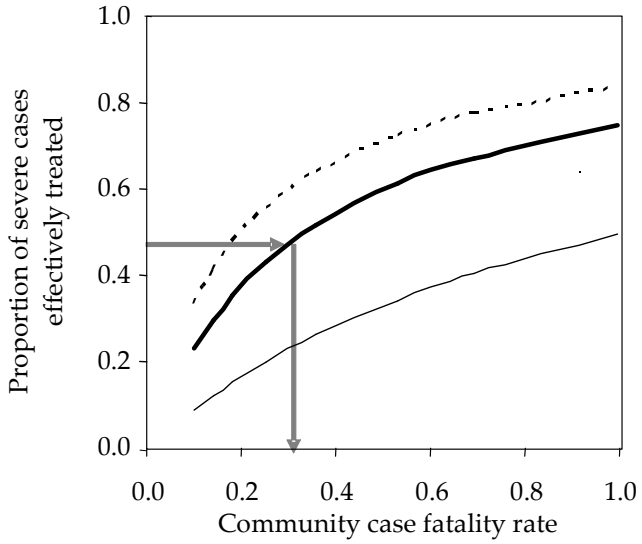
	Die	Survive	Total
Health facility	$Q_h P_h$	$(1-Q_h)P_h$	P_h
Community	$\frac{Q_h \varphi_1 (P_B - P_h)}{1-Q_h+Q_h\varphi_1}$	$\frac{(1-Q_h)(P_B-P_h)}{1-Q_h+Q_h\varphi_1}$	$P_B - P_h$
Total	P_C	$P_B - P_C$	P_B

Q_h = hospital case-fatality rate*; P_h :=incidence of hospital admissions for malaria; P_B = overall severe malaria incidence; P_C = overall incidence of malaria mortality*; φ_1 ratio of odds of community death to in-patient death. * indicates quantities for which we have reasonable estimates. The frequency of hospital admission per capita in rural Africa generally declines steeply with distance from the hospital, so the applicability of estimates to the whole district served is questionable.

The in-patient case-fatality rate in rural hospitals in sub-Saharan Africa, Q_h, is relatively well defined at about 0.1 (Figure 3.6). Paediatric hospital admission rates for the studies reported by Marsh and Snow[4,6] (Figure 3.2) average approximately 30/1000 person-years, and overall malaria mortality rates (from VA studies) is approxiamtely 10 per 1000[44] (Figure 3.7). Combining $Q_h = 1$ with the ratio of inpatient admissions to overall malaria deaths, $P_h / P_c = 30/10 = 3$, gives an estimate of $Q_h P_c / P_h$, the proportion of deaths that occur in hospital, of 0.3. This is similar to the results of a retrospective study of VAs in Tanzania[62] that found that about 33% of children less than five years of age who died of malaria had attended hospital at some time during their terminal illness, though the proportion who died there was lower.

In rural sub-Saharan Africa, since many hospitals are difficult to reach and often provide poor standards of care, attendance is likely to be even less frequent than in the Tanzanian study where public health services have a relatively high ratio to population. However in the research settings that contributed most of the VA and hospital data (many of them the same sites) hospital attendance rates may have been higher. In view of this, we assume that the proportion of cases treated is $P_h / P_B = 0.48$, in agreement with the proportion of severe episodes receiving in-patient treatment in the model of Goodman and others.[67] Using the formulae in Table 3.4, this gives an estimate of 31% for the case fatality rate in the community, corresponding to this level of treatment (arrows in Figure 3.12) and 21% for the overall case fatality rate. This implies that the health system prevents about 33% of malaria deaths. This compares with an estimate of 44% for the proportion of (all cause) deaths prevented by a good Kenyan district hospital.[68]

Figure 3.12 Effects of community case fatality rate on proportion of severe cases



The arrows indicate that effective treatment of 48% of severe episodes corresponds to a community case fatality rate of 31% under the assumptions given above. All values based on an assumption of an average in-patient case-fatality rate of 0.1.

Line style	Proportion of deaths in in-patients $Q_h P_h / P_c$	Ratio of in-patient admissions to overall malaria deaths P_h / P_C
———	0.1	0.9
——	0.3	2.1
------	0.5	3.3

We used the same figure of $P_h / P_B = 0.48$ to obtain an estimate of $\varphi_1 = 2.09$, for the ratio of odds of community death to inpatient death by fitting our stochastic model to verbal autopsy data adjusted for sensitivity and specificity (section 3.3).

Irrespective of the proportion of episodes resulting in admission, the low values of φ_1 that we propose at first sight appear to indicate that in-patient treatment has little benefit. The reality is undoubtedly more complex than this simple model. We dichotomised clinical malaria into severe and uncomplicated classes and assumed each class to be homogeneous in prognosis. In practice there is a continuous range of severity and inpatients are likely to disproportionately represent the most severe cases, many of whom arrive at health facilities when it is too late for treatment to be effective. This selection bias leads to an underestimate of the benefit of seeking treatment. Treatment may be life-saving even when administered less than optimally or based on imperfect diagnoses. Contact is made with formal health facilities at some stage during the terminal illness in many more cases than those who die in hospital.[62] For every case that dies despite making contact with the health services, many more may be saved.

3.C References

1. Greenwood B, Marsh K, Snow R. Why do some African children develop severe malaria? *Parasitol Today*. 1991, 7: 277-281

2. Snow R, Marsh K. The consequences of reducing transmission of *Plasmodium falciparum* in Africa. *Adv Parasitol* 2002, 52: 235-264

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

4. Snow R, Omumbo J, Lowe B, Molyneux CS, Obiero JO, Palmer A, Weber MW, Pinder M, Nahlen B, Obonyo C, Newbold C, Gupta S, Marsh K. Relation between severe malaria morbidity in children and level of *Plasmodium falciparum* transmission in Africa *Lancet* 1997, 349: 1650-1654

5. Smith T, Leuenberger R, Lengeler C. Child mortality and malaria transmission intensity in Africa. *Trends Parasitol* 2001, 17: 145-149

6. Marsh K, Snow R. Malaria transmission and morbidity. *Parassitologia*. 1999, 41: 241-246

7. Reyburn H, Drakeley C, Carneiro I *et al*. The epidemiology of severe malaria due to Plasmodium falciparum at different transmission intensities in NE Tanzania. LSHTM Malaria Centre Report 2002-2003. 2004: 6-7

8. Todd J, de Francisco A, O'Dempsey TJ, Greenwood BM. The limitations of verbal autopsy in a malaria-endemic region. *Ann Trop Paediatr* 1994, 14: 31-36

9. Quigley MA, Armstrong-Schellenberg JR, Snow R. Algorithms for verbal autopsies: a validation study in Kenyan children. *Bull WHO* 1996, 74: 147-154

10. Ezzati M, Vander Hoorn S, Rodgers A, Lopez A, Mathers C, Murray C, the Comparative Risk Assessment Collaborating Group. Estimates of global and regional potential health gains from reducing multiple major risk factors. *Lancet* 2003, 362: 271-280

11. Berkley J, Mwarumba S, Bramham K, Lowe B, Marsh K. Bacteraemia complicating severe malaria in children. *Trans R Soc Trop Med Hyg*. 1999, 93: 283-286

12. Molineaux L. Nature's experiment: what implications for malaria prevention? *Lancet*. 1997, 349: 1636-1637

13. Snow RW, Korenromp EL, Gouws E. Pediatric mortality in Africa: *Plasmodium falciparum* malaria as a cause or risk? *Am J Trop Med Hyg* 2004, 71: 16-24

14. Molineaux L. Malaria and mortality: some epidemiological considerations. *Ann Trop Med Parasitol* 1997, 91: 811-825

15. Giglioli G. Changes in the pattern of mortality following the eradication of hyperendemic malaria from a highly susceptible community. *Bull WHO* 1972, 46: 181-202

16. Molineaux L. The impact of parasitic diseases and their control, with an emphasis on malaria and Africa. In: Vallin J., Lopez A, eds. Health policy, social policy and mortality prospects. Ordina, 1985: 13-44

17. Newman P. Malaria and mortality. *J Am Stat Assoc* 1977, 72: 257-263

18. Bradley DJ. Morbidity and mortality at Pare-Taveta, Kenya and Tanzania, 1954-66: The effects of a period of malaria control. In: Feachem RG, Jamison DT, eds. Disease and mortality in Sub-Saharan Africa. World Bank, 1991

19. Alonso PL, Lindsay SW, Armstrong-Schellenberg JR, Conteh M, Hill AG, David PH, Fegan G, de Francisco A, Hall AJ, Shenton F. The effect of insecticide-treated bed nets on mortality of Gambian children . *Lancet* 1991, 337: 1499-1502

20. Molineaux L, Gramiccia G. The Garki Project. Geneva: World Health Organisation, 1980

21. Williams TN, Mwangi TW, Wambua S, Alexander N, Kortok M, Snow RW, Marsh K. Sickle cell trait and the risk of *Plasmodium falciparum* malaria and other childhood diseases. *J Inf Dis* 2005, 192: 178-186

22. Cuzin-Ouattara N, Van den Broek AH, Habluetzel A, Diabate A, Sanogo-Ilboudo E, Diallo DA, Cousens SN, Esposito F. Wide-scale installation of insecticide-treated curtains confers high levels of protection against malaria transmission in a hyperendemic area of Burkina Faso. *Trans R Soc Trop Med Hyg* 1999, 93: 473-479

23. Ilboudo-Sanogo E, Cuzin-Ouattara N, Diallo DA, Cousens SN, Esposito F, Habluetzel A, Sanon S, Ouedraogo AP. Insecticide-treated materials, mosquito adaptation and mass effect: entomological observations after five years of vector control in Burkina Faso. *Trans R Soc Trop Med Hyg* 2001, 95: 353-360

24. Robert V, Carnevale P, Ouedraogo V, Petrarca V, Coluzzi M. [Transmission of human malaria in a savanna village of South-West Burkina Faso]. *Ann Soc Belg Med Trop*. 1988, 68: 107-121

25. Robert V, Gazin P, Boudin C, Molez JF, Ouedraogo V, Carnevale P. [The transmission of malaria in a wooded savannah area and a rice-growing area

around Bobo Dioulasso (Burkina Faso)]. *Ann Soc Belg Med Trop* 1985, 65 Suppl 2: 201-214

26. Modiano D, Petrarca V, Sirima BS, Nebie I, Diallo D, Esposito F, Coluzzi M. Different response to Plasmodium falciparum malaria in west African sympatric ethnic groups. *Proc Natl Acad Sci USA* 1996, 93: 13206-13211

27. Coosemans M. Recherceh epidemiologique sur le paludisme dans la plaine de la Rusizi et dans l'Imbo sud (République du Burundi). Evaluation des moyens de lutte. Université Catholique de Louvain, 1987

28. Mbogo CN, Snow R, Kabiru EW, Ouma J, Githure JI, Marsh K, Beier JC. Low-level *Plasmodium falciparum* transmission and the incidence of severe malaria infections on the Kenyan coast. *Am J Trop Med Hyg* 1993, 49: 245-253

29. Mbogo CN, Snow R, Khamala CP, Kabiru EW, Ouma J, Githure JI, Marsh K, Beier JC. Relationships between *Plasmodium falciparum* transmission by vector populations and the incidence of severe disease at nine sites on the Kenyan coast. *Am J Trop Med Hyg* 1995, 52: 201-206

30. Beier JC, Oster CN, Onyango FK, Bales JD, Sherwood JA, Perkins PV, Chumo DK, Koech DV, Whitmire RE, Roberts CR. *Plasmodium falciparum* incidence relative to entomologic inoculation rates at a site proposed for testing malaria vaccines in western Kenya. *Am J Trop Med Hyg* 1994, 50: 529-536

31. Faye O, Gaye O, Faye O, Diallo S. [Transmission of malaria in villages far away or situated on the border of a mangrove in Senegal]. *Bull Soc Pathol Exot* 1994, 87: 157-163

32. Trape JF, Pison G, Preziosi MP, Enel C, Desgrées du Lou A, Delaunay V, Samb B, Lagarde E, Molez JF, Simondon F. Impact of chloroquine resistance on malaria mortality. *CR Acad Sci III*. 1998, 321: 689-697

33. Robert V, Dieng H, Lochouarn L, Traore SF, Trape JF, Simondon F, Fontenille D. [Malaria transmission in the rural zone of Niakhar, Senegal]. *Trop Med Int Health* 1998, 3: 667-677

34. Smith T, Charlwood JD, Kihonda J, Mwankusye S, Billingsley P, Meuwissen J, Lyimo E, Takken W, Teuscher T, Tanner M. Absence of seasonal variation in malaria parasitaemia in an area of intense seasonal transmission. *Acta Trop* 1993, 54: 55-72

35. Premji Z, Ndayanga P, Shiff C, Minjas J, Lubega P, MacLeod J. Community based studies on childhood mortality in a malaria holoendemic area on the Tanzanian coast. *Acta Trop* 1997, 63: 101-109

36. Lindsay SW, Snow R, Broomfield GL, Janneh MS, Wirtz RA, Greenwood BM. Impact of permethrin-treated bednets on malaria transmission by the *Anopheles gambiae* complex in The Gambia. *Med Vet Entomol* 1989, 3: 263-271

37. Thomson MC, D'Alessandro U, Bennett S, Connor SJ, Langerock P, Jawara M, Todd J, Greenwood BM. Malaria prevalence is inversely related to vector density in The Gambia, West Africa. *Trans R Soc Trop Med Hyg* 1994, 88: 638-643

38. Bockarie M, Service MW, Barnish G, Maude G, Greenwood BM. Malaria in a rural area of Sierra Leone. III. Vector ecology and disease transmission. *Ann Trop Med Parasitol* 1994, 88: 251-262

39. Akogbeto PM, Nahum A. [Impact of deltamethrin impregnated mosquito nets on the transmission of malaria in the coastal lagoon area, Benin]. *Bull Soc Pathol Exot* 1996, 89: 291-298

40. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006 Suppl 2, 75: 19-31

41. Appawu M, Owusu-Agyei S, Dadzie S, Asoala V, Anto F, Koram K, Rogers W, Nkrumah F, Hoffman SL, Fryauff DJ. Malaria transmission dynamics at a site in northern Ghana proposed for testing malaria vaccines. *Trop Med Int Health* 2004, 9: 164-170

42. Kirkpatrick S, Gelatt CDJr, Vecchi MP. Optimization by simulated annealing. *Science* 220, 671-680. 1983.

43. Press WH, Flannery BP, Teukolsky SA, Vetterling WT. Numerical recipes in C: the art of scientific computing. Cambridge: Cambridge University Press, 1988

44. Korenromp EL, Williams BG, Gouws E, Dye C, Snow R. Measurement of trends in childhood malaria mortality in Africa: an assessment of progress toward targets based on verbal autopsy. *Lancet Infect Dis* 2003, 3: 349-358

45. D'Alessandro U, Olaleye BO, McGuire W, Langerock P, Bennett S, Aikins MK, Thomson MC, Cham MK, Cham BA, Greenwood BM. Mortality and morbidity from malaria in Gambian children after introduction of an impregnated bednet programme. *Lancet* 1995, 345: 479-483

46. Barnish G, Maude G, Bockarie M, Eggelte TA, Greenwood BM, Ceesay S. Malaria in a rural area of Sierra Leone. I. Initial results. *Ann Trop Med Parasitol*. 1993, 87: 125-136

47. Duboz P., Vaugelade J, Debouverie M. Mortalité dans l'enfance dans la région de Niangoloko. 1989. Ouagadougou, ORSTOM.

48. Delauney V, Etard J-F, Préziosi M-P, Marra A, Simondon F. Decline of infant and child mortality rates in rural Senegal over a 37-year period (1963-1999). *Int J Epidemiol* 2001, 30: 1286-1293

49. INDEPTH Network. Population, Health and Survival at INDEPTH Sites. Ottawa: IDRC, 2002

50. Spencer HC, Kaseje DC, Mosley WH, Sempebwa EK, Huong AY, Roberts JM. Impact on mortality and fertility of a community-based malaria control programme in Saradidi, Kenya. *Ann Trop Med Parasitol* 1987, 81 Suppl 1: 36-45

51. Armstrong-Schellenberg JR, Abdulla S, Minja H, Nathan R, Mukasa O, Marchant T, Mponda H, Kikumbih N, Lyimo E, Manchester T, Tanner M, Lengeler C. KINET: a social marketing programme of treated nets and net treatment for malaria control in Tanzania, with evaluation of child health and long-term survival. *Trans R Soc Trop Med Hyg* 1999, 93: 225-231

52. Snow R, Craig M, Newton C, Steketee RW. The public health burden of *Plasmodium falciparum* malaria in Africa: Deriving the numbers. 2003. Bethesda, Maryland., Fogarty International Center, National Institutes of Health.

53. Dietz K, Raddatz G, Molineaux L. A mathematical model of the first wave of *Plasmodium falciparum* asexual parasitaemia in non-immune and vaccinated individuals. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 46-55

54. Snow R, Bastos de Azevedo I, Lowe B, Kabiru EW, Nevill C, Mwankusye S, Kassiga G, Marsh K, Teuscher T. Severe childhood malaria in two areas of markedly different falciparum transmission in east Africa. *Acta Trop* 1994, 57: 289-300

55. Macintosh CL, Beeson JG, Marsh K. Clinical features and pathogenesis of severe malaria. *Trends Parasitol* 2004, 20: 597-603

56. Reyburn H, Mbatia R, Drakeley C, Carneiro I, Mwakasungula E, Mwerinde O, Saganda K, Shao J, Kitua A, Olomi R, Greenwood BM, Whitty CJ. Overdiagnosis of malaria in patients with severe febrile illness in Tanzania: a prospective study. *BMJ* 2004, 329: 1212

57. Makani J, Matuja W, Liyombo E, Snow R, Marsh K, Warrell DA. Admission diagnosis of cerebral malaria in adults in an endemic area of Tanzania: implications and clinical description. *QJM* 2003, 96: 355-362

58. Binka F, Hodgson A, Adjuik M, Smith T. Mortality in a seven-and-a-half-year follow-up of a trial of insecticide-treated mosquito nets in Ghana. *Trans R Soc Trop Med Hyg* 2002, 96: 597-599

59. Brown V, Abdir IM, Rossi M, Barboza P, Paugam A. Epidemic of malaria in north-eastern Kenya. *Lancet* 1998, 352: 1356-1357

60. Ceita JGV, Buck AA, editors. Malaria in São Tomé and Principe. Washington DC: American Institute of Biological Sciences, 1986

61. Smith T, Genton B, Betuela I, Rare L, Alpers MP. Mosquito nets for the elderly? *Trans R Soc Trop Med Hyg* 2002, 96: 37-38

62. de Savigny D, Mayombana C, Mwageni E, Masanja H, Minhaj A, Mkilindi Y, Mbuya C, Kasale H, Reid G. Care-seeking patterns for fatal malaria in Tanzania. *Malar J* 2004, 3: 27

63. Tediosi F, Maire N, Smith T, Hutton G, Utzinger J, Ross A, Tanner M. An approach to model the costs and effects of case management of *Plasmodium falciparum* malaria in sub-saharan Africa. *Am J Trop Med Hyg* 2006 Suppl 2, 75: 90-103

64. Lengeler C, Cattani J, de Savigny D. Net Gain, a new method for preventing malaria deaths. Geneva: IDRC/WHO, 1996

65. Schellenberg D, Menendez C, Kahigwa E, Aponte J, Vidal J, Tanner M, Mshinda H, Alonso PL. Intermittent treatment for malaria and anaemia control at time of routine vaccinations in Tanzanian infants: a randomised, placebo-controlled trial. *Lancet* 2001, 357: 1471-1477

66. Graves P, Gelband H. Vaccines for preventing malaria. *Cochrane Database Syst Rev*. 2003, CD000129

67. Goodman CA, Coleman PG, Mills A. Economic analysis of malaria control in sub-Saharan Africa. Geneva: Global Forum for Health Research, 2000

68. Snow RW, Mung'ala VO, Foster D, Marsh K. The role of the district hospital in child survival at the Kenyan Coast. *Afr J Health Sci* 1994, 1: 71-75

Chapter 4

The effect of malaria transmission intensity on neonatal mortality in endemic areas

Amanda Ross and Thomas Smith

Swiss Tropical Institute, Basel, Switzerland

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4.1 Summary

Estimates of the impact of *Plasmodium falciparum* infections during pregnancy on neonatal mortality have not taken into account how this varies with the level of malaria endemicity and thus do not indicate the possible effects of malaria control strategies that reduce transmission. We now review the relevant literature and propose a mathematical model for the association between *P. falciparum* transmission and neonatal death.

The excess risk of neonatal mortality in malaria-endemic areas appears to be insensitive to the intensity of *P. falciparum* transmission over a wide range of endemicity. Moderate reductions in the overall level of malaria transmission in endemic areas are therefore unlikely to significantly reduce neonatal mortality. The magnitude of the excess risk is very uncertain because existing estimates are heavily dependent on the questionable assumption that the effects are mediated by birth weight. Accurate prediction of the impact of malaria control measures targeted at pregnant women requires direct estimates of malaria-attributable neonatal mortality rates.

4.2 Introduction

In endemic areas, infants are at high risk of *Plasmodium falciparum* mortality, and there is a strong association between all-cause infant mortality and malaria transmission intensity.[1] Infections received in early infancy are unlikely to result in death;[2] however maternal infections during the first, and to a lesser extent later, pregnancies increase the risk of mortality in the newborn.[3-5]

The indirect mortality due to maternal infection could affect estimates of the impact of a malaria intervention. It may not, in the short-term, be amenable to interventions targeted at infants. Nevertheless, effective malaria control may reduce transmission in the community and therefore might be expected to reduce the risk of such mortality. As one component of a project to develop a comprehensive simulation of the likely impact of potential malaria vaccines delivered to infants via the expanded program on immunization,[6] we develop a model for the relationship between malaria transmission and indirect mortality in the neonatal period (birth to 28 days). Most deaths due to post-natal malaria infection occur after the first month of life and a model for these is described in Chapter 3. The magnitude of the impact of maternal malaria infection on neonatal mortality is unclear, as is the mechanism by which it occurs.[5,7] There is little data with which to make direct estimates, due in part to the enormous sample size requirements. In the absence of such data, previous studies have used estimates of the effect of maternal malaria on birth weight, and combined these with independent measures of the association between low birth weight and mortality. The resulting estimates apply either to all endemic areas in Africa taken together, or to a single site (Table 4.1).

Table 4.1. Estimates of neonatal and infant mortality due to malaria in pregnancy derived using birth weight measures

	Primigravidae		Multigravidae		All gravidities	
Reference	%†	Rate‡	%†	Rate‡	%†	Rate‡
Neonatal mortality						
Greenwood and others[8§¶]	42%		6%			
Goodman and others[9§]	24%	11	-	-		
Guyatt and Snow 2001[10#]	18%	7	-	-	11%	4
Infant mortality						
Greenwood and others[8§¶]	18%	14	4%	2		
Guyatt and Snow[10#]	10%	8	-		6%	4
Steketee and others[11]**	-		-		3-8%	-
Murphy and Breman[12#]	-		-		-	3-17

†Estimated percentage of mortality attributable to malaria in pregnancy. ‡Estimated deaths per 1000 live births attributable to malaria in pregnancy. § Estimates are based on the changes in the proportion of low birth weight[8] or in mean birth weight[9] associated with anti-malarial drugs during pregnancy in clinical trials. ¶Estimates apply to The Gambia, all the other estimates apply to sub-Saharan Africa. #Estimates are based on observational studies of maternal infection and low birth weight. ** 8% is a composite of 18% and 4% from Greenwood and others.[8] 3% is derived from the estimated reduction in low birth weight associated with clearing placental and peripheral parasites, and does not include anaemia. The perinatal mortality rate for countries with a Human Development Index between 500 and 800 has been estimated[13]: non-endemic countries had a mean perinatal mortality rate of 30/1000 and endemic countries, 50.5/1000.

Previous estimates of the impact of *P. falciparum* malaria on neonatal mortality have not considered how it varies with the level of transmission. Forecasting the effects of malaria control in endemic areas needs estimates not only of the average contribution of malaria to neonatal mortality, but also of the quantitative relationship between transmission intensity and neonatal mortality. In order to estimate this relationship, we have now summarized available data from clinical trials on birth weight, from between-site comparisons for sites with either entomological or prevalence data together with estimates of mortality, and from observational studies and reviews. We have used these summaries to develop a simple model of neonatal mortality due to malaria in pregnancy over a range of transmission intensities.

4.3 Methods

Our model relates neonatal mortality resulting from malaria infection during pregnancy to the age-specific prevalence of *P. falciparum* in the general population. This allows it to be integrated into a comprehensive simulation[6] and uses our parasitological model[14,15] as a foundation. We model neonatal mortality rather than perinatal mortality (28 weeks gestation to 7 days after birth) so that the predictions can be included in disability-adjusted life year calculations.[16] However, we acknowledge that the increased risk of mortality associated with maternal infection is not necessarily confined to the neonatal period.

There is little data with which to directly relate the risk of indirect malaria neonatal mortality to *P. falciparum* prevalence in young adults. Where available we used proxy variables for the exposure or outcome, which lead us to consider separately the relationship between malaria infection in primigravidae and neonatal mortality and the relationship between parasite prevalence in young adults in the general population and primigravidae. We focus on primigravidae because they suffer the most pronounced effects and have the most data available, and we compute the overall impact on the neonatal mortality rate by assuming that 30% of livebirths are born to primigravidae.

Relationship between malaria infection among primigravidae and neonatal mortality

Data summaries Data summaries were used to provide information on the relationship between malaria infection among the population of primigravidae and the risk of neonatal mortality. We used various sources of information on malaria infection during pregnancy, for both the EIR and *P. falciparum* prevalence. In order to dissect the observed association between infant mortality and transmission intensity[1] into neonatal and post-neonatal mortality, we carried out a literature search for sub-Saharan Africa sites with information on both the EIR and neonatal or post-neonatal mortality rates. The mortality rates were not parity-specific.

In addition, birth weight has been previously used as a proxy in a number of studies estimating mortality in the newborn (Table 4.1) using both observational data and data from controlled clinical trials of anti-malarial drugs in pregnancy. We use data from the trials and assume that, for a particular trial setting, the difference in mean birth weight between the intervention and control groups is an approximate measure of the impact that malaria infection in pregnancy has on birth weight. Although women are not necessarily 100% protected from malaria throughout their pregnancy, the drugs have a large impact on peripheral and placental prevalence.[17] To examine the association between the estimated birth weight difference and EIR, we matched entomological data to the sites of the trials.

We also examined meta-analyses of perinatal mortality rates by maternal peripheral parasite prevalence in malaria-endemic areas,[13] of birth weight by childhood parasite prevalence,[18] and of birth weight by placental prevalence.[19]

Model From the analyses of neonatal mortality and transmission intensity (Results section 4.4) using the data summaries above, we propose that the risk of neonatal mortality attributable to malaria in pregnancy, μ_{PG}, saturates at low transmission levels. Therefore we propose a relationship for primigravidae between the prevalence x_{PG} and the neonatal mortality rate μ_{PG} of the form

$$\mu_{PG} = \mu_{\max}\left[1-\exp\left(-\frac{x_{PG}}{x_{PG}^{*}}\right)\right] \qquad (4.1)$$

where $\mu_{\max}$ and x_{PG}^{*} are constants, and which satisfies the additional constraint that in the absence of malaria $\mu_{PG}=0$. We use an estimate of the efficacy of anti-malarial drugs in pregnancy[20] to assign a value of $\mu_{\max}=0.011$ (11/1000 livebirths among primigravidae). To compute the overall effect on the neonatal mortality rate we assume that 30% of livebirths are born to primigravidae and thus our model predicts an overall risk of malaria-attributable neonatal mortality of $0.3\mu_{PG}$.

Relationship between the prevalence of *P. falciparum* in the general population and prevalence in primigravidae

We relate the prevalence of *P. falciparum* in primigravidae to the age-specific prevalence in the general population. We use data from a review of 27 cross-sectional studies comparing the peripheral prevalence either at antenatal attendance or at delivery in primigravidae and multigravidae.[5] We approximate the prevalence in multigravidae by that of the general population of the same age. We could find little evidence to support this assumption, but it is not a critical assumption for the model predictions and we believe it to be a closer approximation than using the prevalence in the general population for that in primigravidae directly. We fit a statistical model to estimate the prevalence in primigravidae from that in multigravidae. The predicted prevalence in primigravidae, x_{PG}, is constrained to be zero when x_{MG}, the prevalence in multigravidae, is zero. To allow x_{PG} either to increase or saturate at high values of x_{MG} we fit a curve of the form

$$x_{PG} = 1-\frac{1}{1+\left(\frac{x_{MG}}{x_{MG}^{*}}\right)} \qquad (4.2)$$

where x^*_{MG} is a critical value of x_{MG}. This model was fitted in WinBUGS 1.4.[21]

The proportions of women with placental and peripheral parasitaemia at delivery are approximately equal in the same settings,[5] even though in individual women peripheral blood slides are not a good indicator of placental infection.[22,23]

4.4 Results

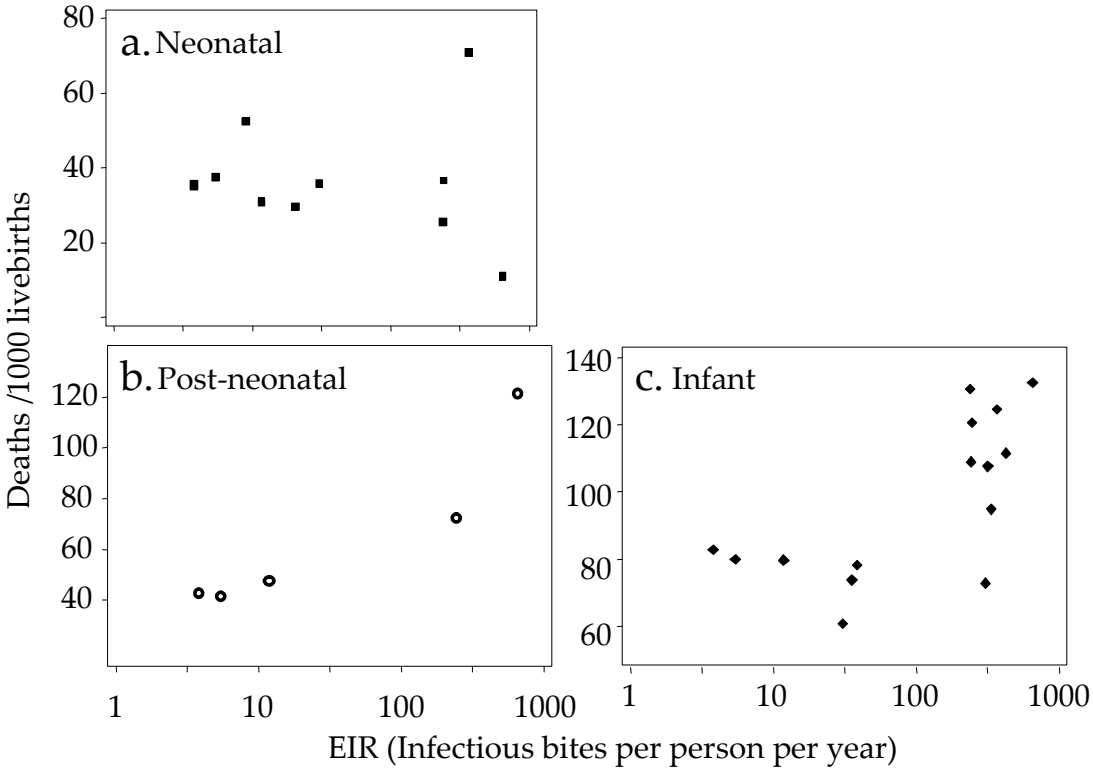
The relationship between malaria infection in primigravidae and neonatal mortality

As reported by Hyder and others,[24] we found few reported neonatal mortality rates from sub-Saharan Africa and we could locate entomological data for only those given in Table 4.2. Among these sites there is no evidence of an association between neonatal mortality and malaria transmission intensity (Figure 4.1a), yet such an association is evident for both post-neonatal and overall infant mortality (Figure 4.1b and c). We acknowledge that there are many differences other than malaria transmission intensity between the studies included in the ecological comparison of mortality rates, and there may be an association between malaria transmission and other diseases, availability of effective treatment, or poverty that may serve to overestimate or underestimate the effect of maternal malaria infection. We conclude that the relationship of transmission intensity with the risk of neonatal mortality is much weaker than that with post-neonatal mortality, although there are few reported post-neonatal mortality rates from settings with entomological data.

Table 4.2. All-cause neonatal, post-neonatal and infant mortality rates from sites with entomological data*

Study site	Reference for entomology data	Year of entomol-ogy data	EIR	Mortality data reference	Year of mortality data	Number of livebirths	Neonatal mortality rate	Post-neonatal mortality rate	Infant mortality rate
Areas I-V, The Gambia	[25]	1991	3.7	[26]	1992	3063	35.9	43	83
Upper River Division, The Gambia	[25]	1991	5.3	[27]	1989-93	26894	37.7	42.4	80.2
Farafenni, The Gambia	[28]	1987	8.9	[29]	1984-87	610	52.5	-	-
Niakhar, Senegal	[30]	1995	11.6	[31]	1995-99	5997	31	48	80
Kilifi, Kenya	[32]	1997-98	20	[33]	1999-2003	2189	29.7	-	-
Bo, Sierra Leone	[34]	1990-91	34.7	[35]	1990	<100	-	-	74.0
Mlomp,Senegal	[36]	1995-96	30	[36]	1995	-	-	-	61
Mlomp, Senegal	[36]	1995-96	30	[37]	1985-89	917	36	-	-
Manhica, Mozambique	Aponte J (pers comm.)		38	[31]	1998-99	1280	-	-	78.5
Yombo, Tanzania	[38]	1992	234	[38]	1992-94	1130	25.7	-	131.0
Saradidi, Kenya	[39]	1986-87	239	[40]	1981-83	1168	36.8	72.8	109.4
Karangasso,	[41]	1985	244	[42]	1986-88	-	-	-	121
Nyanza-Lac, Burundi	[43]	1990-91	312	[43]	1990-91	813	-	-	108
Bandafassi, Senegal	[36]	1995-96	363	[44]	1989-92	1448	71	-	-
Bandafassi, Senegal	[36]	1995-96	363	[31]	1995-99	2122	-	-	124.9
Namawala, Tanzania	[45]	1990-91	329	[46]	1902	1902	-	-	95.2
Navrongo, Ghana	[47]	2001-02	418	[31]	1994-99	20462	-	-	111.9
Muheza, Tanzania	[48]	1987-88	639	[49]	1992-93	361	11.1	121.9	133

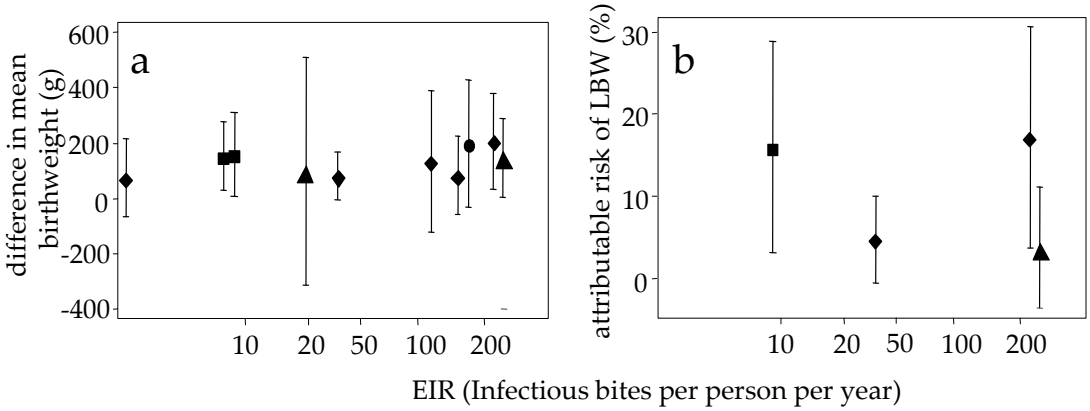
Figure 4.1. Mortality rates by transmission intensity



We found no evidence of an association between the estimated effect of anti-malarial drug interventions on birth weight and EIR (Figure 4.2). The overall pattern observed may be biased by confounders such as drug resistance. Since none of the trial settings had a very low transmission intensity, this is not inconsistent with a review of studies where the proportion of low birth weight (<2500g) babies was lower for studies set in areas with an EIR<1 compared with settings with an EIR≥1. However, among settings with EIR≥1 there was no clear association.[18]

We conclude that there is little or no association between neonatal mortality and transmission intensity once the transmission is above a very low level. This lack of an association enables us to infer that there can be little association also between the prevalence in primigravidae and neonatal mortality. The prevalence of *P. falciparum* in young adults is itself insensitive to transmission intensity.[50]

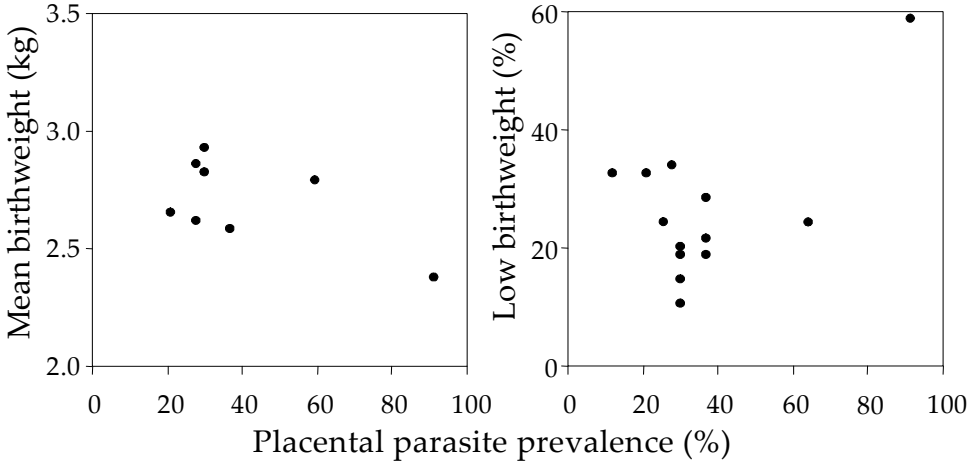
Figure 4.2. Estimated effect of anti-malarial drug interventions on birth weight



a. Estimated mean change in birth weight due to intervention. **b.** Excess risk of low birth weight (LBW) (%LBW in controls-%LBW in drug group). Data from 10 trials comparing anti-malarial drug use to control either placebo or no drug controls[29,51-59] were analyzed. Trials were not included if they compared multiple drugs with no inactive control [60-62] or could not be matched to entomologic data.[63] The estimates refer to primigravidae, or primigravidae and secundigravidae together in the case of one trial. ♦=chloroquine; ■=dapsone-pyrimethamine. ▲=sulphadoxine-pyrimethamine; ● =pyrimethamine. The error bars show the 95% confidence intervals.

Our conclusion is supported by reviews of related outcomes and prevalence. A review of observational studies that found that there was no obvious linear trend between perinatal mortality (28 weeks gestation to the first 7 days) and maternal peripheral parasite prevalence in endemic areas.[13] The association between the proportion of primigravidae with placental parasitaemia and birth weight is weak (Figure 4.3) after accounting for highly influential points (data from Brabin and others[19]), although this may be confounded by the inclusion of studies from South East Asia.

Figure 4.3. Placental prevalence and birth weight



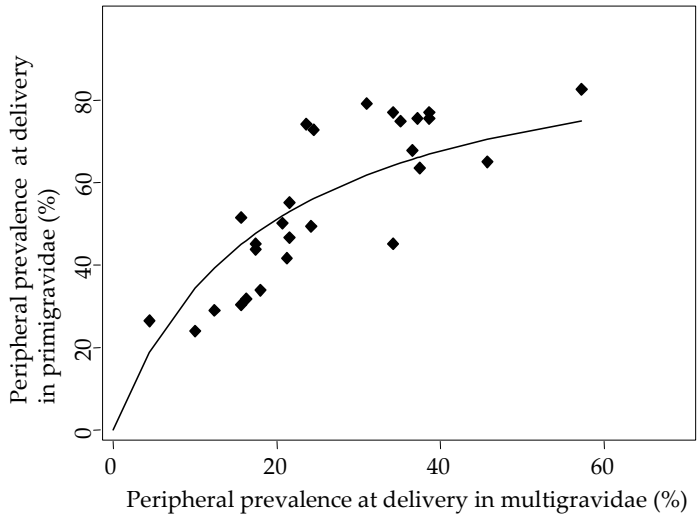
Data for primigravidae from Brabin and others.[19]

These observations contribute only to the shape of our model of the relationship of malaria-attributable neonatal mortality with transmission. Since the malaria-attributable neonatal mortality rate in primigravidae, μ_{PG}, appears to be independent of the transmission intensity across the settings for which we have data, we were not able to use a formal fit to data to obtain estimates of the parameters $\mu_{\max}$ and x^*_{PG} (equation 5.1). We follow Goodman and others[20] in assigning a value of $\mu_{\max} = 0.011$ (11/1000 livebirths among primigravidae). Since saturation seems to occur at lower prevalence than any measured in endemic areas the data only suggest a rough idea of the upper limit of the quantity x^*_{PG}. In the absence of more relevant data, we set $x^*_{PG} = 0.25$.

Relationship between the prevalence of *P. falciparum* in the general population and prevalence in primigravid women

We relate the prevalence of infection in primigravidae, x_{PG}, to that in the multigravidae, x_{MG} (equation 4.2). We obtained a good fit to relationship between x_{PG} and x_{MG} with a value of x^*_{MG} = 0.19 (95% confidence interval = 0.16 – 0.23), which corresponds to the observation that x_{PG} and x_{MG} are approximately proportional when both are low, but as prevalence increases in multigravidae, it approaches 100% in primigravidae and cannot continue to be proportional (Figure 4.4).

Figure 4.4. Relationship between peripheral prevalence at delivery in primigravidae and multigravidae

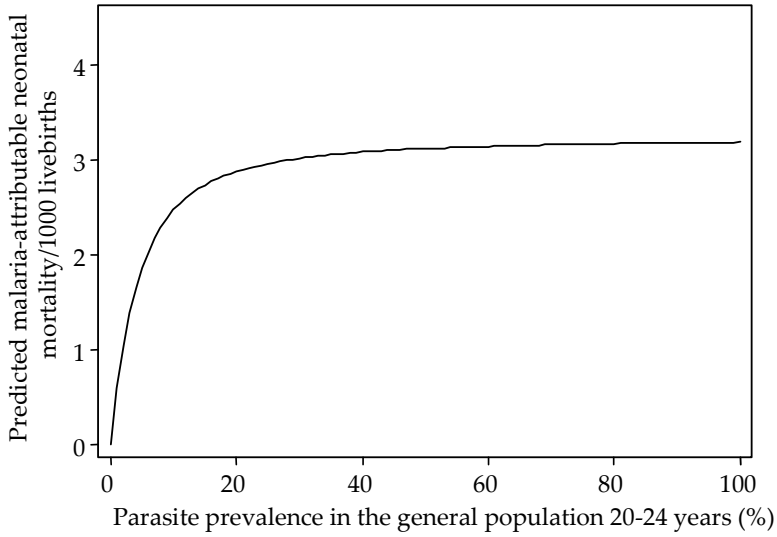


The points represent cross-sectional surveys collated by Brabin and Rogerson.[5] The fitted line corresponds to the model of equation 5.2.

To compute the overall effect on the neonatal mortality rate, we assume that 30% of live births are born to primigravidae and thus our model predicts an overall risk of malaria-attributable neonatal mortality of $0.3\mu_{PG}$. Assuming x_{MG} to be equivalent to the

prevalence of patent *P. falciparum* in adults aged 20 to 24 years in the general population, we can thus combine equations 4.1 and 4.2 to obtain predictions of the malaria-attributable neonatal mortality rate as a function of prevalence as shown in Figure 4.5. Our model predicts very little effect of transmission intensity on neonatal mortality.

Figure 4.5. Predicted malaria attributable neonatal mortality rate as a function of prevalence in the general population aged 20-24 years



4.5 Discussion

Although *P. falciparum* infections during pregnancy in primigravidae have an important impact on the newborn, we found little or no association between neonatal mortality and malaria transmission intensity in stable transmission areas.

This lack of association with transmission intensity is to be expected, if, as is likely, most women in these areas are infected at some stage in their pregnancy, and also that immunity to pregnancy-associated malaria is gained through relatively few infections. Despite problems with the sensitivity of histology,[64] the proportion of placenta with histological evidence of active or past infection is very high even in endemic areas with relatively low transmission: in primigravidae in Kilifi, Kenya it was 77%[23] and in The Gambia it was 76%.[65] A subset of parasites expressing particular cytoadherence properties are thought to account for much of the pathology of malaria in pregnancy.[66-68] It has been suggested that a single infection with such a phenotype may be sufficient to stimulate an immunological reaction,[5] although this is not known. This may both explain why the adverse consequences of maternal infection mainly occur in first, and to a lesser extent second, pregnancies, and why the intensity of superinfection appears to have little effect.

The model would predict little change in mortality from a decrease in transmission intensity unless it reaches a very low level. Trials of insecticide-treated nets provide some data: while increased birth weight was observed in areas with low transmission (Thailand and The Gambia),[69,70] results from areas with more intense transmission are mixed. No impact was observed in Kilifi, Kenya and Navrongo, Ghana,[23,71] but a reduction in the proportion of low birth weight babies was found in Western Kenya.[72] However, the transmission intensity after the introduction of the nets would be more relevant than the baseline transmission intensity.

Since there is considerable uncertainty about the patho-physiology of the effects of *P. falciparum* infection on neonatal mortality, we attempted to avoid assumptions about mechanisms in formulating our predictive model. However, all the available estimates of this effect (Table 4.1), including the one we use, depend on associations with birth weight and assume that the risk of death in babies of the same birth weight is the same whether their mothers had placental malaria or not, and that the relevant effect on the birth weight distribution can easily be summarized either by the mean or by the proportion of birth weights below a standard cut-off. Both these assumptions have been questioned.[17,73] If the full distribution of birth weights is available this should be analyzed as a mixture of the predominant normal distribution and a residual distribution in the form of a tail at low birth weights.[74] It is the relative size of this residual distribution that is the feature associated with mortality.[73] Comparison of three birth weight distributions from areas of high, medium, and low transmission settings suggest that the overall mean and size of the residual tail may move in tandem.[75] This, however, is very indirect support for models that assume the maternal effect to be adequately captured by simple summaries of the effect on birth weight when there is not even convincing evidence of that birth weight is on the causal pathway between maternal infection and neonatal death.

An additional highly uncertain element of our model is the value of 0.25 assigned to the parameter x^*_{PG}. x^*_{PG} determines the prevalence at which neonatal mortality saturates, and data from endemic areas provide only an approximate idea of the upper limit of this quantity because saturation seems to occur at a lower prevalence than any measured in endemic areas. This is one of several reasons why our model is in any case unlikely to be appropriate in areas of unstable transmission such as South East Asia. In such areas, the impact of malaria in pregnancy on the mother is likely to be more severe, and thus the risk associated with individual infections may be higher. In stable endemic areas, acute effects on the mother are less frequent[64,76] presumably because of immunity that has already been acquired prior to pregnancy.

A comprehensive model for the effects of malaria in pregnancy would also need to address the question of the timing and intensity of the infections. Babies born during the rainy season were lighter than those born during the low transmission periods in The Gambia and Mali.[26,29,77] Maternal malaria infection is likely to contribute to this, but the implications for neonatal mortality are unclear. We also do not consider the effects of infection with human immunodeficiency virus (HIV). The prevalence of HIV in

women varies between countries in sub-Saharan Africa,[78] and HIV infection is associated both with an increased prevalence of malaria parasitaemia during pregnancy for all gravities[79-81] and with increased rates of adverse perinatal outcomes.[82]

We are not in a position to provide good estimates of the potential impact of interventions (such as intermittent preventive treatment or vaccination) targeted at pregnant women. This is for two reasons. Firstly, we consider only the impact on the infant and not the health effects for the mother which may be substantial[83] (though the prevalence of anaemia in pregnancy is considered by our model of anaemia[84]). Secondly and most importantly, there is an unacceptable level of uncertainty associated with estimates of malaria in pregnancy associated neonatal mortality that depend on the assumed relationship with birth weight. The burden of neonatal mortality caused by *P. falciparum* will remain highly uncertain so long as we are dependent on indirect assessments.

Despite these uncertainties, we propose that our model is adequate for predicting the effects of preventative interventions targeted at infants and children or the general population on the risk of neonatal mortality associated with maternal infection, and we propose to incorporate equations 4.1 and 4.2 into our general model of the epidemiology of *P. falciparum*.[6] The main predictions relating to neonatal mortality are already evident and are clearly insensitive to the uncertainties documented above. We predict that interventions targeted at infants such as vaccination would have to reduce the infectious reservoir to very low levels to affect indirect neonatal mortality.

4.A References

1. Smith T, Leuenberger R, Lengeler C. Child mortality and malaria transmission intensity in Africa. *Trends Parasitol* 2001, 17: 145-149

2. Snow R, Nahlen B, Palmer A, Donnelly CA, Gupta S, Marsh K. Risk of severe malaria among African infants: direct evidence of clinical protection during early infancy. *J Infect Dis* 1998, 177: 819-822

3. Steketee RW, Nahlen B, Parise M, Menendez C. The burden of malaria in pregnancy in malaria-endemic areas. *Am J Trop Med Hyg* 2001, 64: 28-35

4. World Health Organisation. A strategic framework for malaria prevention and control during pregnancy in the African region. WHO, 2004

5. Brabin B, Rogerson S. The epidemiology and outcomes of maternal malaria. In: Duffy E, Fried M, eds. Malaria in pregnancy - deadly parasite, susceptible host. London: Taylor and Francis, 2003

6. Smith T, Killeen GF, Maire N, Ross A, Molineaux L, Tediosi F, Hutton G, Utzinger J, Dietz K, Tanner M. Mathematical modeling of the impact of malaria vaccines on the clinical epidemiology and natural history of *Plasmodium falciparum* malaria: Overview. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 1-10

7. Menendez C. Malaria during pregnancy: A priority area of malaria research and control. *Parasitol Today* 1995, 11: 178-183

8. Greenwood AM, Armstrong-Schellenberg JR, Byass P, Snow R, Greenwood BM. Malaria chemoprophylaxis, birth weight and child survival. *Trans R Soc Trop Med Hyg* 1992, 86: 483-485

9. Goodman CA, Coleman PG, Mills A. Cost-effectiveness of malaria control in sub-Saharan Africa. *Lancet* 1999, 354: 378-385

10. Guyatt HL, Snow R. Malaria in pregnancy as an indirect cause of infant mortality in sub-Saharan Africa. *Trans R Soc Trop Med Hyg* 2001, 95: 569-576

11. Steketee RW, Wirima JJ, Campbell CC. Developing effective strategies for malaria prevention programs for pregnant African women. *Am J Trop Med Hyg* 1996, 55: 95-100

12. Murphy SC, Breman JG. Gaps in the childhood malaria burden in Africa: cerebral malaria, neurological sequelae, anemia, respiratory distress, hypoglycaemia, and complications of pregnancy. *Am J Trop Med Hyg* 2001, 64: 57-67

13. Van Geertruyden J-P, Thomas F, Erhart A, D'Alessandro U. The contribution of malaria in pregnancy to perinatal mortality. *Am J Trop Med Hyg* 2004, 71 Suppl 2: 35-40

14. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationship between the entomologic inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 11-18

15. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 19-31

16. Tediosi F, Hutton G, Maire N, Smith TA, Ross A, Tanner M. Predicting the cost-effectiveness of introducing a pre-erythrocytic malaria vaccine into the expanded program on immunization in Tanzania. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 131-143

17. Garner P, Gulmezoglu AM. Drugs for preventing malaria-related illness in pregnant women and death in the newborn. *Cochrane Database Syst Rev* 2003, CD000169

18. Guyatt HL, Snow RW. Impact of malaria during pregnancy on low birth weight in sub-Saharan Africa. *Clin Microbiol Rev* 2004, 17: 760-769

19. Brabin BJ, Romagosa C, Abdelgalil S, Menendez C, Verhoeff FH, McGready R, Fletcher KA, Owens S, D'Alessandro U, Nosten F, Fischer PR, Ordi J. The sick placenta-the role of malaria. *Placenta* 2004, 25: 359-378

20. Goodman C, Coleman PG, Mills A. The cost-effectiveness of antenatal malaria prevention in sub-Saharan Africa. *Am J Trop Med Hyg* 2001, 64: 45-56

21. Winbugs. Cambridge, England: MRC-BSU, 2003

22. Ismail MR, Ordi J, Menendez C. Placental pathology in malaria: a histological, immunohistochemical, and quantitative study. *Hum Pathol* 2000, 31: 85-93

23. Shulman CE, Dorman EK, Talisuna AO, Lowe BS, Nevill C, Snow RW, Jilo H, Peshu N, Bulmer JN, Graham S, Marsh K. A community randomized controlled trial of insecticide-treated bednets for the prevention of malaria and anaemia among primigravid women on the Kenyan coast. *Trop Med Int Health* 1998, 3: 197-204

24. Hyder A, Wali SA, McGuckin J. The burden of disease from neonatal mortality: a review of South Asia and sub-Saharan Africa. *Br J Obst Gynaecol* 2003, 110: 894-901

25. Thomson MC, D'Alessandro U, Bennett S, Connor SJ, Langerock P, Jawara M, Todd J, Greenwood BM. Malaria prevalence is inversely related to vector density in The Gambia, West Africa. *Trans R Soc Trop Med Hyg* 1994, 88: 638-643

26. D'Alessandro U, Olaleye BO, McGuire W, Langerock P, Bennett S, Aikins MK, Thomson MC, Cham MK, Cham BA, Greenwood BM. Mortality and morbidity from malaria in Gambian children after introduction of an impregnated bednet programme. *Lancet* 1995, 345: 479-483

27. Jaffar S, Leach A, Greenwood AM, Jepson A, Muller O, Ota MO, Bojang K, Obaro S, Greenwood BM. Changes in the pattern of infant and childhood mortality in Upper River Division, The Gambia, from 1989 to 1993. *Trop Med Int Health* 1997, 2: 28-37

28. Lindsay SW, Snow R, Broomfield GL, Janneh MS, Wirtz RA, Greenwood BM. Impact of permethrin-treated bednets on malaria transmission by the *Anopheles gambiae* complex in The Gambia. *Med Vet Entomol* 1989, 3: 263-271

29. Greenwood BM, Greenwood AM, Snow RW, Byass P, Bennett S, Hatib-N'Jie AB. The effects of malaria chemoprophylaxis given by traditional birth attendants on the course and outcome of pregnancy. *Trans R Soc Trop Med Hyg* 1989, 83: 589-594

30. Robert V, Dieng H, Lochouarn L, Traore SF, Trape JF, Simondon F, Fontenille D. [Malaria transmission in the rural zone of Niakhar, Senegal]. *Trop Med Int Health* 1998, 3: 667-677

31. INDEPTH Network. Population, Health and Survival at INDEPTH Sites. Ottawa: IDRC, 2002

32. Mbogo CN, Mwangangi JM, Nzovu J, Gu W, Yan G, Gunter JT, Swalm C, Keating J, Regens JL, Shilulu JI, Githure JI, Beier JC. Spatial and temporal heterogeneity of *Anopheles* mosquitoes and *Plasmodium falciparum* transmission along the Kenyan coast. *Am J Trop Med Hyg* 2003, 68: 734-742

33. English M, Muhoro A, Aluda M, Were S, Ross A, Peshu N. Outcome of delivery and cause-specific mortality and severe morbidity in early infancy: a Kenyan district hospital birth cohort. *Am J Trop Med Hyg* 2003, 69: 228-232

34. Bockarie M, Service MW, Barnish G, Maude G, Greenwood BM. Malaria in a rural area of Sierra Leone. III. Vector ecology and disease transmission. *Ann Trop Med Parasitol* 1994, 88: 251-262

35. Barnish G, Maude G, Bockarie M, Eggelte TA, Greenwood BM, Ceesay S. Malaria in a rural area of Sierra Leone. I. Initial results. *Ann Trop Med Parasitol* 1993, 87: 125-136

36. Trape JF, Pison G, Preziosi MP, Enel C, Desgrées du Lou A, Delaunay V, Samb B, Lagarde E, Molez JF, Simondon F. Impact of chloroquine resistance on malaria mortality. *CR Acad Sci III* 1998, 321: 689-697

37. Pison G, Trape JF, Lefebvre M, Enel C. Rapid decline in child mortality in a rural area of Senegal. *Int J Epidemiol* 1993, 22: 72-80

38. Premji Z, Ndayanga P, Shiff C, Minjas J, Lubega P, MacLeod J. Community based studies on childhood mortality in a malaria holoendemic area on the Tanzanian coast. *Acta Trop* 1997, 63: 101-109

39. Beier JC, Oster CN, Onyango FK, Bales JD, Sherwood JA, Perkins PV, Chumo DK, Koech DV, Whitmire RE, Roberts CR. *Plasmodium falciparum* incidence relative to entomologic inoculation rates at a site proposed for testing malaria vaccines in western Kenya. *Am J Trop Med Hyg* 1994, 50: 529-536

40. Spencer HC, Kaseje DC, Mosley WH, Sempebwa EK, Huong AY, Roberts JM. Impact on mortality and fertility of a community-based malaria control programme in Saradidi, Kenya. *Ann Trop Med Parasitol* 1987, 81 Suppl 1: 36-45

41. Robert V, Carnevale P, Ouedraogo V, Petrarca V, Coluzzi M. [Transmission of human malaria in a savanna village of South-West Burkina Faso]. *Ann Soc Belg Med Trop* 1988, 68: 107-121

42. Duboz P., Vaugelade J, Debouverie M. Mortalité dans l'enfance dans la région de Niangoloko. 1989. Ouagadougou, ORSTOM

43. Delacollette C, Barutwanayo M. [Mortality and morbidity at young ages in a stable hyperendemic malaria region, community Nyanza-Lac, Imbo South, Burundi]. *Bull Soc Pathol Exot* 1993, 86: 373-379

44. Desgrées du Lou A, Pison G, Aaby P. Role of immunizations in the recent decline in childhood mortality and the changes in the female/male mortality ratio in rural Senegal. *Am J Epidemiol* 1995, 142: 643-652

45. Smith T, Charlwood JD, Kihonda J, Mwankusye S, Billingsley P, Meuwissen J, Lyimo E, Takken W, Teuscher T, Tanner M. Absence of seasonal variation in malaria parasitaemia in an area of intense seasonal transmission. *Acta Trop* 1993, 54: 55-72

46. Armstrong-Schellenberg JR, Abdulla S, Minja H, Nathan R, Mukasa O, Marchant T, Mponda H, Kikumbih N, Lyimo E, Manchester T, Tanner M, Lengeler C. KINET: a social marketing programme of treated nets and net treatment for malaria control in Tanzania, with evaluation of child health and long-term survival. *Trans R Soc Trop Med Hyg* 1999, 93: 225-231

47. Appawu M, Owusu-Agyei S, Dadzie S, Asoala V, Anto F, Koram K, Rogers W, Nkrumah F, Hoffman SL, Fryauff DJ. Malaria transmission dynamics at a site in northern Ghana proposed for testing malaria vaccines. *Trop Med Int Health* 2004, 9: 164-170

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

49. Salum FM, Wilkes TJ, Kivumbi K, Curtis CF. Mortality of under-fives in a rural area of holoendemic malaria transmission. *Acta Trop* 1994, 58: 29-34

50. Molineaux L, Gramiccia G. The Garki Project. Geneva: World Health Organisation, 1980

51. Cot M, Roisin A, Barro D, Yada A, Verhave J-P, Carnevale P, Breart G. Effect of chloroquine chemoprohylaxis during pregnancy on birth weight: results of a randomised trial. *Am J Trop Med Hyg* 1992, 46: 21-27

52. Cot M, Le Hesran JY, Mialhes P, Esveld M, Etya'ale D, Breart G. Increase of birth weight following chloroquine chemoprophylaxis during the first pregnancy: results of a randomized trial in Cameroon. *Am J Trop Med Hyg* 1995, 53: 581-585

53. Fleming AF, Ghatoura GBS, Harrison KA, Briggs ND, Dunn DT. The prevention of anaemia in pregnancy in primigravidae in the guinea savanna of Nigeria. *Ann Trop Med Parasitol* 1986, 80: 211-233

54. Hamilton PJS, Gebbie DAM, Wilks NE, Lothe F. The role of malaria,folic acid and haemoglobin AS in pregnancy at Mulago hospital. *Trans R Soc Trop Med Hyg* 1972, 66: 594-602

55. Menendez C, Todd J, Alonso PL, Lulat S, Francis N, Greenwood BM. Malaria chemoprophylaxis, infection of the placenta and birth weight in Gambian primigravidae. *J Trop Med Hyg* 1994, 97: 248

56. Morley D, Woodland M, Cuthbertson WFJ. Controlled trial of pyrimethamine in pregnant women in an African village. *BMJ* 1964, 1: 667-668

57. Ndyomugyenyi R, Magnussen P. Chloroquine prohylaxis, iron-folic acid supplementation or case management of malaria attacks in primigravidae in western Uganda: effects on maternal parasitaemia and haemoglobin levels and on birthweight. *Trans R Soc Trop Med Hyg* 2000, 94: 413-418

58. Parise ME, Ayisi JG, Nahlen BL, Schultz LJ, Roberts JM, Misore A, Muga R, Oloo AJ, Steketee RW. Efficacy of sulfadoxine-pyrimethamine for prevention of placental malaria in a n area of Kenya with a high prevalence of malaria and human immunodeficiency virus infection. *Am J Trop Med Hyg* 1998, 59: 813-822

59. Shulman CE, Dorman EK, Cutts F, Kawuondo K, Bulmer JN, Peshu N, Marsh K. Intermittent sulphadoxine-pyrimethamine to prevent severe anaemia secondary to malaria in pregnancy: a randomised placebo-controlled trial. *Lancet* 1999, 353: 632-636

60. Mutabingwa T, Malle L, De Goos GA, Oosting J. Malaria chemosuppression in pregnancy. II: its effect on maternal haemoglobin levels, placental malaria and birthweight. *Trop Geogr Med* 1993, 45: 49-55

61. Steketee RW, Wirima JJ, Hightower AW, Slutsker L, Heymann DL, Breman JG. The effect of malaria and malaria prevention in pregnancy on offspring birthweight, prematurity, and intrauterine growth retardation. *Am J Trop Med Hyg* 1996, 55: 33-41

62. Verhoeff FH, Brabin BJ, Chimsuku L, Kazembe P, Russell WB, Broadhead RL. An evaluation of the effects of intermittent sulfadoxine-pyrimethamine treatment in pregnancy on parasite clearance and risk of low birthweight in rural Malawi. *Ann Trop Med Parasitol* 1998, 92: 141-150

63. Challis K, Osman N, Cotiro M, Nordahl G, Dgedge M, Bergström S. Impact of a double dose of sulphadoxine-pyrimethamine to reduce prevalence of pregnancy malaria in southern Mozambique. *Trop Med Int Health* 2004, 9: 1066-1073

64. Nosten F, Rogerson S, Beeson JG, McGready R, Mutabingwa TK, Brabin B. Malaria in pregnancy and the endemicity spectrum: what can we learn? *Trends Parasitol* 2004, 20: 425-432

65. Menendez C, Ordi J, Ismail MR, Ventura PJ, Aponte J, Kahigwa E, Font F, Alonso PL. The impact of placental malaria on gestational age and birth weight. *J Infect Dis* 2000, 181: 1745

66. Craig AG. Pregnancy-associated malaria--on the brink? *Trends Parasitol* 2004, 20: 201-204

67. Smith JD, Deitsch KW. Pregnancy-associated malaria and the prospects for syndrome-specific antimalaria vaccines. *J Exp Med* 2004, 200: 1093-1097

68. Hviid L. The immuno-epidemiology of pregnancy-associated *Plasmodium falciparum* malaria: a variant surface antigen-specific perspective. *Parasite Immunol* 2004, 26: 477-486

69. D'Alessandro U, Langerock P, Bennett S, Francis N, Cham K, Greenwood BM. The impact of a national impregnated bed net programme on the outcome of pregnancy in primigravidae in The Gambia. *Trans R Soc Trop Med Hyg* 1996, 90: 487-492

70. Dolan G, ter Kuile F, Jacoutot V, White N, Luxemburger C, Malankirii L, Chongsuphajaisiddhi T, Nosten F. Bed nets for the prevention of malaria and anaemia in pregnancy. *Trans R Soc Trop Med Hyg* 1993, 87: 620-626

71. Browne E, Maude G, Binka F. The impact of insecticide-treated bednets on malaria and anaemia in Kassena-Nankana district, Ghana: a randomized controlled trial. *Trop Med Int Health* 2001, 6: 667-676

72. ter Kuile F, Terlouw D, Phillips-Howard P, Hawley W, Friedman J, Kariuki S, Shi Y, Kolczak M, Lal A, Vulule J, Nahlen B. Reduction of malaria during pregnancy by permethrin-treated bed nets in an area of intense perennial malaria transmission in Western Kenya. *Am J Trop Med Hyg* 2003, 68 (Suppl 4): 50-60

73. Wilcox AJ. On the importance--and the unimportance--of birthweight. *Int J Epidemiol* 2001, 30: 1233-1241

74. Umbach DM, Wilcox AJ. A technique for measuring epidemiologically useful features of birthweight distributions. *Stat Med* 1996, 15: 1333-1348

75. Brabin BJ, Agbaje SOF, Ahmed Y, Briggs ND. A birthweight nomogram for Africa, as a malaria control indicator. *Ann Trop Med Parasitol* 1999, 93: S43-S57.

76. Steketee RW, Wirima JJ, Slutsker L, Heymann DL, Breman JG. The problem of malaria and malaria control in sub-Saharan Africa. *Am J Trop Med Hyg* 1996, 55: 2-7

77. Bouvier P, Breslow N, Doumbo O, Robert C-F, Picquet M, Mauris A, Dolo A, Dembele HK, Delley V, Rougemont A. Seasonality,malaria, and impact of prophylaxis in a West African village II Effect on birthweight. *Am J Trop Med Hyg* 1997, 56: 384-389

78. World Health Organisation, UNAIDS. AIDS epidemic update: December 2004. 2004.

79. van Eijk AM, Ayisia JG, ter Kuile FO, Misore AO, Otieno JA, Rosen DH, Kager PA, Steketee RW, Nahlen BL. HIV increases the risk of malaria in women of all gravidities in Kisumu, Kenya. *AIDS* 2003, 17: 595-603

80. Verhoeff FH, Brabin BJ, Hart CA, Chimsuku L, Kazembe P, Broadhead RL. Increased prevalence of malaria in HIV-infected pregnant women and its implications for malaria control. *Trop Med Int Health* 1999, 4: 5-12

81. ter Kuile FO, Parise ME, Verhoeff FH, Udhayakumar V, Newman RD, van Eijk AM, Rogerson SJ, Steketee RW. The burden of co-infection with human immunodeficiency virus type 1 and malaria in pregnant women in sub-Saharan Africa. *Am J Trop Med Hyg* 2004, 71 (Suppl 2): 41-54

82. Brocklehurst P, French R. The association between maternal HIV infection and perinatal outcome: a systematic review of the literature and meta-analysis. *Br J Obstet Gynaecol* 1998, 836-848

83. Shulman CE, Dorman EK. Importance and prevention of malaria in pregnancy. *Trans R Soc Trop Med Hyg* 2003, 7: 30-35

84. Carneiro I, Smith T, Lusingu J, Malima R, Utzinger J, Drakeley C. Modeling the relationship between the population prevalence of *Plasmodium falciparum* malaria and anemia. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 82-89

Chapter 5

Relationships between host infectivity to mosquitoes and asexual parasite density in *Plasmodium falciparum*

Amanda Ross, Gerry Killeen and Thomas Smith

Swiss Tropical Institute, Basel, Switzerland
Ifakara Health Research and Development Centre, Ifakara, Tanzania

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5.1 Summary

We describe a statistical model for the relationship between asexual parasite densities of *Plasmodium falciparum* and the infectivity of the host to mosquitoes. The model takes into account the delay between asexual parasitaemia and infectivity resulting from the time course of gametocytaemia. It also allows for the need for the blood meal to contain gametocytes of both sexes if infection is to take place. We show that by fitting this model to data from malariatherapy patients it can explain observed patterns of infectiousness of the human host and is consistent with distributions of gametocyte densities in malariatherapy patients.

By integrating this model into an individual-based simulation of human populations exposed to endemic *P. falciparum* transmission, we are able to predict the contributions of different host age groups to the infectious reservoir. Comparison of model predictions with published estimates of this quantity confirms that infected adults hosts are likely to make a significant contribution to the reservoir of transmission, and points to the need for improved population-based estimates of this age-dependence in infectivity of humans in endemic areas.

5.2 Introduction

Transmission of malaria from humans to the vector occurs when a female anopheline imbibes the sexual stages of the parasite (gametocytes) that arise as a result of developmental switching of a small proportion of the population of erythrocytic parasites. However not all anophelines feeding on gametocyte-infected hosts become infected and cryptic gametocytaemia can result in mosquito infections.[1,2] Oocyst rates in mosquitoes in endemic areas can be similar regardless of whether gametocytes or trophozoites are detected in the human host. Some studies have found little or no relationship of infectivity with gametocyte densities[3,4] while others found these to be related.[5,6] In artificially-induced infections, on average, infectivity to vectors rapidly reaches a persistent low but non-zero level as the infection proceeds.[7] However the decrease in infectivity is by no means as steep as that in measured gametocyte densities. There is a general decline in infectiousness with age but even highly immune hosts contribute to the infectious reservoir.[8,9]

Community effects of vaccines or of other interventions targeting the human host (herd immunity) result either directly or indirectly from changes in infectivity. To understand these, mathematical models are needed. Infectivity of the human host is a key determinant of the intensity of malaria transmission, but in contrast to field data, models of malaria transmission dynamics have generally assumed that the probability of transmission to a vector is equal across all infected humans and throughout the course of infection,[10] or that complete transmission-blocking immunity is acquired as a result of repeated exposure.[11]

As one component of a project to develop simulation models for the potential impact of a malaria vaccine, we now develop a new model for infectivity. We avoid directly incorporating the poorly understood processes of gametocytogenesis and the subsequent fate of gametocytes. Instead, we implicitly model gametocytogenesis by estimating infectivity from analyses of the association between lagged asexual blood stage densities and infectivity. We use data from deliberate infection of human subjects with *Plasmodium falciparum* between 1940 and 1963 as a treatment for tertiary syphilis[7,12] and fit a statistical model to these data to estimate the relationship between recent asexual parasitaemia and infectivity.

5.3 Methods

5.3.1 Model for infectivity as a function of parasite density

We define $\Upsilon(i,t)$ as the density of asexual parasites in patient *i* that gave rise to the gametocytes present at time *t*. Our stochastic simulation model of malaria epidemiology[13,14] uses five-day time steps. To incorporate transmission to the vector into this model, we propose that $\Upsilon(i,t)$ should be estimated as a weighted sum of the asexual parasite densities measured 10, 15 and 20 days prior to the feeding experiment. Thus if β_1, β_2 and β_3 are constants, then

$$\Upsilon(i,t) = \beta_1 Y(i,t-10) + \beta_2 Y(i,t-15) + \beta_3 Y(i,t-20) \qquad (5.1)$$

This quantity is constrained to be zero when there is no history of patent infection in the last 20 days, or when patent infection first arose in the last five-day time period. We justify the choice of the lag periods of 10, 15 and 20 days by observing that previous analyses of gametocyte dynamics in the same malariatherapy patients suggested that gametocytes spend an average of 7.4 days sequestered before appearing in the peripheral circulation and have a subsequent mean circulation time of 6.4 days.[15]

We considered a number of models for infectivity. In our preferred model, the ratio of functional female gametocytes per blood meal to $\Upsilon(i,t)$ is log normally distributed, with some geometric mean, ρ, so that the density of functional female gametocytes in the host blood, $y_g(i,t)$, is related to $\Upsilon(i,t)$ by

$$\ln(y_g(i,t)) \sim \text{Normal}(\ln(\rho\Upsilon(i,t)),\sigma_g^2) \qquad (5.2)$$

For a mosquito to become infected with *P. falciparum,* it must imbibe at least one female and one male gametocyte in the same blood meal. We assume that functional gametocytes are those able to give rise to sporozoites. We define y_g^* to be the density of female gametocytes necessary for infection of the mosquito, corresponding to 1

functional gametocyte per blood meal. The probability that at least one functional female gametocyte is taken up is then $\Pr(y_g(i,t) > y_g^*)$ and it follows that

$$\begin{aligned}\Pr(y_g(i,t) > y_g^*) &= \Phi\left[\frac{\ln\left(\rho\Upsilon(i,t)\right) - \ln(y_g^*)}{\sigma_g}\right] \\ &= \Phi\left[\frac{\ln\left(\Upsilon(i,t)\right)}{\sigma_g} + \rho^*\right]\end{aligned} \tag{5.3}$$

where Φ is the percentile of the cumulative standard normal distribution, and $\rho^* = \left(\ln(\rho) - \ln\left(y_g^*\right)\right)/\sigma_g$ is a constant which depends on the volume of the blood meal, the viability of the gametocytes, and the variability in the system. If the volume of the blood meal is assumed to be $3\,\mu l$ [16] (and there is assumed to be no concentration of erythrocytes in these species[17,18]), then the ratio of the functional female gametocyte density to $\Upsilon(i,t)$ is given by $\rho/3$.

If the sex-ratio of functional male to female gametocytes is 1:1, then the probability that at least one functional male gametocyte is taken up is also $\Pr(y_g(i,t) > y_g^*)$, and the probability that a mosquito is infected with both male and female gametocytes is $\Pr(y_g(i,t) > y_g^*)^2$. The number of mosquitoes out of a batch of $n_{fed}(i,t)$ mosquitoes fed on patient *i*, at time *t*, that are infected follows a Binomial distribution

$$n_{inf}(i,t) \sim Binomial\left(n_{fed}(i,t), \Pr(y_g(i,t) > y_g^*)^2\right) \tag{5.4}$$

Our estimate of the proportion of mosquitoes feeding on individual *i* at time point *t* that would become infected, $I_m(i,t)$, is then the expected proportion from this binomial

$$I_m(i,t) = E\left[\frac{n_{inf}(i,t)}{n_{obs}(i,t)}\right]. \tag{5.5}$$

5.3.2 Data and model fitting

We fitted the model given by equations 5.1-5.5 to archive data provided by Dr. W. Collins (Centers for Disease Control, Atlanta) collected by the United States Public Health Service in South Carolina and Georgia between 1940 and 1963. At that time, malariatherapy was a recommended treatment of neurosyphilis.[12] We obtained archive data for 392 patients inoculated with varying species, strains and parasite stages of *Plasmodium.* In each case, densities of asexual parasitaemia and of gametocytaemia were recorded on a daily basis. Batches of between 5 and 60 caged *Anopheles quadrimaculatus* or *An. albimanus* mosquitoes were allowed to feed on gametocytaemic

patients and then kept to allow oocysts to develop in the midgut before dissection to record infection status. Details of the methods have been previously published.[12]

We excluded patients inoculated or co-inoculated with *Plasmodium* species other than *P. falciparum* and those who were inoculated for a second time or who were treated with any of the antimalarial drugs used and those with insufficient data. We initially explored various possible predictors of the proportion of the mosquitoes that became infected. These comprised day since first parasitaemia, fever, the concurrent asexual parasite density, and the history of parasitaemia for the preceding 40 days. To maintain compatibility with our population model of asexual parasitaemia,[13] we simplified the history by considering parasite density only at five-day intervals.

The model described by equations 5.1 - 5.5 was selected as the most appropriate on the basis of these exploratory analyses. We treated it as a hierarchical Bayesian model, and fitted it using the Metropolis-Hastings algorithm in the program WinBUGS version 1.4.[19] β_1 was fixed at a value of 1 and we used imprecise log normal prior distributions for β_2 and β_3, and a gamma prior for $1/\sigma_g^2$.

5.3.3 Age-pattern of infectivity

Field estimates of the relative contribution to the infectious reservoir by different age groups have been made for settings in Liberia,[9] The Gambia,[8] Tanzania,[8] western Kenya,[20] Papua New Guinea[21] and Cameroon.[22] The estimates from Africa are based on feeding of insectary *An. gambiae*, while those from Papua New Guinea used a colony of *An. farauti.*

To validate our model against these data we simulated the epidemiology of malaria in field settings with entomological data using our stochastic simulation model.[13,14,23] This model makes predictions of parasite densities using a five-day time step assuming an annually recurring stable pattern of transmission as input. We used this model to implement an individual-based simulation of a population of at least 10,000 simulated individuals exposed from birth to the local transmission patterns. For each individual and time point during the year we use equations 5.1-5.5 to make predictions of $I_m(i,t)$.

There is evidence that the risk of being bitten by an anopheline is approximately proportional to body size.[24] To allow for this, we make an estimate of the contribution to the infectious reservoir for age group *j*, weighted proportionately to $A(a(i,t))$, the estimated body surface area of the host, where $a(i,t)$ is the age of individual *i* at time *t*.[14] The proportion of infectiousness contributed by group *j* is then

$$= \frac{\sum_{i,t}\left(A(a(i,t))I_m(i,t)J_j(i,t)\right)}{\sum_{i,t}\left(A(a(i,t))I_m(i,t)\right)}$$

where $J_j(i,t) = 1$ if individual *i* is in age group *j* at time *t*, and $J_j(i,t) = 0$ otherwise, and where the summations are over the whole simulated population and a whole year of follow-up time.

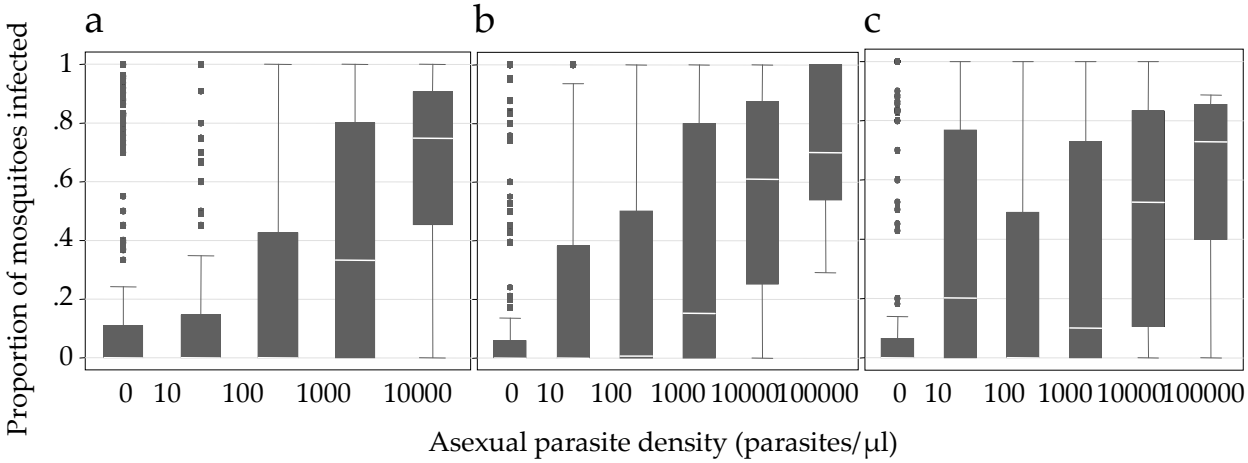
5.4 Results

5.4.1 Data description

Seventy one patients with 730 feeding experiments (median of 4 experiments per individual (90% central range = 1-29) were included in the analyses. A total of 22,431 mosquitoes were analyzed (median of 28 mosquitoes per experiment, 90% central range = 5-60).

A total of 562 (77%) of blood slides were positive for *P. falciparum* 10 days prior to the feeding experiments, with median density 1,792 parasites/μL (90% central range=30-25,990). Parasite prevalence and densities on the other days analysed were comparable. A total of 565 (77%) of the 730 samples on the day of the feeding experiment were gametocytaemic, with median gametocyte densities of 110 (90% central range 10-3330). Overall, 5420 (24%) out of 22431 mosquitoes became infected. However there was considerable variation in the observed proportions of mosquitoes infected in any one experiment (Figure 5.1). For the simulation model, we aimed to describe the average relationship between asexual parasite density and infectivity as accurately as possible for all values of the parasitaemia history.

Figure 5.1. Boxplot of the proportion of mosquitoes infected in the experiments for each category of asexual parasitaemia
(a) 10 days previously, (b) 15 days previously, (c) 20 days previously



The box indicates the 25th and 75th percentiles and the central line is the median. The upper whisker extends to the largest value below the 75th percentile plus the box height multiplied by 1.5. Similarly for the lower whisker. Values outside the whiskers are plotted individually.

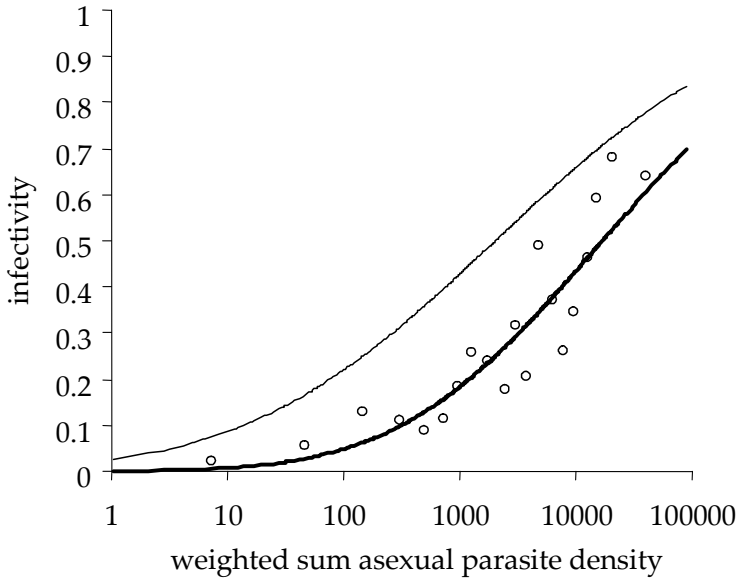
In analyses that considered only single predictors of the proportion of mosquitoes infected, asexual parasitaemia 20, 15, 10 and 5 days previously and the reciprocal of days since first parasitaemia were found to be most closely related to the proportion of mosquitoes that are infected. In multivariable models, the effects of parasite density five days previously and the reciprocal of time since first asexual parasitaemia did not improve the fit.

5.4.2 Model for infectivity

We rejected a number of models for infectivity before selecting the model given by equations 5.1 to 5.5. Fixed effects models that ignored variation between patients or samples in the relationship predicted very high proportions of infected mosquitoes for hosts with a history of high parasite densities. There is substantial variation in infectivity in the data even at high values of the weighted sum of asexual parasitaemia $\Upsilon(i,t)$ (Figure 5.1) and models with fixed effects of the weighted sum of asexual parasitaemia gave a better fit only if there was allowance for overdispersion in infectivity. However allowing for overdispersion in the response led to problems of convergence, with the estimates of random effects highly correlated with those of $\Upsilon(i,t)$.

Our preferred model (equations 5.1 to 5.5) introduces random variation into the explanatory variable, $\Upsilon(i,t)$, intended to simulate random variation in the process of gametocytogenesis. The fit was good (Figure 5.2 and Table 5.1) except at very high values of the weighted sum of asexual density $\Upsilon(i,t)$. There is little data at these high values, and although proportions of 100% infected mosquitoes were observed in almost one-third of the experiments with the highest 5% of values of $\Upsilon(i,t)$, there was also considerable variation, with some proportions as low as 17%. This variation coupled with the relatively small amount of data makes it difficult to specify a model which fits well in this range.

Figure 5.2. Relationship of infectivity to weighted sum of asexual densities , $\Upsilon(i,t)$



The thin line represents the estimated probability that the mosquito is infected with a female gametocyte, $\Pr(y_g(i,t) > y_g^*)$, and the thick line is the estimated probability of infection with both male and female gametocytes, $\Pr(y_g(i,t) > y_g^*)^2$. Circles represent the mean proportion of mosquitoes infected within categories of the weighted asexual parasite sum.

Table 5.1. Parameter estimates

	Description	Point Estimate	95% Credible interval
β_1	Effect of asexual density (lag 10 days) on expected gametocytaemia	1	Fixed*
β_2	Effect of asexual density (lag 15 days) on expected gametocytaemia	0.46	0.38 – 0.55
β_3	Effect of asexual density (lag 20 days) on expected gametocytaemia	0.17	0.13 – 0.23
ρ	Location parameter for the distribution of the ratio of gametocytes to asexual parasites	0.00031	0.00027 - 0.00036
σ_g	Standard deviation of the distribution of the ratio of gametocytes to asexual parasites	3.91	3.72 – 4.10

*To ensure identifiability

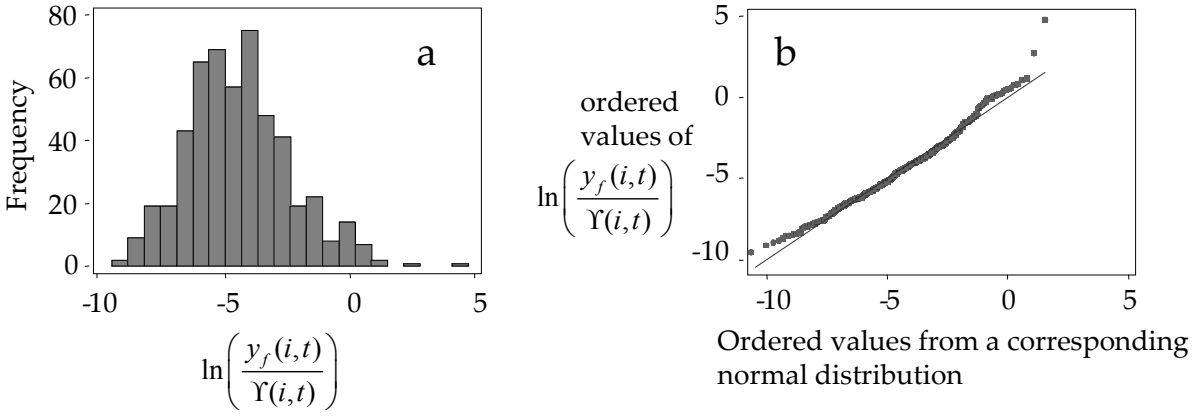
5.4.3 Gametocyte densities

From equation 5.2 it follows that

$$\ln\left(\frac{y_g(i,t)}{\Upsilon(i,t)}\right) \sim \text{Normal}(\ln(\rho), \sigma_g^2) \qquad (5.6)$$

To test this assumption of log-normality, we plotted the logarithm of the ratio of the observed density of female gametocytes $y_f(i,t)$ to the weighted sum of the asexual parasite densities (Figure 5.3a).

Figure 5.3. Distribution of the observed density of female gametocytes



a. Histogram of $\ln(y_f(i,t)/\Upsilon(i,t))$, the logarithm of the observed ratio of the density of female gametocytes to the weighted sum of asexual parasite densities.
b. Normal quantile plot of $\ln(y_f(i,t)/\Upsilon(i,t))$. For each data point (x, y), y is the observed value and x is the value expected for the same quantile of the corresponding normal distribution. Points lying along the straight line indicate that the data fits a normal distribution.

The observed distribution of this ratio is somewhat truncated because the limit of detection in the malariatherapy dataset was 10 gametocytes/μl. However, the normal quantile plot (Figure 5.3b) suggests that the log-normal approximation is roughly valid, with a mean of approximately -5 for $\ln\left(y_f(i,t)/\Upsilon(i,t)\right)$.

Thus, on average $y_f(i,t) \approx \Upsilon(i,t)\exp(-5)$, and the expectation of the normal distribution in equation 5.6 is

$$E\left[\ln\left(\frac{y_g(i,t)\exp(-5)}{y_f(i,t)}\right)\right] \approx \ln(\rho). \qquad (5.7)$$

Using our best estimate of ρ (Table 5.1), and scaling per microlitre, this gives an approximation for the proportion of gametocytes that are functional of $y_g(i,t)/(3y_f(i,t)) \approx \rho \exp(5)/3 \approx 0.015$.

5.4.4 Age pattern of infectivity

The field estimates of the relative contribution of different age groups to the infectious reservoir are presented in Table 5.2. Using the transmission patterns for Ifakara and Farafenni, we estimated the relative contributions from our model predictions. The field studies make the assumption that people of different ages are equally frequently bitten by mosquitoes. Thus the most comparable estimates are those not weighted by body surface area. The unweighted contributions that we predicted are shown for Ifakara, and these are similar to those from the field study.[8] Weighting the contributions of different ages of hosts to the infectious reservoir proportionately to the estimated surface area of the host as well as to the representation of the age-group in the population led to higher predicted contributions from adults.

Table 5.2. Estimated contributions of different age groups to the infectious reservoir for *P. falciparum*

	Site	Age group (years)					Details
		<1	1-4	5-9	10-19	≥20	
Field estimates of % contribution of different age groups*	Farafenni, The Gambia[8]	-	17.5	21.7	22.2	37.9	Membrane feeds of blood from gametocyte carriers
	Ifakara, Tanzania[8]	-	30.9	25.2	15.7	28.1	Membrane feeds of blood from gametocyte carriers
	Kano, Kenya[20]	-	38.1	33.9	11.7	16.3	Direct feeds on an unselected population sample
	Madang, Papua New Guinea[21]	-	21.0	30.7	28.3	6.0	Direct feeding of random population sample
	Madang, Papua New Guinea[21]	-	53.0	7.6	31.0	10.4	Direct feeding of gametocyte carriers
Model estimates for Ifakara (Idete)	Mean of $I_m(i,t)$ (%)	8.0	13.0	7.0	4.0	3.0	Assuming Ifakara (Idete) transmission pattern[25]
	% of total population	3.0	10.1	14.6	24.7	47.6	From Drakeley and others[8]
	Model-based estimate of % of infectious reservoir	4.8	26.3	20.5	19.8	28.6	Not weighted by body surface area
		1.7	14.2	16.3	25.0	42.9	Weighted by body surface area
	Model-based estimate of % of infectious reservoir	0.3	7.9	13.6	30.7	47.5	Assuming Farafenni transmission pattern[26] weighted by body surface area

* A population-based study in two villages in Cameroon[22] suggests a similar pattern with age and transmission intensity, using different age groups.

The studies of Drakeley and others[8] investigated infectivity using feeds only on blood from demonstrable gametocyte carriers, and this makes it difficult to be confident in comparing the results with our population-based estimates. The assumption that individuals without detectable gametocytes do not transmit to mosquitoes is questionable. However the estimated contributions of different age-groups were similar to those for Ifakara in settings where population-based assessments have been carried out. The general pattern in our simulations was in agreement with the data: at higher transmission (as in Kano, Kenya) the contribution of the younger age groups was increased, and at lower transmission (as in Farafenni, or in Papua New Guinea) the older age groups are predicted to be relatively more important.

5.5 Discussion

We have fitted a statistical model to describe the relationship between *P. falciparum* asexual parasite densities and infectivity. The model makes use of the requirement for infectivity, that both male and female gametocytes must occur in the same blood meal, to provide an explanation for the non-linear relationship between infectivity and gametocyte densities in the malariatherapy dataset.[7]

We developed this model specifically for inclusion in our mathematical model of populations exposed to endemic *P. falciparum* transmission, and this dictated some simplifications. To correspond to our simulations we used a five-day time step, which makes less use of the available data than would a model based on shorter time intervals. We also avoided directly fitting a model to gametocyte densities because we required a model for infectivity as a function of asexual parasite densities.

The latter simplification allowed us to avoid any strong assumptions about the problematical relationship between observed and functional gametocytaemia or about the poorly understood factors affecting gametocytogenesis.[27,28] Our examination of the distribution of gametocyte densities confirmed that our model for gametocyte densities is a reasonable approximation. Our estimate for the percentage of gametocytes that are functional is low, in agreement with estimates from life tables[29,30] which indicate that many parasites are killed between the macrogamete and oocyst stages.

We assumed a 50:50 ratio of functional male:female gametocytes. The median proportion of male gametocytes recorded in the malariatherapy dataset was 0.43 with a 90% central range of 0-1, while other studies have found lower proportions (approximately 0.25) of male gametocytes.[31-33] However male gametocytes produce more gametes than female gametocytes,[27] and thus perhaps y_g^* for male gametocytes should be lower than for female gametocytes, compensating for any imbalance in the sex ratio. We considered the sensitivity of $I_m(i,t)$, the proportion of mosquitoes feeding on individual i at time t who become infected, to variations in the ratio of functional male:female gametocytes. $I_m(i,t)$ showed little sensitivity to variations in the assumed sex ratio of gametocytes over a wide range. This supports the claim that the sex ratio of

gametocytes is unlikely to be of major importance in determining infectivity;[27] and studies to date have shown little or no effect of sex ratio on infectivity.[34-37]

We do not consider heterogeneity between individuals or parasite clones in their innate propensity to produce gametocytes because our interest focuses on overall infectivity. Moreover, we ignore any direct effects of fever on infectivity[38] and of drug treatment on gametocytaemia. These effects are likely to be of epidemiological importance mainly in low transmission areas.

To apply our model to endemic areas in Africa, we must make several further simplifications. The malariatherapy experiments used *An. quadrimaculatus* and *An. albimanus* in controlled conditions, which differ from African field conditions where the main vectors are *An. funestus* and the *An. gambiae* complex. We consider the extent to which our approach leads to an overall bias in the estimates of the proportion of mosquito bites resulting in infections of the vector in a separate paper.[39]

We also assume that the relationship between asexual parasitaemia and infectivity in adults infected for the first time can be generalized to those with varying levels of acquired immunity, which entails assuming that there is no acquisition of transmission-blocking immunity and no effect of antibodies to gametocytes. This is supported by our finding that the model predicts age patterns that concur with those from field data. Although children have often been thought to contribute most of the malaria transmission to vectors because of their higher parasite densities, those studies that have estimated the contributions of different ages of hosts to the infectious reservoir have found that adults make a substantial contribution (Table 5.2).

Our model supports the conclusions of these field studies and indeed suggests that the contribution of adults has been underestimated because of the usual assumption that each potential host is bitten equally frequently. This corroborates the assumption that transmission-blocking immunity does not markedly increase with age, and that naturally acquired immune responses to gametocytes are also not of epidemiological importance. However, these conclusions rest on a limited evidence base. There is a need for improved population-based estimates of age- and exposure- dependence in the infectivity of humans to mosquitoes in endemic areas.

5.A References

1. Muirhead-Thomson RC. Factors determining the true reservoir of infection of *Plasmodium falciparum* and *Wucheriria bancrofti* in a West African village. *Trans R Soc Trop Med Hyg* 1954, 48: 208-236

2. Haji H, Smith T, Meuwissen JT, Sauerwein R, Charlwood JD. Estimation of the infectious reservoir of *Plasmodium falciparum* in natural vector populations based on oocyst size. *Trans R Soc Trop Med Hyg* 1996, 90: 494-497

3. Haji H, Smith T, Charlwood JD, Meuwissen JH. Absence of relationships between selected human factors and natural infectivity of *Plasmodium falciparum* to mosquitoes in an area of high transmission. *Parasitol* 1996, 113: 425-431

4. Lensen A, Mulder L, Tchuinkam T, Willemsen L, Eling W, Sauerwein R. Mechanisms that reduce transmission of *Plasmodium falciparum* malaria in semiimmune and nonimmune persons. *J Infect Dis* 1998, 177: 1358-1363

5. Draper CC. Observations on the infectiousness of gametocytes in hyperendemic malaria. *Trans R Soc Trop Med Hyg* 1953, 47: 160-165

6. Tchuinkam T, Mulder B, Dechering K, Stoffels H, Verhave JP, Cot M, Carnevale P, Meuwissen JH, Robert V. Experimental infections of *Anopheles gambiae* with *Plasmodium falciparum* of naturally infected gametocyte carriers in Cameroon: factors influencing the infectivity to mosquitoes. *Trop Med Parasitol* 1993, 44: 271-276

7. Jeffery GM, Eyles DE. Infectivity to mosquitoes of *Plasmodium falciparum* as related to gametocyte density and duration of infection. *Am J Trop Med Hyg* 1955, 781-789

8. Drakeley CJ, Akim NI, Sauerwein R, Greenwood BM, Targett G. Estimates of the infectious reservoir of *Plasmodium falciparum* malaria in The Gambia and in Tanzania. *Trans R Soc Trop Med Hyg* 2000, 94: 472-476

9. Muirhead-Thomson RC. The malarial infectivity of an African village population to mosquitoes (*Anopheles gambiae*); a random xenodiagnostic survey. *Am J Trop Med Hyg* 1957, 6: 971-979

10. Macdonald G, Cuellar CB, Foll CV. The dynamics of malaria. *Bull WHO* 1968, 38: 743-755

11. Dietz K, Molineaux L, Thomas A. A malaria model tested in the African savannah. *Bull WHO* 1974, 50: 347-357

12. Collins WE, Jeffery GM. A retrospective examination of mosquito infection on humans infected with *Plasmodium falciparum*. *Am J Trop Med Hyg* 2003, 68: 366-371

13. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 19-31

14. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationships between the entomological inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 11-18

15. Eichner M, Diebner HH, Molineaux L, Collins WE, Jeffery GM, Dietz K. Genesis, sequestration and survival of *Plasmodium falciparum* gametocytes: parameter estimates from fitting a model to malariatherapy data *Trans R Soc Trop Med Hyg* 2001, 95: 497-501

16. Jeffery GM. Blood meal volume in *Anopheles quadrimaculatus*, *A. albimanus* and *Aedes aegypti. Exp Parasitol* 1956, 5: 371-375

17. Chege GM, Beier JC. Blood acquisition and processing by three *Anopheles* (Diptera: Culicidae) species with different innate susceptibilities to *Plasmodium falciparum*. *J Med Entomol* 1998, 35: 319-323

18. Vaughan JA, Noden BH, Beier JC. Concentration of human erythrocytes by anopheline mosquitoes (Diptera: Culicidae) during feeding. *J Med Entomol* 1991, 28: 780-786

19. WinBUGS. Cambridge, England: MRC-BSU, 2003.

20. Githeko AK, Brandling-Bennett AD, Beier M, Atieli F, Owaga M, Collins FH. The reservoir of *Plasmodium falciparum* malaria in a holoendemic area of western Kenya. *Trans R Soc Trop Med Hyg* 1992, 86: 355-358

21. Graves PM, Burkot TR, Carter R, Cattani J, Lagog M, Parker J, Brabin B, Gibson FD, Bradley DJ, Alpers MP. Measurement of malarial infectivity of human populations to mosquitoes in the Madang area, Papua, New Guinea. *Parasitol* 1988, 96: 251-263

22. Bonnet S, Gouagna LC, Paul RE, Safeukui I, Meunier JY, Boudin C. Estimation of malaria transmission from humans to mosquitoes in two neighbouring villages in south Cameroon: evaluation and comparison of several indices. *Trans R Soc Trop Med Hyg* 2003, 97: 53-59

23. Smith T, Killeen GF, Maire N, Ross A, Molineaux L, Tediosi F, Hutton G, Utzinger J, Dietz K, Tanner M. Mathematical modeling of the impact of malaria vaccines on the clinical epidemiology and natural history of *Plasmodium falciparum* malaria: Overview. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 1-10

24. Port GR, Boreham PFL, Bryan JH. The relationship of host size to feeding by mosquitos of the *Anopheles gambiae* Giles Complex (Diptera, Culicidae). *Bull Entomol Res* 1980, 70: 133-144

25. Charlwood JD, Smith T, Lyimo E, Kitua A, Masanja H, Booth M, Alonso PL, Tanner M. Incidence of *Plasmodium falciparum* infection in infants in relation to exposure to sporozoite-infected anophelines. *Am J Trop Med Hyg* 1998, 59: 243-251

26. Lindsay SW, Snow R, Broomfield GL, Janneh MS, Wirtz RA, Greenwood BM. Impact of permethrin-treated bednets on malaria transmission by the *Anopheles gambiae* complex in The Gambia. *Med Vet Entomol* 1989, 3: 263-271

27. Carter R, Graves PM. Gametocytes. In: Wernsdorfer WH, McGregor I, eds. Malaria. Principles and practice of malariology.1. Edinburgh: Churchill Livingstone, 1988: 253-306

28. McKenzie FE, Bossert WH. The dynamics of *Plasmodium falciparum* blood-stage infection. *J Theor Biol* 1997, 188: 127-140

29. Vaughan JA, Noden BH, Beier JC. Population dynamics of *Plasmodium falciparum* sporogony in laboratory-infected *Anopheles gambiae*. *J Parasitol* 1992, 78: 716-724

30. Vaughan JA, Noden BH, Beier JC. Sporogonic development of cultured *Plasmodium falciparum* in six species of laboratoy-reared *Anopheles* mosquitoes. *Am J Trop Med Hyg* 1994, 51: 233-243

31. Osgood SM, Eisen RJ, Schall JJ. Gametocyte sex ratio of a malaria parasite: experimental test of heritability. *J Parasitol* 2002, 88: 494-498

32. Paul REL, Coulson TN, Raibaud A, Brey PT. Sex determination in malaria parasites. *Science* 2000, 287: 128-131

33. Robert V, Sokhna CS, Rogier C, Ariey F, Trape JF. Sex-ratio of *Plasmodium falciparum* gametocytes in inhabitants of Dielmo, Senegal. *Parasitol* 2003, 127: 1-8

34. Noden BH, Beadle PS, Vaughan JA, Pumpini CB, Kent MD, Beier JC. *Plasmodium falciparum*: the population structure of mature gametocyte cultures has little effect on their innate fertility. *Acta Trop* 1994, 58: 13-19

35. Ponnudurai T, Meuwissen JTET, Leeuwenberg ADEM, Verhave JP, Lensen AHW. The production of mature gametocytes of *Plasmodium falciparum* in continuous cultures of different isolates infective to mosquitoes. *Trans R Soc Trop Med Hyg* 1982, 76: 242-250

36. Robert V, Read AF, Essong J, Tchuinkam T, Mulder B, Verhave JP, Carnevale P. Effect of gametocyte sex ratio on infectivity of *Plasmodium falciparum* to *Anopheles gambiae*. *Trans R Soc Trop Med Hyg* 1996, 90: 621-624

37. Boyd MF, Stratham-Thomas WK, Kitchen SF. On the relative susceptibility of *Anopheles quadrimaculatus* to *Plasmodium vivax* and *Plasmodium falciparum*. *Am J Trop Med* 1935, 15: 485-493

38. Sinden RE. Asexual blood stages of malaria modulate gametocyte infectivity to the mosquito vector - possible implications and control. *Parasitol* 1991, 103: 191-196

39. Killeen GF, Ross A, Smith T. Infectiousness of malaria-endemic human populations to vectors. *Am J Trop Med Hyg* 2006, 76 (Suppl 2): 38-45

Chapter 6

Modelling the impact of intermittent preventive treatment against malaria in infants

Amanda Ross, Melissa Penny, Nicolas Maire, Alain Studer, Ilona Carneiro, David Schellenberg, Brian Greenwood, Marcel Tanner, Thomas Smith

Swiss Tropical Institute, London School of Hygiene and Tropical Medicine
Ifakara Health Research and Development Centre

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6.1 Summary

Trials of intermittent preventive treatment against malaria in infants (IPTi) using sulphadoxine-pyrimethamine (SP) have shown promising results in reducing clinical malaria episodes. The impact of IPTi in different epidemiological settings and over time is unknown and predictions are hampered by the lack of knowledge about how IPTi works. We investigated mechanisms proposed for the action of IPTi and made predictions of the likely impact on morbidity and mortality.

We used a comprehensive model of malaria epidemiology to simulate recently published trials of IPTi using SP with site-specific characteristics as inputs. This baseline model was then modified to represent hypotheses concerning the duration of action of SP, the temporal pattern of fevers caused by individual infections, potential benefits of avoiding fevers on immunity and the effect of sub-therapeutic levels of SP on parasite dynamics. The baseline model reproduced the pattern of results reasonably well. None of the models based on alternative hypotheses improved the fit between the model predictions and observed data suggesting that the pattern of trial results can be accounted for by differences between the trial sites together with known features of malaria epidemiology.

Predictions suggest that IPTi would have a beneficial impact over a wide range of transmission intensities. IPTi was predicted to avert a greater number of episodes where IPTi coverage was higher, the health system treatment coverage lower, and for drugs which were more efficacious and had longer prophylactic periods. The predicted cumulative benefits were proportionately greater for severe malaria episodes and malaria-attributable mortality than for acute episodes in the settings modelled. Modest increased susceptibility was predicted between doses and following the last dose, but these were outweighed by the cumulative benefits. The impact on transmission intensity was negligible.

6.2 Introduction

Intermittent preventive treatment in infants (IPTi) involves giving antimalarial drugs at scheduled times during the first year of life, irrespective of whether the infants have malaria infections.[1] The limited number of doses is intended to retain the benefits of weekly or fortnightly chemoprophylaxis whilst avoiding the disadvantages: thus reducing malaria morbidity and mortality while minimising difficulties in sustainability, accelerating drug resistance or impairing the development of natural immunity.

IPTi trials to date have shown a strong, albeit variable, protective efficacy against clinical episodes of malaria in the first year of life.[2] How the impact of IPTi varies over time and in different epidemiological settings is unknown. Prediction is hampered by the lack of knowledge of both how IPTi works and the extent to which different trial characteristics may account for the variability in the observed estimates. Trial

characteristics which have been highlighted are levels of drug resistance, transmission intensity, seasonality, IPTi schedule, and other interventions for malaria control (such as insecticide-treated nets (ITN) and treatment coverage).[2-4] We use these characteristics as inputs to a stochastic simulation model of malaria epidemiology. We then modify this model to represent hypotheses that have been proposed for the mechanism of IPTi to investigate which of these hypotheses are consistent, and which cannot be reconciled, with the observed trial results. The hypotheses, defined in section 6.3, concern the duration of action of SP, the temporal pattern of fevers caused by individual infections, the potential benefits for acquired immunity of avoiding episodes and the effect of sub-therapeutic levels of SP on parasite dynamics. We then use the model which best fits our criteria to make predictions of the impact of IPTi in different epidemiological settings and with varying drug characteristics.

6.3 Methods

Model 1 (Baseline model): Model of malaria epidemiology taking into account between-trial differences

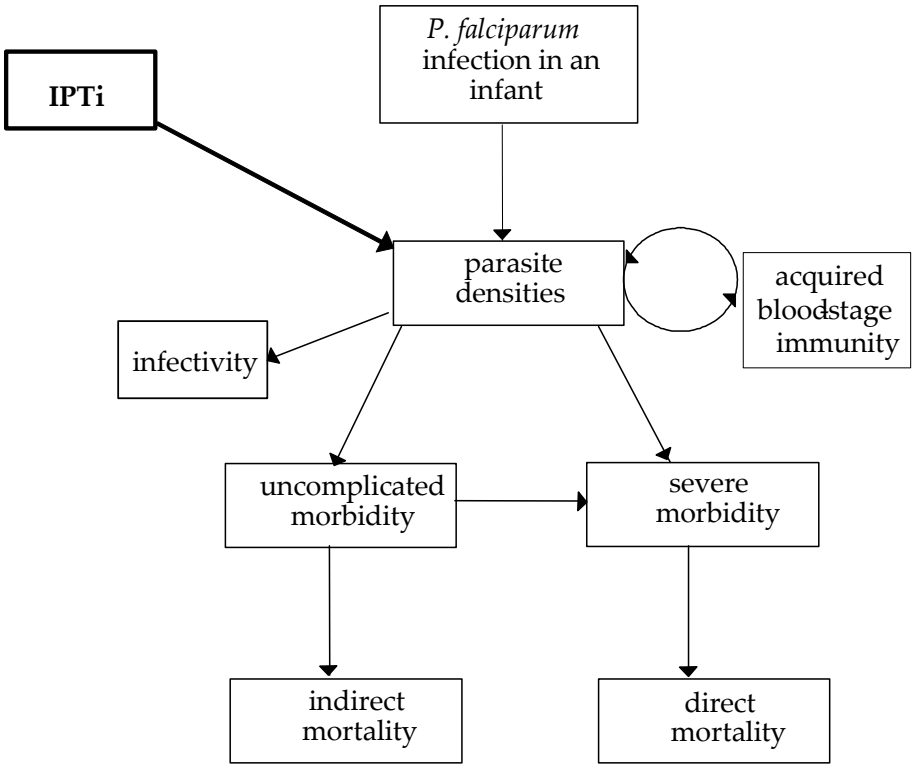
We combine our model of malaria epidemiology with an added component for the action of SP[5] and input the different trial characteristics such as transmission intensity and treatment coverage. This allows us to see if the between-trial differences in combination with this model can account for the heterogeneity in observed efficacy estimates.

Model for malaria epidemiology

The model is individual-based and stochastic, and is fully described elsewhere (Appendix A.1).[6] Briefly, there is a simulated population of individuals who are updated at five-day timesteps via model components representing new infections, parasite densities, acquired immunity, morbidity, mortality and infectivity to mosquitoes (Figure 6.1). The course of parasite densities over an infection are described by averaged empirical data (described in [7]). Immunity to asexual parasites is derived from a combination of cumulative exposure to both inoculations and parasite densities, and maternal immunity.[7] The inclusion of acquired immunity allows us to model potential effects of IPTi on immunity through loss of exposure. The probability of a clinical attack of malaria depends on the current parasite density and a pyrogenic threshold (described in Chapter 2). The pyrogenic threshold responds dynamically to recent parasite load, increasing or saturating through exposure to parasites and decaying with time, and thus is individual- and time- specific. Severe malaria can arise in two ways, either as a result of overwhelming parasite densities or through uncomplicated malaria with concurrent non-malaria co-morbidity (Chapter 3). Mortality can be either direct (following severe malaria) or indirect (uncomplicated malaria in conjunction with co-morbidity, or during the neonatal period as a result of

maternal infection (Chapter 3). The parameter values for this model were estimated by fitting to data from a total of 61 malaria field studies of various different aspects of malaria epidemiology,[8,9] and are given in Appendix A.1.

Figure 6.1 Simplified processes in the baseline model



Simulation of sulphadoxine-pyrimethamine and drug resistance

The benefits of SP depend on a combination of the drug concentration and the frequency of mutations conferring drug resistance present in the population,[5] however the exact time-course and killing action of SP is not known.[10] Hastings and Watkins quantify the chances of failing treatment with correct dosing for dihydrofolate reductase (*dhfr*) wildtype, 108, doubles, triples at 0, 0, 0, 50% respectively, while periods of preventive effect are 52, 12, 12, 2 days.[5] We simulate the action of SP according to these numbers rounded to the 5 day time steps used by the simulation model. Although *dhps* mutations have been isolated at the sites, they are not considered in this study.

Simulation of clinical episodes

The primary trial outcome was clinical episodes, defined as detected fever or history of fever together with parasitaemia, and infants were regarded as not at risk for the

following 21 days.[3] In our simulations, only fevers presenting for treatment were counted as episodes and the infant was classified as not at risk for the following 4 five-day periods.

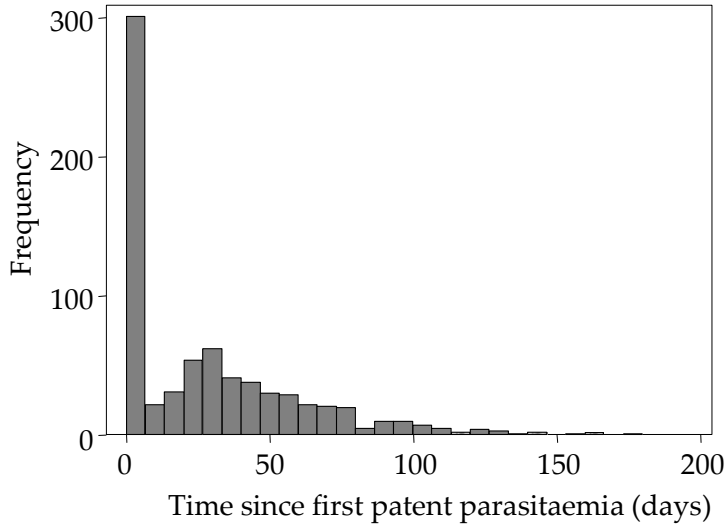
Model 2: Alternative time duration for SP action

The duration of the prophylactic period for SP is not well established. We vary the duration of SP action from the baseline model, which has a prophylactic period of 50 days for wildtype infections,[5] to 30 days. This alternative time period was chosen because drug concentrations of sulfadoxine and pyrimethamine alone decline log-linearly, but in combination they are synergistic and an isobologram suggests that there is a sharp drop in SP action after approximately a month.[11] Observations from field studies also suggest that the apparent effect of SP lasts for roughly one month.[12-14] Simulated infections are either sensitive or resistant, and the resistant infections are unaffected by drug treatment.

Model 3: The timing of episodes produced by a single infection

In non-immune adults inoculated with *P. falciparum* as treatment for neurosyphilis, untreated infections can persist for many months, during which clinical attacks recur at irregular intervals[15] (Figure 6.2). The infections cleared or prevented by IPTi would therefore have caused repeated fevers, some of which could have occurred 3 months or more after infection.

Figure 6.2 Timing of fevers* resulting from single infections in 334 neurosyphilis patients



*One fever counted per five-day interval. This data was collected by the United States Public Health Service in South Carolina and Georgia between 1940 and 1963 and was provided by Dr W Collins (Centers for Disease Control and Prevention, Atlanta, GA).

The timing of fevers is not well characterized by the baseline model which tends to produce too little variation, missing both early and late fevers. We therefore use an alternative, simple simulation model based on the empirical timing of fevers to examine whether the temporal pattern of fevers resulting from individual infections can account for the pattern of trial results for episodes.

Model 3 is different to the other models in that it is not based on model 1, other than the algorithms for the number of infections producing blood-stage parasites in each infant.[16] For each successful infection, we randomly selected one of the 334 malariatherapy patients' timing of fevers. Concurrent infections did not interact and there was no acquired immunity. SP was assumed to act in the same way as for the baseline model.

Model 4: High parasite densities may not be efficient for acquiring immunity

Overwhelming parasite densities may not contribute as much to the accrual of immunity as would the same total number of parasites experienced in smaller doses over a longer period of time. Such densities cause fever, and the fever itself may also hinder the acquisition of immunity, possibly through the loss of T- and B-cells**.** We modify the baseline model to reduce the contribution of parasite density to acquired immunity in the presence of a fever.

In the baseline model (model 1), immunity is modelled as a function of both the number of distinct infections that the individual has experienced and his or her cumulative parasite load. The cumulative exposure to parasites for individual *i* of age *a* at time *t*, $X_y(i,t)$, is defined as the cumulative sum of daily densities of asexual parasites/microlitre of blood since birth up to time *t*. This can be partitioned into the cumulative sum up to time *t-1*, the previous five-day time-step, and the sum of the densities over the last five days, $Y_5(i,t)$,

$$X_y(i,t) = \int_{t-a}^{t-1} Y(i,\tau) \quad d\tau \quad + \quad Y_5(i,t)$$

For model 4, we include a parameter β_f which fixes the contribution of the current density as $Y_5(i,t)$ if a malarial fever is absent $(\beta_{f=0} = 1)$, but may differ from this if a fever is present.

$$X_y(i,t) = \int_{t-a}^{t-1} Y(i,\tau) \quad d\tau \quad + \quad \beta_f Y_5(i,t)$$

We fitted the new parameter β_f to the same datasets used to fit model 1, simultaneously with the previously defined parameters.[6,8] These parameters estimates

are given in Appendix A.1. To calculate $X_y(i,j,t)$ to correspond to the published model,[7] we subtract the contribution of infection *j* to avoid double-counting.

Model 5: Surviving infections are attenuated by SP allowing extended low-level exposure beneficial to stimulating immunity

Waning drug concentrations or partial drug resistance may allow parasites to survive in the presence of SP whilst restricting their growth.[5] This may allow an extended time for the immune system to mount a response to the parasite, which could facilitate the development of immunity to malaria.[17,18] It is not known if attenuated infections can lead to enhanced immunity in this way, although there is some experimental data from mice that suggests that this may be possible.[19] Low levels of blood stage infection in humans can induce immunity.[20] We hypothesize that infections beginning when SP concentrations have decreased to sub-therapeutic concentrations have reduced densities and longer durations compared to when there is no SP, and that this enhances the development of immunity.

We modify the baseline model so that a simulated infection beginning within a window period after SP treatment has a longer duration and lower densities. The window period begins as the prophylactic action ends, and the duration depends on the *dhfr* mutations assigned to the simulated infection (wildtype: 10 days; 108/double/triple: 30 days). We simulate parasite densities in the same way as for the baseline model, except that we reduce all densities from the infection by a third and extend the duration by a factor of 3. This value was arbitrarily chosen to represent an upper limit for plausible values. The potential consequence of model 5 is to increase the amount of time that an infant has low-level parasitaemia, which in turn increases the time that the pyrogenic threshold is high.

Data sources: The field trials

A model of IPTi should capture the approximate time-course of efficacy of IPTi trials. The most detailed, standardised age-groups are those provided by a systematic analysis of six IPTi trials using SP[3] (Table 6.1). For practical reasons, we omit studies not included in this report.[21-24] All six included trials were carefully conducted and independently monitored.[3]

A critical input for the models is transmission intensity. Reported seasonal entomological inoculation rates (EIR) and/or age-prevalence curves were available from three trial sites (Manhiça, Ifakara and Navrongo),[13,25-27] but not for the remaining three (Tamale, Kumasi and Lambaréné).[28-30] Thus the formal comparison of models and empirical data was restricted to simulations from the former three trials, whilst the latter were used to validate model output against general patterns in the trial results.

Table 6.1. Study sites and trial characteristics

	Schellenberg *et al*[25,26]	Chandramohan *et al*[13]	Macete *et al*[27]	Kobbe et al[29]	Mockenhaupt et al[30]	Grobusch et al[28]
Study site	Ifakara, Tanzania	Navrongo, Ghana	Manhiça, Mozambique	Kumasi, Ghana	Tamale, Ghana	Lambaréné, Gabon
Pattern of seasonality[§]	Perennial	Marked seasonality	Perennial	Perennial	Perennial	Perennial
Study period	1999-2001	2000-2004	2002-2005	2003-2005	2003-2005	2002-2006
Transmission Intensity (Infectious bites/adult/year)	29[31] in 1999-2000	418[32] in 2001-02	38 in 2001-02	approx 400	NK (high)	Approx 50
ITN coverage	67%	17%	0%	20%	<1%	5%
Untreated net coverage			15%	20%	<1%	85%
Day 14 ACPR* (95% CI)	66% (55,76)[33]	78% (69, 85)[34]	83% (73, 90)[35]	‡	86%(79,91)[36]	79% (64,90) †[37]
Trial characteristics						
Primary outcome: Protective efficacy first dose to 12 mo[3]	58.8 (40.9, 71.3)	29.3 (17.3, 39.6)	20.1 (2.0, 34.9)	20.9 (8.9, 31.3)	33.3 (20.7, 43.9)	22.0 (-25.4, 51.5)
Number of infants enrolled (placebo/active)	351/350	1242/1243	755/748	535/535	600/600	595/594
Level of randomisation	Individual	Community	Individual	Individual	Individual	Individual
Schedule of IPTi doses (months)	2, 3 and 9	3, 4, 9 and 12	3, 4 and 9	3,9 and 15	3,9 and 15	3,9 and 15
Mean age at doses (months)	2.2, 3.3 and 9.2	3.0, 4.0, 9.5 and 12.6	3.3, 4.4, 9.4	2.8	2.4, 8.1, 14.3	3.1,9.3, 15.3
Coverage	100%, 95%, 84%	95%, 95%, 90%, 91%.	100%,96%,91%	100%,100%,99%	100%,98%,98%	
Method of case detection	Passive	Passive	Passive	Passive+Active (monthly)	Passive+Active (3-monthly)	Passive+Active (monthly)
First-line treatment	SP	CQ and SP	CQ or SP + AQ	Artesunate+AQ	Artesunate	Artesunate (+AQ)
Rescue treatment	quinine	quinine	quinine			
Routine iron supplementation	Yes	Yes	No	No	No	No

*ACPR=adequate parasitological cure rates in clinical cases (6 months – 5 years, or <5 years) †Children aged 1-10 years

ITN=insecticide treated net. NK=not know; [§] Roca-Feltrer *et al*, in prep

‡ 79% infants with triple *dhfr* and/or *dhps* mutations at IPTi-3 [29]

For some age-groups, efficacy estimates for episodes defined with two different parasite density cut-offs are available: fever plus parasitaemia of any density, and fever plus high parasite density. In most cases there is little difference.[3,13,25-27] However, a discrepancy arises in Navrongo[3,13] for children over one year of age where the estimated efficacy for high density episodes (≥5000/μl) suggests an increase in episodes in the IPTi group compared to the placebo group which is not apparent for episodes with parasitaemia of any density. In this case, we use the high density definition because it is likely to be more specific in a high transmission area and as age increases.[38,39]

Specifying model input values for the trial sites

Transmission intensity, seasonality and ITN use

We based our model inputs on the published data for seasonality and transmission intensities (Table 6.1). In Ifakara, the extensive coverage of insecticide-treated nets (ITNs) may have substantially decreased transmission from the reported EIR of 30 per year. Our baseline model does not explicitly include a component for the impact of ITNs, this is currently being implemented.[40] However the most relevant consequence for modelling trials of IPTi would be the reduction in transmission to the infants. It is likely that ITN use would also decrease onward transmission, but is not expected to alter the sporozoite load of an infectious mosquito, nor would a lower sporozoite load be likely to lead to less severe outcomes in humans.[41-43]

For the Ifakara trial, we gauged the effective overall EIR by comparing observed age-prevalence[44,45] and age-incidence curves for uncomplicated episodes[44] and malaria hospital admissions[46] to simulated age curves for a range of annual EIR values. The best-fitting age patterns were produced by an EIR of approximately 4. We also considered the effects of decreasing transmission intensity and reduced seasonality.[46] Decreasing transmission has been proposed as a possible explanation for the high protective efficacy estimates observed in the Ifakara trial.[47] For Manhiça and Navrongo, we did not adjust the overall EIR for ITN use. The inputs for the Manhiça field site have been previously characterized for the baseline model.[48] We validated our input EIR value for Navrongo by comparing the simulated age-prevalence curve against two sets of survey data.[7,49] In addition, we restricted the simulations for Ifakara to infants who reached 2 months of age between August and April in order to correspond to the recruitment period.

Treatment of clinical episodes

Only simulated fevers presenting for treatment were counted as episodes and the infant was classified as not at risk for the following 4 five-day periods to correspond to the trial definitions.[3] The proportion of malaria fevers that presented for treatment in

the trials is unknown. We estimated this proportion by assuming that fevers were treated with a constant probability. We adjusted this probability until our simulations of the time to first treated episode matched the published Kaplan-Meier curves for the placebo groups. The closest matches were found for Ifakara, Manhiça and Navrongo using 20%, 4% and 7% respectively. The value of 4% in infants in Manhiça was similar to the previous estimate of 5% for children 1-4 years in a vaccine trial.[48] The pattern of the estimates is plausible because the Ifakara study area was centred around a town with relatively good access to the health facility whereas the other trial settings were rural. We assumed that 48% of the severe episodes presented for treatment in all trials (Chapter 3).

Clinical episodes presenting for treatment were given SP in Ifakara and SP with chloroquine (CQ) in Navrongo (Table 6.1). In Manhiça, the national policy changed from CQ to amodiaquine (AQ) plus SP during the course of the trial. We simulate CQ and SP as clearing all infections, sensitivity analyses show that this assumption is not critical. The rescue treatment in the Ifakara, Manhiça and Navrongo trials was quinine which was given if the infant was admitted to hospital with malaria, presented within 2 weeks of an IPTi or placebo dose, or presented within 14 days of receiving SP. We simulate quinine as clearing all infections within a five-day time-step.

Frequencies of dhfr mutations

Each simulated infection was assigned a genotype (*dhfr* wildtype, 108 or double mutations, or triple mutations)**.** The frequency of *dhfr* mutations in each trial site is uncertain. Fourteen day adequate clinical and parasitological cure rates (ACPR) are available (Table 6.1), but they underestimate the true failure rate.[10,50] It is not possible to determine by how much the rate is underestimated for an individual site because the 14 day parasitological failure rates have a low predictive value.[50,51] Estimating *dhfr* genotype frequencies from data on the prevalence of mutations in infected humans is also difficult, because a combination of mutations may be formed in various ways when there are multiple infections. We aim to determine only whether the trial results can be reproduced for a reasonable assumed value of the frequency of *dhfr* mutations combined with the baseline model, and so we simulate the trials over a range of assumed frequencies. The lower bound of this range was provided by converting the lower confidence interval of the 14 day failure rates into *dhfr* genotype frequencies using simulations of the trials which had reported the 14 day failure rates. The value producing the best-fitting predictions within this range was chosen.

Scenarios used for predicting the impact of IPTi outside of the trial settings

We predicted age-specific protective efficacy and cumulative protective efficacy up to the age of four years.

We define the cumulative protective efficacy as $1-\left(\frac{c_i / pyar_i}{c_p / pyar_p}\right)$

where c is the cumulative number of episodes in the IPTi (i) or placebo (p) groups and *pyar* are the person-years at risk.

We also predicted the number of acute episodes, severe episodes and combined direct and indirect malaria deaths that would be averted for a period of 20 years following the introduction of IPTi in a population aged 0 to 90 years. We assumed a reference scenario with IPTi doses at 3, 4 and 9 months and then changed the values of different variables one by one to investigate their effects on the predicted impact (Table 6.2). The simulations were based on a population of 200,000 individuals, with an approximately stationary age-distribution matching that of the demographic surveillance site in Ifakara, Tanzania, in 1997-99.[52]

Table 6.2. Variables that vary between scenarios**

Variable	Description	Levels
Intensity of transmission	Infected bites per adult per year prior to the introduction of IPTi‡	High transmission: 200 Moderate transmission: 100 **Reference: 21** Low transmission: 6
Treatment coverage	Proportion of malaria fevers treated	**4%,** 30%
Drug resistance	Frequency of 3 different genotypes	100%, 0%, 0% **80%, 10%,10%** 20%, 40%, 40% 0%, 0%, 100%
Prophylactic period	Time in days that drug clears blood-stage infections for each of the 3 different genotypes †	0,0,0 days (treatment only) **50, 10, 0 d** (corresponds to SP) 100, 20, 0 days
IPTi schedule	Age at doses	**3, 4 and 9 months** Single doses 1.5-24 months*
IPTi coverage	Proportion of eligible infants receiving all 3 IPTi doses (coverage with first, second and third dose)	**89% (95%,95%,99%)** 50% (79%,79%,79%) 100% (100%,100%,100%)

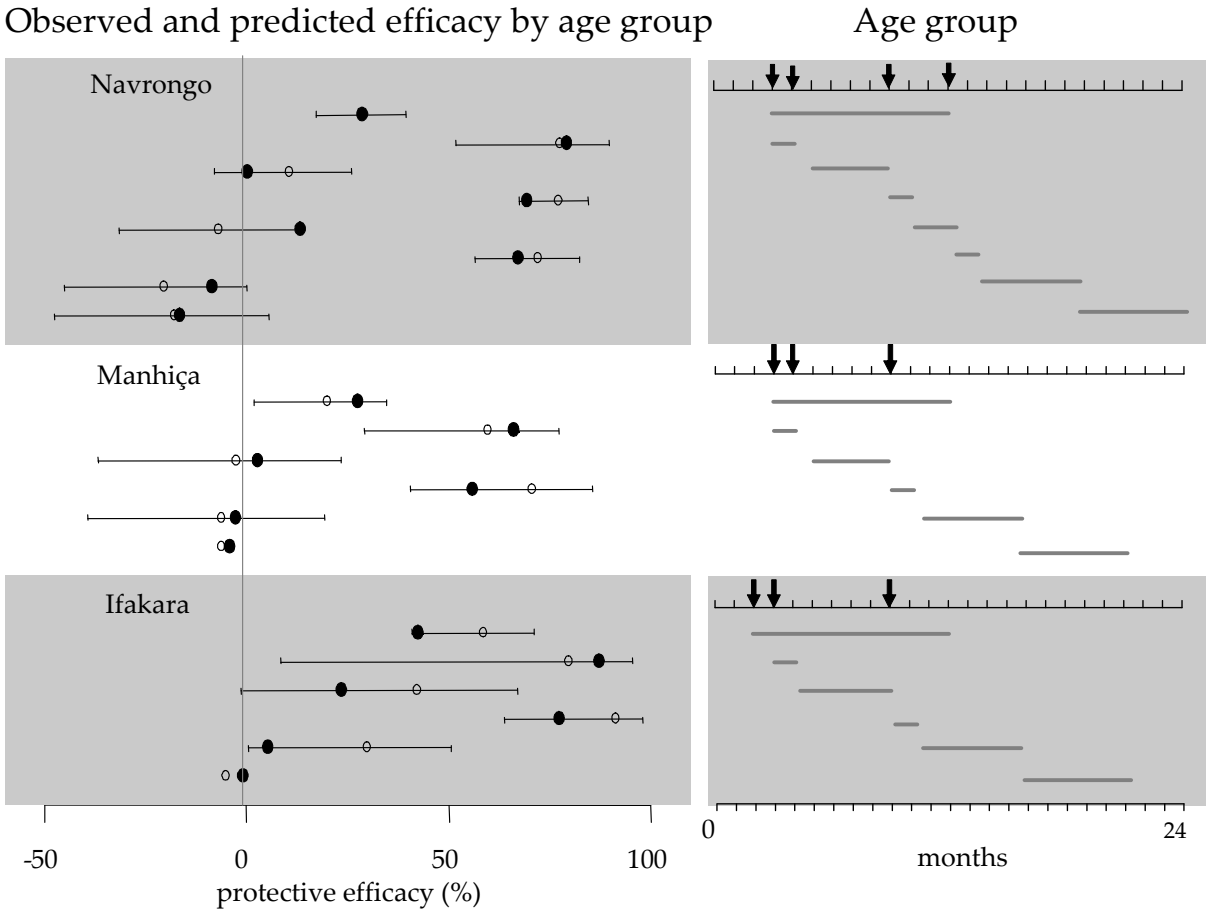
** One variable was varied at a time. In each scenario, the variables not being evaluated were fixed at the reference levels (indicated in **bold**). ‡The seasonality follows that of Namawala, Tanzania.[53] Each simulation assumes a recurring pattern of the vectoral capacity. †The proportion of infections cleared by the genotypes are set at 100%,100% and 50%. *We investigated the effect of age at dose by simulating a single IPTi dose at varying ages.

6.4 Results

Comparison of models with different mechanisms for IPTi

The agreement between the baseline model predictions and observed trial estimates was generally good (Figure 6.3 and Table 6.3). However, the continued positive protective effects of IPTi observed in Ifakara between doses and after the last dose were not fully captured. The Ifakara trial results for the periods between doses and after the last dose could be matched by reducing the transmission intensity as found in another study [47], but only if the intensity was reduced by at least 70% in the second year.

Figure 6.3. Comparison of trial estimates and baseline model predictions of protective efficacy of IPTi with SP against clinical episodes, by age group



Left side ○ Trial estimate with 95% confidence interval ● Baseline model predictions. Protective efficacy = percentage reduction in incidence of clinical episodes in IPTi group compared to placebo group

Right side The age groups that the morbidity surveillance refers to are illustrated. The arrows point to the scheduled ages at IPTi doses.

Table 6.3. Model fit for acute episodes assessed by weighted sums of squares

	Description of model	Navrongo	Manhiça	Ifakara	Total
Model 1	Baseline	0.618	0.046	0.239	0.903
Model 2	30 day SP action	0.557	0.039	0.386	0.982
Model 3	Repeat episodes	1.515	0.534	0.089	2.138
Model 4	Avoiding fevers	0.699	0.043	0.180	0.922
Model 5	Attenuated infections	2.845	0.423	0.128	3.396

We calculated the squared difference between the trial estimate and the predicted protective efficacy, weighted them by the number of person years at risk/100 and summed them to give a measure of the goodness-of-fit. A smaller value indicates a better fit. The three trials with EIR measurements were formally used to test the models, the remaining three were used only to validate the model output against general patterns in the trial results.

We compared the fit of the different models using weighted sums of squares (Table 6.3). None of the alternative models substantially improved agreement over that of the baseline model. However, models 2 and 4 also produced predictions which fell within the confidence intervals of the estimates of protective efficacy obtained in the trials (not shown) and could not be ruled out as providing an explanation for the effects of IPTi. Altering the duration of SP action (model 2) improved the fit slightly in the case of Navrongo and Manhiça, but reduced the fit for Ifakara, in comparison to the baseline model. Overall, however, the predictions were similar to those of the baseline model which can most likely be attributed to the similarity of the assumed action of SP since only the duration was altered. The predictions made by model 4 (where fevers penalized the acquisition of immunity) did not substantially differ from those of model 1. The greatest difference was seen in the results for Ifakara, where the assumption of benefits to acquired immunity from avoiding fevers increased the predicted efficacy between doses and after the last dose. Models 3 and 5 both incorporated processes to lengthen the duration of SP action beyond the duration of active drug concentrations. Their results were not consistent with Navrongo trial estimates, since they failed to capture the lack of effect of IPTi between doses and after the last dose. However, these models best predicted the high efficacy estimates observed between doses and after the last dose in the Ifakara trial.

Model 1 also adequately predicted the impact of IPTi on hospital admissions (Table 6.4).

Table 6.4. Observed and predicted protective efficacy of IPTi for severe episodes presenting for treatment

	Observed hospital admissions with parasitaemia	Observed all-cause admissions	Predicted admissions due to severe malaria (model 1)
	first dose - 12months		
Ifakara	58.5 (28.7, 75.8)	29.2 (6.6, 46.2)	48.7
Navrongo	50.2 (22.6, 68.0)	17.7 (-0.1, 32.3)	32.9
Manhiça	22.5 (-16.0, 48.2)	24.6 (7.2, 38.7)	30.0
	5 months after last dose		
Ifakara	15.3 (-65.0, 56.5)	-4.9 (-47.1, 25.2)	18.5
Navrongo	-14.2 (-95.9, 33.4)	-16.3 (-53.0, 11.6)	0.04
Manhiça	-32.0 (-114, 18.2)	8.1 (-25.7, 32.8)	-1.7

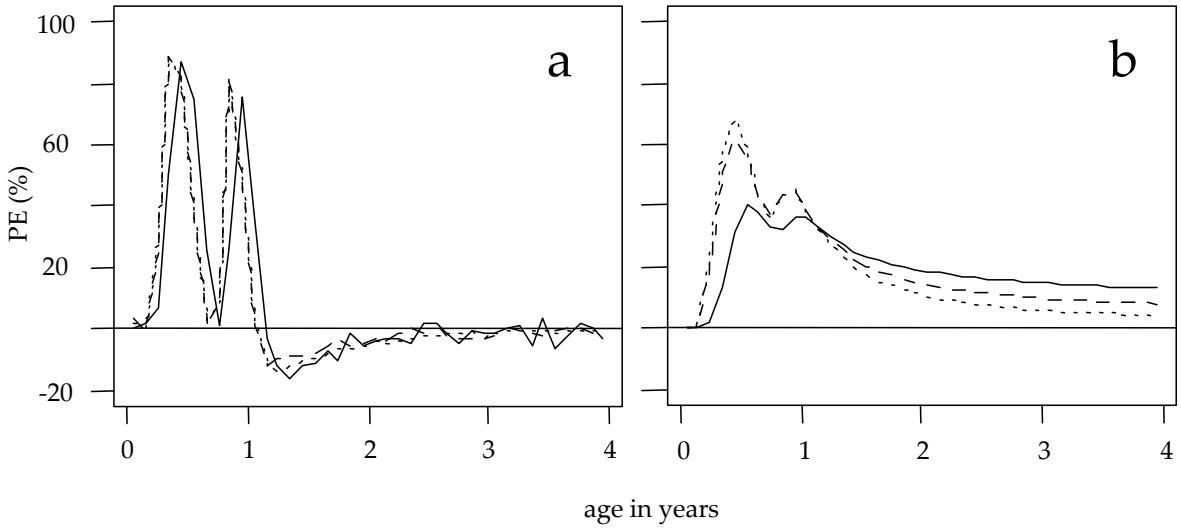
We also compared the output of the baseline model with the three trials not included in the formal comparison. They reported efficacy estimates in line with the Manhiça and Navrongo trials. Only previously unobserved features were used for further validation of the baseline model. In the trial in Kumasi,[29] IPTi doses were given at 3, 9 and 15 months of age. Kumasi villages with higher incidence of malaria in the placebo group show a linear increase in observed protective efficacy in the following 6 months.[54] Model 1 did not reproduce this result. Our simulated protective efficacy showed either no change or a slight decrease over a wide range of incidence values. The observed association may be due to different health system coverage in the different villages,[55] or the additional influence of increased acquired immunity on SP efficacy.[54] Alternatively, the observation may be due to different specificities of case definitions in the different villages.[54] The model would be able to capture the effect of treatment coverage if it is known, but at present is unable to capture the effect of increased immunity on SP action or effects of different specificities since non-malaria fevers are not modelled. Model 1 also did not fully capture the large negative efficacy observed for severe malarial anaemia in the post-intervention period in the trial in Tamale.[30]

Predicted impact of IPTi using the baseline model

Predicted patterns of protective efficacy by age were similar for acute malaria episodes, severe episodes and malaria-attributable mortality using model 1 (Figure 6.4a), with a small negative efficacy after the final dose for all outcomes. The slight delay in the peak protective efficacy for mortality is due to the inclusion of indirect malaria deaths, which occur as a result of an acute episode in conjunction with a co-morbidity and occur 30

days after the acute episode (Chapter 3). In contrast, the cumulative protective efficacy varied by outcome in the settings modelled (Figure 6.4b), at four years of age, the greatest effect was on mortality, followed by severe episodes. This is due partly to different predicted age-distributions of episodes in the placebo group and partly to age-dependent components in the model for severe episodes and mortality. The cumulative efficacy did not fall below zero for this or any of the other scenarios we have simulated.

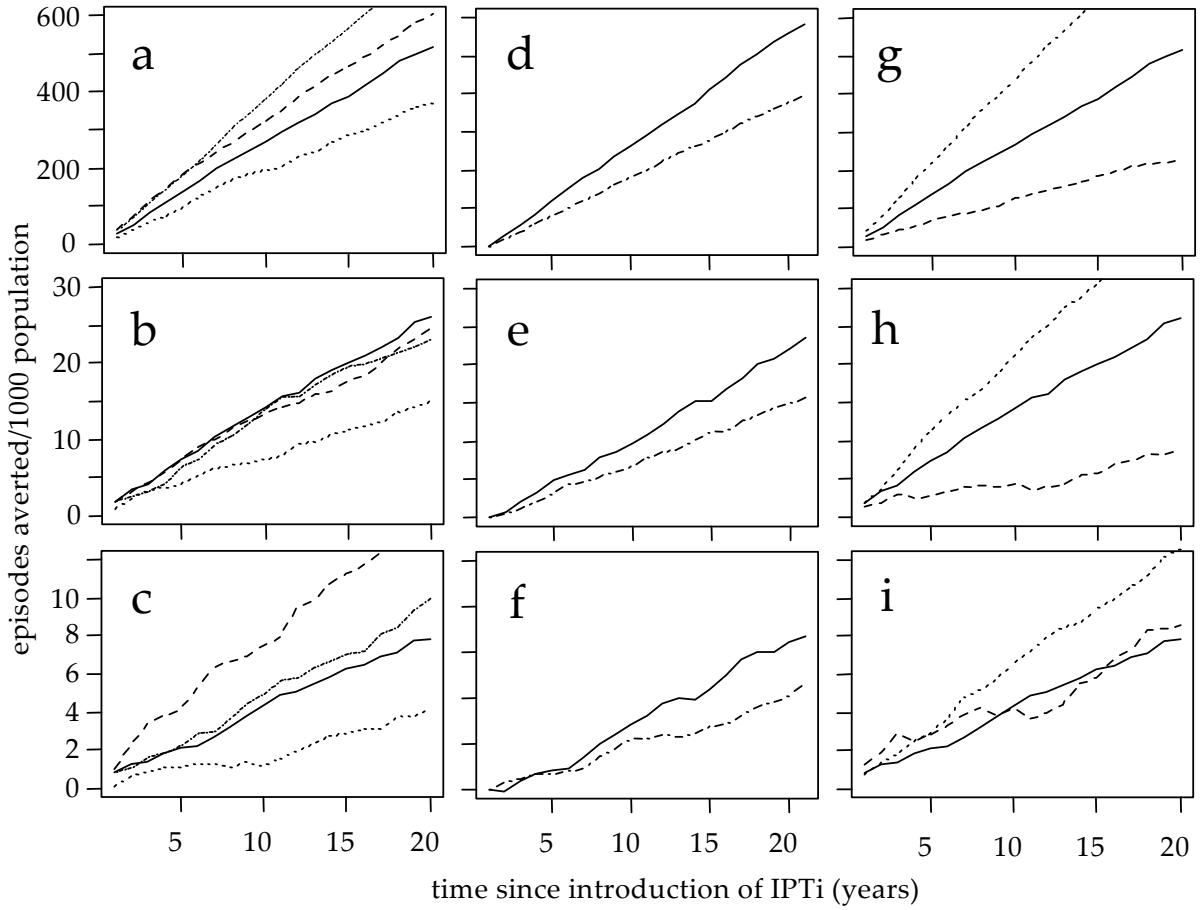
Figure 6.4 Predicted protective efficacy and cumulative protective efficacy by age



a. Predicted efficacy by age. b., Predicted cumulative efficacy by age for the reference scenario (Table 6.2) using model 1. IPTi doses were given following an EPI schedule at 3, 4 and 9 months of age. Dotted line=clinical malaria episodes; dashed line=severe episodes; solid line=malaria-attributable mortality.

The predicted number of episodes averted increased steadily over 20 years from the introduction of an IPTi programme (Figure 6.5). The linear increase reflects the negligible impact of IPTi on transmission and the short-term effects of IPTi in individuals. The predicted number of clinical episodes averted was greatest for moderate transmission settings (Figure 6.5a), but the number of deaths averted was greatest for higher transmission settings (Figure 6.5c). The number of deaths averted was greater for settings with a lower proportion of fevers treated and for IPTi drugs with a longer prophylactic period (Figure 6.5). Higher IPTi coverage and greater drug efficacy (or lower drug resistance) were also predicted to avert a greater number of episodes (not shown). The small predicted negative efficacy following the last dose as shown in Figure 6.4 was reduced both in settings where the impact of IPTi was less, such as with low drug efficacy or a high proportion of treated fevers, and in settings where there was low transmission intensity and thus little acquired immunity.

Figure 6.5 Predicted number of episodes averted by time since start of IPTi programme



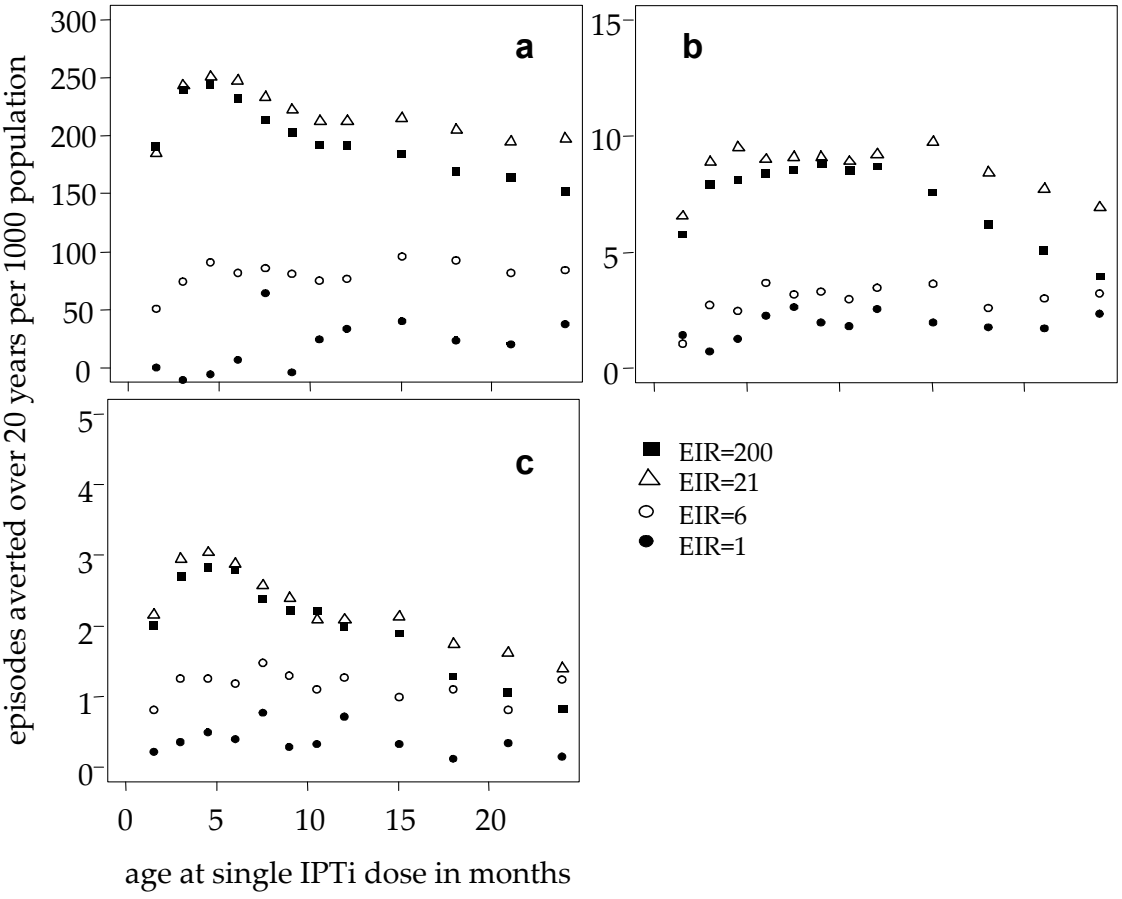
First column	Effect of varying EIR from 6 (dotted line), 21(solid line), 100 (dash-dot line), 200 (dashed line) on a) clinical malaria episodes b) severe episodes, c) deaths
Second column	Effect of varying health system coverage from 4% fevers treated (solid line) to 30% (dashed line) on d) clinical malaria episodes, e) severe episodes, f) deaths
Third column	Effect of varying prophylactic period for sensitive infections from treatment effect only (dashed line), 50 days (solid line), 100 days (dotted line) on g) clinical malaria episodes, h) severe episodes, i) deaths

Variables not being evaluated were fixed at the reference levels defined in Table 6.2

We simulated the number of episodes averted for varying Expanded Programme on Immunization (EPI) schedules (not shown). Predictions suggested that the spacing of doses was important, with a greater number of episodes averted for doses at 4, 6, 9 months compared to 4, 5, 9 months. For simplicity, we show the effect of age at the time of doses by simulating a single dose although the number of episodes averted with a single dose is lower than with the three dose schedule. A single SP dose was predicted to have a beneficial impact for all of the transmission intensites and ages up to 24 months. The age at which the maximum number of acute episodes and deaths were averted for a single SP dose was approximately 5 months for both high and moderate transmission intensities, but there is no obvious peak within the first 24 months for low transmission intensities (Figure 6.6). For severe episodes, two peaks are apparent for the high and moderate transmission intensities. These reflect a shift

between two types of severe malaria in the model. At younger ages, the majority of severe episodes averted are caused by an acute episode in conjunction with co-morbidity, and at older ages, overwhelming parasitaemia. For a single dose at older ages, the number of episodes averted by a single dose is greater for moderate transmission intensities. At low transmission intensities, it is been proposed that doses at later ages would avert the greatest number of episodes,[56] and our predictions are consistent with this. However, there is greater uncertainty in our predictions for low transmission intensities due to the effects of heterogeneity.[6,8]

Figure 6.6 Predicted number of episodes averted per 1000 population over 20 years by a single dose of IPTi by age at dose: a) acute episodes b) severe episodes c) malaria-attributable mortality



6.5 Discussion

Trial-specific inputs together with the baseline model reproduced the pattern of trial results reasonably well. Although there was no clear 'best model', none of the alternative models substantially improved agreement. This indicates that known features of malaria epidemiology together with the duration of SP action can account for the trial results and the variability between them. However, other hypotheses involving interactions between drug concentrations and acquired immunity or fevers and acquired immunity could not be ruled out as possible mechanisms.

Predictions using the baseline model suggest that IPTi using SP is effective over a wide range of transmission intensities at reducing malaria clinical episodes and malaria-attributable mortality in infants. Small negative protective efficacy values were predicted for a short time following the prophylactic periods, but these were outweighed by the cumulative benefits. The predicted short-term impact of IPTi on an individual's level of immunity and negligible effect on transmission intensity produced a steady rate of cases averted in the community over time from the start of an IPTi programme. IPTi was predicted to avert a greater number of episodes where IPTi coverage was higher, the health system treatment coverage lower, and for drugs which were more efficacious. A greater number of episodes were also averted with longer drug prophylactic periods, agreeing with considerations that the prophylactic period is important for IPTi.[57] The predicted reductions in mortality were not as large as those observed with ITN programmes or interruption of transmission. IPTi has a similar effect on severe episodes as a pre-erythrocytic vaccine with assumed characteristics,[58] but a much lower impact on uncomplicated episodes. This is likely to be due to the age-distribution of episodes and the longer-lasting effect of the pre-erythrocytic vaccine. The predictions also point to when IPTi is likely to not be useful. The number of cases averted is predicted to be fewer where IPTi coverage is lower, the health system treatment coverage is higher, and for short-acting drugs. At very low transmission intensities the predicted number of cases averted is few, however the model is likely to be less reliable at low transmission intensities[6,8] and so it is not easy to determine a transmission intensity below which IPTi is not useful.

This study offers possible explanations for the very strong positive protective efficacy observed in the Ifakara trial between doses and following the last dose. Three models produced predictions consistent with the observed results (Table 6.3), two describing a process for the continued positive benefits of IPTi, either by enhancing the acquisition of immunity (model 5) or by clearing infections which may have caused future clinical episodes (model 3), and model 1 in conjunction with sharply decreasing transmission. Neither model 3 nor 5 reproduced the results from the other sites as well as model 1. In the case of model 5, it is not easy to see why enhancing immunity should work only at low transmission intensities. However, it is possible that effects resulting from the timing of episodes (model 3) are only apparent at low transmission intensities. They may be otherwise obscured by processes such as interactions between infections or

acquired immunity. However, it is also possible that infections have shorter durations in infants than in adults.[59] Model 1 was able to reproduce the Ifakara trial results only if the initial EIR of 4 decreased by 70% or more in the second year. This is a substantial decrease and it is not known whether the transmission intensity did decrease so markedly over the study period (1999-2001). The incidence of uncomplicated episodes halved between 1995 and 2000.[46] Decreasing transmission intensity was shown to be consistent with the Ifakara results in another modelling study, but also required a substantial decrease of 22% per month.[47] Although the causes underlying the Ifakara results remain unknown, it seems reasonable that transmission intensity, either low or decreasing, is likely to have played a role. The potential contribution of the high coverage of ITNs to the large impact of IPTi in Ifakara has been noted elsewhere.[60]

The predicted impact on indirect malaria mortality was greater than that on direct malaria mortality. The predictions for indirect malaria mortality, and to a lesser extent, severe episodes rely on age-dependent co-morbidity functions. In a trial setting with access to good health care, the age-pattern of comorbidity may be quite different to that implicitly assumed by our models, which were fitted to other datasets (Chapter 3). In this case, the impact of IPTi on severe malaria and malaria-attributable mortality would be expected to be lower. Reductions in mortality have not been observed in the field trials reported to date, but the trials were not powered for this outcome.

This is the most comprehensive model to date, but still has certain limitations. The model predictions are unlikely to be reliable for low transmission intensities due to factors such as micro-heterogeneity and in-migration,[8] and thus it is difficult to determine the range of transmission intenities where IPTi is not useful.

We were unable to capture the effects of drug levels on parasite population dynamics by the current within-host model which relies on empirical averaged parasite densities. A within-host model which will capture immune development more explicitly is in preparation, and will include several immune responses, fevers and antigenic variation. It will also allow a more realistic model of the action of SP.

The model component for the action of SP was compatible with our model of malaria epidemiology. It is a simple model derived from dose-response curves and isobolograms.[5] SP is assumed to act on the infection, either clearing it or not. This model would be unable to account for certain observed effects such as density-dependent cure rates or effects of acquired immunity. A more refined model would allow the drug concentrations to affect individual parasites. Such a model has been formulated by Gatton and colleagues.[61] All SP models to date have been constructed using data on SP concentrations in adults. There is evidence that SP is cleared more quickly in children and requires a greater dose per kilogram to reach the same concentrations,[62] but little is known about infants. Data on the pharmacokinetics of SP in infants and the impact on infections with *dhfr* and *dhps* mutations are needed.[63] Adverse side-effects of SP are beyond the scope of this model. Although very rare,

these have been reported.[29] The model also does not incoporate the effect of IPTi on levels of drug resistance, which has been modelled elsewhere.[64,65]

We did not include the impact of IPTi on anaemia in our model. Whilst anaemia is an important consequence of malaria, the lack of knowledge about the dynamic effects of malaria and anaemia on one another limits our ability to construct a satisfactory model. We have previously used a model relating anaemia to the population prevalence of parasitaemia[66] to predict the impact of pre-erythrocytic vaccines.[58] However, in the case of IPTi, the short-term blood-stage effects of the drugs and use of iron supplementation in some of the trials rendered this model unsuitable. A model of anaemia may be able to account for the severe malarial anaemia rebound which was observed in the trial in Tamale.[30]

In conclusion, several models reproduced the trial data adequately so a single clearly preferred hypothesis for the secondary effect of IPTi on anti-malarial immunity cannot be identified. The previously published model adopted as our baseline model,[6] with additional components for the action of SP, can reproduce the trial results using known features of malaria epidemiology. We propose that this model is suitable for making predictions of the impact of IPTi. These predictions suggest that IPTi would have a beneficial impact across a wide range of settings. These analyses contribute to a growing database of the likely effectiveness of different malaria control strategies generated using this common simulation platform.[8]

6.A References

1. IPTi Consortium. www.ipti-malaria.org. 2008.

2. ter Kuile F, Steketee R. Intermittent preventive treatment in infants - adjusting expectations and seeing opportunity. *J Inf Dis* 2006, 194: 269-72

3. IPTi Statistical Working Group report. September 2007.

4. Schellenberg D, Cisse B, Mendendez C. The IPTi Consortium: research for policy and action. *Trends Parasitol* 2006, 22: 296-300

5. Hastings I, Watkins WM. Tolerance is the key to understanding antimalarial drug resistance. *Trends Parasitol* 2006, 22: 71-77

6. Smith T, Killeen GF, Maire N, Ross A, Molineaux L, Tediosi F, Hutton G, Utzinger J, Dietz K, Tanner M. Mathematical modeling of the impact of malaria vaccines on the clinical epidemiology and natural history of *Plasmodium falciparum* malaria: Overview. *Am J Trop Med Hyg* 2006 (Suppl 2), 75: 1-10

7. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 19-31

8. Smith T, Maire N, Ross A, Penny M, Chitnis N, Schapira A, Studer A, Genton B, Lengeler C, Tediosi F, de Savigny D, Tanner M. Towards a comprehensive simulation model of malaria epidemiology and control. *Parasitol* in press

9. Maire N. Using volunteer computing to simulate the epidemiology and control of malaria: malariacontrol.net. Distributed & Grid Computing - Science made transparent for everyone. Principles, applications and supporting communities. Rechenkraft.net, in press

10. White N. Assessment of the pharmacodynamic properties of antimalarial drugs in vivo. *Antimicrob Agents Chemother* 1997, 41: 1413-1422

11. Watkins WM, Mberu EK, Winstanley PA, Plowe CV. The efficacy of antifolate antimalarial combinations in Africa: a predictive model based on pharmacodynamic and pharmacokinetic analyses. *Parasitol Today* 1997, 13: 459-464

12. Coulibaly D, Diallo DA, Thera M, Dicko A, Guindo A, Koné A, Cissoko Y, Coulibaly S, Djimde A, Lyke K, Doumbo OK, Plowe CV. Impact of pre-season treatment on incidence of falciparum malaria and parasite density at a site for testing malaira vaccines in Bandiagara, Mali. *Am J Trop Med Hyg* 2002, 67: 604-10

13. Chandramohan D, Owusu-Agyei S, Carneiro I, Awine T, Amponsa-Achiano K, Mensah N, Jaffar S, Baiden R, Hodgson A, Binka F, Greenwood B. Cluster randomised trial of intermittent preventative treatment for malaria in infants in area of high, seasonal transmission in Ghana. *BMJ* 2005, 331: 727-733

14. Cairns M, Carneiro I, Milligan P, Owusu-Agyei S, Awine T, Gosling R, Greenwood B, Chandramohan D. Duration of protection against malaria and anaemia provided by intermittent preventive treatment in infants in Navrongo, Ghana. *PLoS ONE*. 2008, 3(5): e2227

15. Collins WE, Jeffery GM. A retrospective examination of the patterns of recrudescence in patients infected with *Plasmodium falciparum*. *Am J Trop Med Hyg* 1999, 61: 44-48

16. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationship between the entomologic inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 11-18

17. Schellenberg D, Aponte J, Menendez C, Alonso P. Intermittent preventive malaria treament in infants (correspondence). *Lancet* 2006, 366: 545-546

18. Sutherland C, Drakeley C, Schellenberg D. How is childhood development of immunity to *Plasmodium falciparum* enhanced by certain antimalarial interventions? *Malar J* 2007, 6: 161

19. Belnoue E, Costa F, Frankenberg T, et al. Protective T cell immunity against malaria liver stage after vaccination with live sporozoites under chloroquine treatment. *J Immunol* 2004, 172: 2487-2495

20. Pombo D, Lawrence G, Hirunpetcharat C, Rzepcyzk C, Bryden M, Cloonan N, Anderson K, Mahakunkijcharoen Y, Martin L, Wilson D, Elliott S, Elliott S, Eisen D, Weinberg J, Good M. Immunity to malaria after administration of ultra-low doses of red cells infected with *Plasmodium falciparum*. *Lancet* 2002, 360: 610-617

21. Cisse B, Sokhna C, Boulanger D, Milet J, Ba E, Richardson K, Hallett R, Sutherland C, Simondon K, Simondon F, Alexander N, Gaye O, Targett G, Lines J, Greenwood B, Trape J-F. Seasonal intermittent preventive treatment with artesunate and sulfadoxine-pyrimethamine for prevention of malaria in Senegalese children a randomised, placebo-controlled, double-trial. *Lancet* 2006, 367: 659-667

22. Massaga JJ, Kitua A, Lemnge MM, Akida J, Malle LN, Ronn AM, Theander TG, Bygbjerg IC. Effect of intermittent treatment with amodiaquine on anaemia and malarial fevers in infants in Tanzania: a randomised placebo-controlled trial. *Lancet* 2003, 361: 1853-1860

23. Desai MR, Mei JV, Kariuki SK, Wannemuehler KA, Phillips-Howard PA, Nahlen BL, Kager PA, Vulule JM, ter Kuile FO. Randomized, controlled trial of daily iron supplementation and intermittent sulfadoxine-pyrimethamine for the treatment of mild childhood anaemia in Western Kenya. *J Inf Dis* 2003, 187: 658-666

24. Dicko A, Sagara I, Sissoko M, Guindo O, Diallo A, Kone M, et al. Impact of intermittent preventive treatment with sulphadoxine-pyrimethamine targeting the transmission season on the incidence of clinical malaria in children of 6 months to 10 years in Kambili, Mali. *Am J Trop Med Hyg* 2004, 71 (suppl): abstract 18

25. Schellenberg D, Menendez C, Kahigwa E, Aponte J, Vidal J, Tanner M, Mshinda H, Alonso PL. Intermittent treatment for malaria and anaemia control at time of routine vaccinations in Tanzanian infants: a randomised, placebo- controlled trial. *Lancet* 2001, 357: 1471-1477

26. Schellenberg D, Menendez C, Aponte J, Kahigwa E, Tanner M, Mshinda H, Alonso P. Intermittent preventative antimalarial treatment for Tanzanian infants: follow-up to age 2 years of a randomised, placebo-controlled trial. *Lancet* 2005, 365: 1481-1483

27. Macete E, Aide P, Aponte J, Sanz S, Mandomando I, Espasa M, Sigauque B, Dobano C, Mabunda S, Dgedge M, Alonso P, Menendez C. Intermittent preventive treatment for malaria control administered at the time of routine vaccinations in Mozambican infants: a randomized, placebo-controlled trial. *J Inf Dis* 2006, 194: 276-85

28. Grobusch MP, Lell B, Schwarz N, Gabor J, Dörneman J, Pötschke M, Oyakhirome S, Kiessling G, Necek M, Längin M *et al*. Intermittent preventive treatment in infants against malaria in Gabon - a randomized, double-blind, placebo-controlled trial. *J Inf Dis* 2007, 196: 1595-1602

29. Kobbe R, Kreuzberg C, Adjei S, Thompson B, Langefeld I, Thompson P, Abruquah H, Kreuels B, Ayim M, Busch W, Marks F, Amoah K, Opoku E, Meyer C, Adjei O, May J. A randomized controlled trial of extended intermittent preventive antimalarial treatment in infants. *Clin Infect Dis* 2007, 45: 16-25

30. Mockenhaupt F, Reither K, Zanger P, Roepcke F, Danquah I, Saad E, Ziniel P, Dzisi S, Frempong M, Agana-Nsiire P, Amoo-Sakyi F, Otchwemah R, Cramer J, Anemana S, Dietz E, Bienzle U. Intermittent preventive treatment in infants as a means of malaria control: a randomized, double-blind, placebo-controlled trial in northern Ghana. *Antimicrob Agents Chemother* 2007, 51: 3273-3281

31. Drakeley C, Schellenberg D, Kihonda J, Sousa CA, Arez AP, Lopes D, Lines J, Mshinda H, Lengeler C, Armstrong SJ, Tanner M, Alonso P. An estimation of the entomological inoculation rate for Ifakara: a semi-urban area in a region of intense malaria transmission in Tanzania. *Trop Med Int Health* 2003, 8: 767-774

32. Appawu M, Owusu-Agyei S, Dadzie S, Asoala V, Anto F, Koram K, Rogers W, Nkrumah F, Hoffman SL, Fryauff DJ. Malaria transmission dynamics at a site in northern Ghana proposed for testing malaria vaccines. *Trop Med Int Health* 2004, 9: 164-170

33. Schellenberg D, Kahigwa E, Drakeley C, Malende A, Wigayi J, Msokame C, Aponte J, Tanner M, Mshinda H, Menendez C, Alonso PL. The safety and efficacy of sulfadoxine-pyrimethamine, amodiaquine, and their combination in the treatment of uncomplicated *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2002, 67: 17-23

34. Oduro A, Anyorigya T, Hodgson A, Ansah P, Anto F, Ansah N, Atuguba F, Mumuni G, Amankwa J. A randomized comparative study of chloroquine amodiaquine and sulphadoxine-pyrimethamine for the treatment of uncomplicated malaria in Ghana. *Trop Med Int Health* 2007, 10: 279-284

35. Abacassamo F, Enosse S, Aponte J, Gomez-Olive X, Quinto L, Mabunda S, Barreto A, Magnussen P, Ronn AM, Thompson R, Alonso PL. Efficacy of chloroquine, amodiaquine, sulphadoxine-pyrimethamine and combination

therapy with artesunate in Mozambican children with non-complicated malaria. *Trop Med Int Health* 2004, 9: 200-208

36. Mockenhaupt F, Ehrhardt S, Dzisi S, Bousema J, Wassilew N, Schreiber J, Anemana S, Cramer J, Otchwemah R, Sauerwein R, Eggelte T, Bienzle U. A randomised, placebo-controlled, double-blind trial on sulfadoxine-pyrimethamine alone or in combined with artesunate or amodiaquine in uncomplicated malaria. *Trop Med Int Health* 2005, 10: 512-520

37. Alloueche A, Bailey W, Barton S, Bwika J, Chimpeni P, Falade C, Fehintola F, Horton J, Jaffar S, Kanyok T *et al*. Comparison of chlorproguanil-dapsone with sulphadoxine-pyrimethamine for the treatment of uncomplicated falciparum malaria in young African children: double-blind randomised controlled trial. *Lancet* 2004, 363: 1843-1848

38. Smith T, Hurt N, Teuscher T, Tanner M. Is fever a good sign for clinical malaria in surveys of endemic communities? *Am J Trop Med Hyg* 1995, 52: 306-310

39. Chandler C, Drakeley C, Reyburn H, Carneiro I. The effect of altitude on parasite density case definitions. *Trop Med Int Health* 2006, 11: 1178-1184

40. Chitnis N, Steketee RW, Smith T. A mathematical model for the dynamics of malaria in mosquitoes feeding on a heterogeneous host population. *J Biologic Dynamics,* in press

41. Lines J, Armstrong-Schellenberg JR. For a few parasites more: inoculum size, vector control and strain-specific immunity to malaria. *Parasitol Today* 1992, 8: 381-382

42. Glynn JR, Collins WE, Jeffery GM, Bradley DJ. Infecting dose and severity of malaria. *Trans R Soc Trop Med Hyg* 1995, 89: 281-283

43. Boyd M. The influence of sporozoite dosage in vivax malaria. *Am J Trop Med* 1940, 20: 279

44. Schellenberg DM, Aponte JJ, Kahigwa EA, Mshinda H, Tanner M, Menendez C, Alonso PL. The incidence of clinical malaria detected by active case detection in children in Ifakara, southern Tanzania. *Trans R Soc Trop Med Hyg* 2003, 97: 647-654

45. Henning L, Schellenberg D, Smith T, Henning D, Alonso P, Tanner M, Mshinda H, Beck H-P, Felger I. A prospective study of Plasmodium falciparum multiplicity of infection and morbidity in Tanzanian children. *Trans R Soc Trop Med Hyg* 2004, 98: 687-694

46. Schellenberg D, Menendez C, Aponte J, Guinovart C, Mshinda H, Tanner M, Alonso PL. The changing epidemiology of malaria in Ifakara Town, southern Tanzania. *Trop Med Int Health* 2004, 9: 68-76

47. Gosling R, Ghani A, Deen J, von Seidlein L, Greenwood B, Chandramohan D. Can changes in malaria transmission intensity explain lack of rebound and contribute to high protective efficacy of intermittent preventive treatment for malaria in infants? *Malar J* 2008, 7: 54

48. Maire N, Aponte J, Ross A, Thompson R, Alonso P, Utzinger J, Smith T. Modeling a field trial of the RTS,S/ASO2A malaria vaccine. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 104-110

49. Binka F, Morris SS, Ross DA, Arthur P, Aryeetey ME. Patterns of malaria morbidity and mortality in children in northern Ghana. *Trans R Soc Trop Med Hyg* 1994, 88: 381-385

50. Stepniewska K, Taylor W, Mayxay M, Price R, Smithuis F, Guthman J-P, Barnes K, Myint HY, Adjuik M, Olliaro P, Pukrittayamee S, Looareesuwan S, Hien T, Farrar J, Nosten F, Day N, White N. In vivo assessment of drug efficacy against *Plasmodium falciparum* malaria: duration of follow-up. *Antimicrob Agents Chemother* 2004, 48: 4271-4280

51. Ringwald P. Susceptibility of *Plasmodium falciparum* to antimalarial drugs: Report on global monitoring, 1996-2004. 2005. World Health Organization.

52. INDEPTH Network. Population, Health and Survival at INDEPTH Sites. Ottawa: IDRC, 2002.

53. Smith T, Charlwood JD, Kihonda J, Mwankusye S, Billingsley P, Meuwissen J, Lyimo E, Takken W, Teuscher T, Tanner M. Absence of seasonal variation in malaria parasitaemia in an area of intense seasonal transmission. *Acta Trop* 1993, 54: 55-72

54. Kobbe R, Adjei S, Kreuzberg C, Kreuels B, Thompson B, Thompson P, Marks F, Busch W, Tosun M, Schreiber N, Opoku E, Adjei O, Meyer C, May J. Malaria incidence and efficacy of intermittent preventive treatment in infants. *Malar J* 2007, 6: 163

55. Kreuels B, Kobbe R, Adjei S, Kreuzberg C, von Reden C, Bäter K, Klug S, Busch W, Adjei O, May J. Spatial variation of malaria incidence in young children from a geographically homogenous area with high endemicity. *J Inf Dis* 2008,97: 85-97

56. Chandramohan D, Webster J, Smith L, Awine T, Owusu-Agyei S, Carneiro I. Is the Expanded Programme on Immunisation the most appropriate delivery

system for intermittent preventive treatment of malaria in West Africa? *Trop Med Int Health* 2007, 12: 743-750

57. White N. Intermittent presumptive treatment for malaria. *PLoS Med* 2005, 2: 28-33

58. Maire N, Tediosi F, Ross A, Smith T. Predictions of the epidemiologic impact of introducing a pre-erythrocytic vaccine into the expanded program on immunization in sub-Saharan Africa. *Am J Trop Med Hyg* 2006, 75: 111-118

59. Walton GA. On the control of malaria in Freetown, Sierra Leone. 1. *Plasmodium falciparum* and *Anopheles gambiae* in relation to malaria occurring in infants. *Ann Trop Med Parasitol* 1947, 41: 380-407

60. Menendez C, Schellenberg D, Macete E, Aide P, Kahigwa E, Sanz S, Aponte J, Sacarlal J, Mshinda H, Tanner M, Alonso P. Varying efficacy of intermittent preventive treatment for malaria in infants in two similar trials: public health implications. *Malar J* 2007, 26: 132

61. Gatton M, Martin L, Cheng Q. Evolution of resistance to sulfadoxine-pyrimethamine in *Plasmodium falciparum*. *Antimicrob Agents Chemother* 2004, 48: 2116-2123

62. Barnes K, Little F, Smith P, Evans A, Watkins W, White N. Sulfadoxine-pyrimethamine pharmacokinetics in malaria: pediatric dosing implications. *Clin Pharmacol Ther* 2006, 80: 582-596

63. White N. How antimalarial drug resistance affects post treatment prophylaxis. *Malar J* 2008, 7: 9

64. Alexander N, Sutherland C, Roper C, Cissé B, Schellenberg D. Modelling the impact of intermittent preventive treatment for malaria on selection pressure for drug resistance. *Malar J* 2007, 6: 9

65. Prudhomme O'Meara W, Smith D, Mckenzie F. Potential impact of intermittent preventive treatment (IPT) on spread of drug-resistant malaria. *PLoS Med*. 2006, 3: 141

66. Carneiro I, Smith T, Lusingu J, Malima R, Utzinger J, Drakeley C. Modeling the relationship between the population prevalence of *Plasmodium falciparum* malaria and anemia. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 82-89

Chapter 7: Discussion

The development of an integrated model of malaria was motivated by the need for predictions of the likely impact of interventions. This is the first major attempt to model the dynamic effects of malaria transmsission, parasitological status, morbidity, mortality and cost-effectiveness using model components fitted to field data. This thesis contributes the elements that consider the immediate consequences of human infection: uncomplicated morbidity, severe morbidity, mortality and transmission to the vector. The integrated model was also applied to specific questions concerning one potential intervention, IPTi.

A detailed discussion on the findings was given in each chapter. In this discussion, the model components are set in context and strengths and limitations are discussed with an emphasis on the special features of models of downstream events.

7.1 The context of the models

7.1.1 Integrated models

There is no other malaria model which combines parasitology, burden of disease, health systems, transmission and economics. The integrated models that exist mostly fall into two categories. Those in the first category are constructed by combining biological components, for example embedding a within-host model into a population model,[1] including both human and mosquito populations,[2] or incorporating both biological and environmental components.[3] Whilst these models extend the scope of biological models, they do not include morbidity, mortality or cost-effectiveness. These measures are, however, included in the second category of integrated models,[4-7] which aim to predict the epidemiological impact and cost-effectiveness of different interventions. The models are constructed by combining empirical estimates of the effectiveness of interventions (often as a percentage reduction), studies on disease burden, and unit costs. There is no explicit consideration of the dynamics of transmission and immunity, in particular the loss of population immunity which results from reduced exposure.[8] One previous model does straddle biology and economics, combining a model of the transmission of drug resistant parasites with economic data.[9]

7.1.2 Models of acute malaria episodes

There are few mathematical models of morbidity and mortality due to malaria.[1,10] This may be partly due to the focus on transmission of early models of malaria, and partly because the mechanisms are unclear.[11] Some models incorporate fever, and these vary widely in both their purpose and in the platform on which the fever component rests. The primary purpose of some of these models is to investigate the effect of fever on parasite dynamics due to temperature-induced mortality.[12-14] Others explored the nature of acquired immunity.[15-17] Some, including the component reported in Chapter 3, make predictions of the impact of interventions on morbidity.[18,19]

An early model by Aron assumed that there was a class of susceptibles who became ill on infection.[16] The assumption that non-immunes experience symptoms immediately following successful blood-stage infection is contradicted by data from deliberate infection of patients with *P. falciparum* as therapy for neurosyphilis. Subsequent models have used a pyrogenic asexual density threshold. Initially, a constant pyrogenic threshold was used.[15,18] An analysis suggesting that pyrogenic thresholds increase during the course of an infection prompted time-specific pyrogenic thresholds to be incorporated into a model investigating antigenic variation.[20] Wide variation between individuals prompted individual-specific thresholds in a model describing the first wave of parasitaemia.[19] These were estimated by fitting the model to each individual in turn. The model component in Chapter 2 has a pyrogenic threshold which responds dynamically to the parasite load, and is therefore both individual- and time-specific.

7.1.3 Models of severe malaria episodes

Only three previously published models describe the processes leading to severe malaria. Gupta and others assumed that severe malaria occurs following successful blood-stage infection in non-immunes, and that immunity is acquired after a number of infections. They concluded that whilst strain-transcending immunity was important for non-cerebral severe malaria,[21] cerebral malaria was a discrete syndrome caused by a discrete set of *P. falciparum* antigenic types for which strain-specific immunity is important.[22] Again, the assumption that non-immunes experience symptoms immediately following successful blood-stage infection is contradicted by the malaria therapy data. The notion of distinct syndromes in severe malaria has been questioned,[23] as has the structuring of parasite populations into a limited number of strains (Chapter 2).

Dietz and others[19] added a component for severe morbidity to their within-host model of the first wave of parasitaemia. The incidence of severe malaria was assumed to be related to a large parasite biomass, and a common threshold of asexual parasite density was used for all patients. The model described in Chapter 3 incorporates two processes for severe malaria. One is a large parasite biomass (related to asexual parasite density) similar to the model by Dietz and others. The other is a novel process allowing the

recognition that severe malaria may be triggered by an uncomplicated malaria episode in conjunction with co-morbidity.

7.1.4 Models of malaria-associated mortality

Malaria mortality has been incorporated into simple models of the impact of an intervention. The models combine estimates of the effectiveness of an intervention from clinical trials or other field data, estimates of the disease burden and unit costs to estimate the reduction in morbidity, mortality and the cost-effectiveness of an intervention.[5,6,24] These models do not consider the dynamics of transmission and immunity, and thus ignore feedbacks such as a reduction in transmission and loss of population immunity. Recently, Hastings and others constructed models which predict direct and indirect malaria mortality from the frequency of a mutation encoding drug resistance.[25]

The model components described in Chapters 3 and 4 includes both direct (following an episode of severe malaria), and indirect mortality (where the host is weakened by comorbidity, or via maternal infection). By specifying the processes leading to death, the integrated model has the advantage of being able to predict effects of interventions where estimates of the impact are not available and to take dynamic effects into account.

Models of neonatal mortality are few and have been reviewed in Chapter 4. They depend heavily on birth weight as a mediating variable, but with present data this is difficult to avoid. Chapter 4 provides a review of the evidence of neonatal mortality due to maternal infection across a wide range of transmission intensities. Recent analyses have added to the information available on the association between maternal infection and infant mortality,[26] and the timing of infections and adverse birth outcomes.[27]

7.1.5 Models of infectivity of humans to mosquitoes

As an essential part of the transmission cycle, infectivity of humans to mosquitoes has attracted more attention than morbidity and has long been incorporated into models of malaria transmission. Early compartmental models define a fixed period of human infectivity.[28,29] Dietz and colleagues[30] allows differences between classes of human hosts in infectiousness: individuals classed as immune are not infective to mosquitoes, and positives can be infective or non-infective (infective positives convert to non-infective positives). An individual-based model by Gu *et al* [2] allows the probability of infecting a feeding mosquito to be less than 1. Infectivity was fixed at probability *c*, after the incubation period.

These models do not incorporate the wide variation in infectivity observed in the malariatherapy experiments both between individuals and over the course of an

infection. Densities are an obvious way of incorporating this variation and since infection of the mosquito requires gametocytes to be taken up in a blood meal, the process is likely to be density-driven. Gametocyte densities themselves have been included in some models either as the focus[31] or as part of the parasite dynamics.[32] A recent model by Gatton and colleagues[33] relates asexual parasite densities to subsequent infectivity by explicitly modelling gametocytogenesis. The components for gametocytes and subsequent infectivity were added to their individual-based stochastic within-host model.[20] Stochasticity is included at three points: the conversion rate of asexual parasites to gametocytes follows a lognormal distribution, as does the lifespan of gametocytes, and the number of gametocytes ingested by the mosquito follows a negative binomial distribution. In comparison, the model component described in Chapter 5 does not explicitly include gametocytogenesis. Instead it was modelled implicitly by estimating infectivity from analyses of the association between lagged asexual densities and infectivity. This allows us to avoid modelling unnecessary variables which have an uncertain quantitative relationship with infectivity, and implicitly takes into account the timecourse of gametocytaemia. Stochasticity is introduced both via the stochasticity already present in the parasite densities and via a lognormal distribution for the ratio of functional female (or male) gametocytes per blood meal to the weighted sum of asexual parasite densities. Both models address the need for both a functional male and female gametocyte to be taken up in the blood meal. The males and females are assumed to be independently distributed in the blood, but this may not be the case.[34]

7.2 Strengths and limitations of model components for downstream events

Many features of the model components are both strengths and limitations.

7.2.1 Integration into a comprehensive model

Whilst each component contributes a novel sub-model, a great strength of this work is that the components are integrated into a comprehensive model. The whole is greater than the sum of its parts: together the components provide the power and flexibility to provide predictions of key indicators of interest to programme planners of the impact of many different types of interventions on a common platform. The model can also suggest where to look for counter-intuitive effects.

The scope of the predictions is not unlimited. The model does not incorporate interactions with specific other diseases, such as HIV. It is unable to forecast effects of an intervention beyond those on malaria, for example the effect of DDT on the environment. It does not predict social outcomes, such as equitable access to health care, nor does it take into account spatial information. Very recently, there has been

some interest in the goal of elimination. It is not advisable, however, to use this model to make predictions of whether malaria interventions might lead to elimination in an area. There are factors, such as microheterogeneity in transmission and in-migration, which are not accounted for in the model but which become increasingly important in low transmission settings. In addition, much of the data used for estimating the parameter values came from areas with medium or high transmission intensities.

A potential disadvantage to an integrated model is that it can be perceived to be complicated. Adding even very simple sub-models together can produce a model which is difficult to conceptualise. The cut-off for deeming a model too complicated is highly individual. A complicated model can lead to a lack of acceptance,[35] and can deter questions and hide crucial assumptions.[36,37] It may be difficult to identify the drivers of an effect other than by making predictions.

7.2.2 Fitting the model to data

All of the parameters in the model were fitted to data to ensure that the elements of the model conform as far as possible to reality and to minimise uncertainty in the predictions. This is a substantial improvement over most malaria models. There have been only limited efforts to optimize models of malaria by formal fitting to data, most models being superficially validated against field observations. The model by Dietz and others[30] was the first fitted to data.[38] Cancre and others[39] used MCMC methods to fit parameters to the model by Struchiner and others.[38]

Fitting the integrated model to data presented several challenges. One source of difficulty was gaps in the data available such as the unknown burden of malaria in adults, the lack of direct data on malaria-attributable neonatal mortality, the absence of a proxy variable for immunity and uncertainty in health system parameters. In addition, many sites with carefully collected information on morbidity or prevalence could not be matched exactly to entomological data.

The second obstacle was the enormous computational power required to estimate the parameters of the stochastic simulation model. Some components could be fitted to data without requiring other parts of the model and thus were relatively straightforward. The components for infectivity (Chapter 5) and malaria-attributable neonatal mortality (Chapter 4), as well as anaemia[40] were estimated in this way. However, where simulations were necessary, the computational demands were much greater. Harnessing sufficient power to fit the parameters was achieved by distributed computing, initially using computers within the Swiss Tropical Institute and later through volunteer computing via the internet. This is described in detail elsewhere.[8,41] The parameter estimates from the different methods of fitting are given in Appendix A.1. Overall, they did not differ substantially. A probalistic sensitivity analysis of the parameter estimates is currently underway.

Whilst the problem of sufficient computational power has been solved, other issues remain. Parameters of the components can be fitted simultaneously but it is unclear how best to weight the different outcomes of parasite prevalence, densities, multiplicity of infections, incidence of acute and severe episodes and mortality. Currently they each have approximately equal weight. The best algorithm for fitting is also unclear.[8] Many diverse fields such as fisheries[42] and climate modelling face the same challenge of fitting to data on multiple outcomes, and this is an active area of research in computational sciences.

Finally, estimating parameters for biologically plausible models can lead to collinearity.[43] It was found, when fitting the indirect mortality component, that the parameter values for non-malaria infant mortality and the co-morbidity prevalence at birth were highly correlated.

7.2.3 Heterogeneity

Important patterns at the community level derive ultimately from differences among individuals.[11,37] Although the model is an individual-based one, it was fitted to aggregated data. The predictions for an aggregated number of simulated individuals fit well to the data, however the extent to which variations in longitudinal patterns from individuals are captured has not yet been investigated. Some heterogeneity between simulated individuals arises because of effects of host age, seasonality of transmission and propensity for high parasite densities and stochasticity additionally occurs throughout the model. Further sources of heterogeneity are not included. Stochastic models can easily capture heterogeneity in transmission between hosts by sampling from distributions without explicitly stating what the sources of variation are. A component for between-individual micro-heterogeneity in transmission has been recently developed.[44]

Heterogeneity in single variables is known to have important effects for modelling such as sustaining parasites in a population.[11,36,45,46] Heterogeneity can also bias predictions of the impact of interventions.[35,47] A recent paper showed that there were differential effects of heterogeneity in biting rates on R_0 depending on the population size.[48]

A myriad of heterogeneities affect malaria epidemiology, falling into roughly into four categories (i) heterogeneity of transmission; (ii) biological heterogeneity of the host in susceptibility and response to malaria infection[49-52]; (iii) heterogeneity in host behaviour, including quality of housing, use and knowledge of protective measures such as ITNs and treatment-seeking strategies; and (iv) heterogeneity in the risk of co-morbidity and nutrition.

Heterogeneities in the four categories are unlikely to be uncorrelated. A factor, such as socio-economic status (SES), may be associated with several different types of heterogeneity. SES can influence transmission through quality of housing, knowledge of protection measures and use of ITNs.[53,54] Risks of co-morbidity and malnutrition may

be associated with SES.[55] SES may influence treatment-seeking behaviour. Amongst carers of children in rural Tanzania, treatment-seeking and knowledge of danger signs were worse in poorer families.[56] Such inequalities tend to persist over time.[57]

The effects of multiple sources of heterogeneity on models for predicting the effects of interventions are not established. It is not known whether the effect of heterogeneity in biting rates between individuals on R_0 is altered in the presence of other kinds of heterogeneity. This is likely to depend on the extent to which they covary. Covariance between the four categories is plausible, and the degree to which it occurs likely to vary from site to site. An analysis of the effect of heterogeneity in parameters of the Macdonald model on the estimation of R_0 reported a change in R_0 if a parameter co-varied with a second parameter.[35] R_0, however, may not be the most important measure unless elimination is under consideration.

More generally, covariation may describe sub-groups within a population. Ignoring this structure may create problems for an integrated model when estimating the parameters of the individual components, or when simulating an intervention trial since different subgroups can respond differently to an intervention (for example[58]). A sub-population may be reached neither by the health system nor by any of several interventions. Further work is needed to determine the circumstances under which multiple covarying sources of heterogeneity may affect conclusions from a comprehensive model about the impacts of different interventions.

7.2.4 Validation

The model has been validated to a limited extent. The validation criteria were aimed at both the individual model components and the model predictions of the impact of an intervention.

Validation of the model components was possible for some outcomes. For example, the model component for infectivity was constructed using the malariatherapy data and then validated against field data (Chapter 5). In other cases, however, all the available relevant data was used for fitting the component parameters, such as for morbidity and mortality (Chapters 2, 3 and 4). Even where validation criteria are aimed at a particular component, they were frequently conditional on other components such as parasitology.

The model predictions have been validated against data from trials of interventions. The model has reproduced the results from the trial of a pre-erythrocytic vaccine[59] and the IPTi trials (Chapter 6). Although important for gaining credibility, the process of comparing model output to trial data is limited for validation.[60] Parts of the model may well be conceptually acceptable even though the integrated model is unable to reproduce the trial results. Conversely, the integrated model may be validated even though parts of the model are unsound.[61] The best model in terms of formulation may not be the best-fitting model, as found by Nedelman who fitted the Garki model and

several variants to the Garki data.[62] A further problem when using trial data to validate the model output is that the required model inputs, such as transmission intensity, may not be known. They are a prerequisite for validation.[63] In Chapter 6, the characteristics of some IPTi trial sites could not be determined. This uncertainty made it less likely that a model could be rejected on the grounds of fit since a range of values was simulated for the uncertain inputs such as the frequency of drug resistant infections. Others have found similar problems. Najera used the Macdonald model to help plan a field trial of interventions, but found that most of the necessary input variables were lacking or not sufficiently known.[64]

7.3 Implications of the application of the model to IPTi

The application of the integrated model to IPTi demonstrates that, with site-specific inputs, the model can reproduce the results of several trials and that it can provide information and observations which are potentially useful to decision-makers. Although more than one hypothesis remains in contention, it was shown that no interactions between SP concentrations, fever and acquired immunity need be assumed to reproduce the observed pattern of trial results. Predictions suggest that IPTi with a reasonably efficacious long-acting drug would be effective in a wide range of epidemiological settings. To complement these results, predictions of cost-effectiveness are planned, as well as a comparison of the effectiveness of IPTi and seasonal IPTc in different settings. In the longer-term, predictions of IPTi should be integrated with those of other interventions to be able to inform an integrated malaria control programme.

The predictions were influenced by the individual model components. The differing predicted cumulative efficacy of IPTi on different outcomes was driven by the age-dependent co-morbidity functions for both severe malaria and indirect mortality. Avoiding an acute episode during ages when the risk of co-morbidity is highest reduced the cumulative risks. The model predicts little impact on transmission intensity; the infants contribute little to the infectious reservoir because they make up a small percentage of the population, and have a low parasite prevalence and small body size.

The study was conducted under the co-ordination of the IPTi Consortium. The consortium was formed to develop and co-ordinate a research and implementation agenda that would resolve outstanding scientific questions about IPTi and move the intervention into policy and practice. Alongside other evidence of the suitability of IPTi, modelling can play a role in evaluating applicability, but timing, communication and overcoming resistance to modelling are crucial to whether decision-makers can fully capitalise on these results.

7.4 Outlook

The model components described here contribute to the most comprehensive integrated model of malaria to date. The modular structure of the integrated model allows all of the model components to be improved and extended in the future. Models for vector control, within-host parasite dynamics, heterogeneity, drug resistance, decay of acquired immunity are all in development. The model will be further validated and improved as an iterative process.

The model has been applied to IPTi in this thesis, and to pre-erythrocytic vaccines.[65,66] It can be easily adapted to predict the dynamic effects of different interventions, and different combinations of interventions. The accumulating collection of predictions will be a valuable resource for a rational basis for decisions about malaria control strategies. Bringing about a sustained reduction in the intolerable burden of malaria is long overdue, and the model makes a contribution to achieving this goal.

7.A References

1. McKenzie FE, Bossert WH. An integrated model of *Plasmodium falciparum* dynamics. *J Theor Biol* 2005, 232: 411-426

2. Gu W, Killeen G, Mbogo CM, Regens JL, Githure JI, Beier JC. An individual-based model of *Plasmodium falciparum* malaria transmission on the coast of Kenya. *Trans R Soc Trop Med Hyg* 2003, 97: 43-50

3. Depinay JM, Mbogo CM, Killeen G, Knols B, Beier J, Carlson J, Dushoff J, Billingsley P, Mwambi H, Githure J, Toure AM, McKenzie FE. A simulation model of African Anopheles ecology and population dynamics for the analysis of malaria transmission. *Malar J* 2004, 3: 29

4. Rowe A and Steketee RW. Predictions of the impact of malaria control efforts on all-cause child mortality in sub-Saharan Africa *Am J Trop Med Hyg* 2007, 77 (6 Suppl): 48-55

5. Morel C, Lauer J, Evans D. Cost effectiveness analysis of strategies to combat malaria in developing countries. *BMJ* 2005, 331: 1299

6. Jones G, Steketee RW, Black RE, Bhutta ZA, Morris SS. How many child deaths can we prevent this year? *Lancet* 2003, 362: 65-71

7. Goodman CA, Coleman PG, Mills A. Economic analysis of malaria control in sub-Saharan Africa. Geneva: Global Forum for Health Research, 2000

8. Smith T, Maire N, Ross A, Penny M, Chitnis N, Schapira A, Studer A, Genton B, Lengeler C, Tediosi F, de Savigny D, Tanner M. Towards a comprehensive simulation model of malaria epidemiology and control. *Parasitol* 2008, in press

9. Yeung S, Pontavornpinyo P, Hasting I, Mills A, White N. Antimalarial drug resistance, artemisin-based combination therapy, and the contribution of modeling to elucidating policy choices. *Am J Trop Med Hyg* 2004, 71 (Suppl 2):1 79-186

10. Marsh K. Preface. *Am J Trop Med Hyg*. 2006, 75 (Suppl 2): i

11. Anderson RM, May RM. Infectious diseases of humans: dynamics and control. Oxford: Oxford University Press, 1991

12. Molineaux L, Dietz K. Review of intra-host models of malaria. *Parassitologia* 1999, 41: 221-231

13. Gravenor MB, Kwiatkowski D. An analysis of the temperature effects of fever on the intra-host population dynamics of *Plasmodium falciparum*. *Parasitol* 1998, 117 (Pt 2): 97-105

14. Kwiatkowski D, Nowak M. Periodic and chaotic host-parasite interactions in human malaria. *Proc Natl Acad Sci USA* 1991, 88: 5111-5113

15. Paget-McNicol S, Gatton M, Hastings I, Saul A. The *Plasmodium falciparum var* gene switching rate, switching mechanism and patterns of parasite recrudescence described by mathematical modelling. *Parasitol* 2002, 124: 225-235

16. Aron JL. Dynamics of acquired immunity boosted by exposure to infection. *Math Biosci* 1983, 64: 249-253

17. Gatton ML, Cheng Q. Modeling the development of acquired clinical immunity to *Plasmodium falciparum* malaria. *Infect Immun* 2004, 72: 6538-6545

18. Saul A. Testing the Vaccine. In: Good MF, Saul A, eds. Molecular immunological considerations in malaria vaccine development. Boca Raton: CRC Press 1994: 245-259

19. Dietz K, Raddatz G, Molineaux L. A mathematical model of the first wave of *Plasmodium falciparum* asexual parasitaemia in non-immune and vaccinated individuals. *Am J Trop Med Hyg* 2006, 75 Suppl 2: 46-55

20. Gatton M, Cheng Q. Investigating antigenic variation and other host-parasite interactions in *Plasmodium falciparum* infection in naive hosts. *Parasitol* 2004, 128: 367-376

21. Gupta S, Snow R, Donnelly CA, Marsh K, Newbold C. Immunity to non-cerebral severe malaria is acquired after one or two infections. *Nat Med* 1999, 5: 340-343

22. Gupta S, Snow R, Donnelly C, Newbold C. Acquired immunity and postnatal clinical protection in childhood cerebral malaria. *Proc R Soc Lond B Biol Sci* 1999, 266: 33-38

23. Macintosh CL, Beeson JG, Marsh K. Clinical features and pathogenesis of severe malaria. *Trends Parasitol* 2004, 20: 597-603

24. Rowe A, Steketee R, Arnold F, Wardlaw T, Basu S, Bakyaita N, Lama M, Winston C, Lynch M, Cibulskis R, Shibuya K, Ratcliffe A, Nahlen B, for the Roll Back Malaria Monitoring and Evaluation Reference Group. Evaluating the impact of malaria control efforts on mortality in sub-Saharan Africa. *Trop Med Int Health submitted*

25. Hastings I, korenromp E, Bloland P. The anatomy of a malaria disaster: drug policy choice and mortality in African children. *Lancet Infect Dis*. 2007, 7: 739-48

26. Haghdoost A, Alexander N, Smith T. Maternal malaria during pregnancy and infant mortality rate: critical literature review and a new analytical approach. *J Vector Borne Dis* 2007, 44: 98-104

27. Wort U, Hastings I, Mutabingwa T, Brabin B. The impact of endemic and epidemic malaria on the risk of stillbirth in two areas of Tanzania with different transmission patterns. *Malar J* 2006, 5: 89

28. Ross R. The prevention of malaria. 2nd ed. London: Murray, 1911

29. Macdonald G. The epidemiology and control of malaria. London: Oxford University Press, 1957

30. Dietz K, Molineaux L, Thomas A. A malaria model tested in the African savannah. *Bull World Health Org* 1974, 50: 347-357

31. Eichner M, Diebner HH, Molineaux L, Collins WE, Jeffery GM, Dietz K. Genesis, sequestration and survival of *Plasmodium falciparum* gametocytes: parameter estimates from fitting a model to malariatherapy data. *Trans R Soc Trop Med Hyg* 2001, 95: 497-501

32. McKenzie FE, Bossert WH. The dynamics of *Plasmodium falciparum* blood-stage infection. *J Theor Biol* 1997, 188: 127-140

33. Gatton M, Cheng Q. *Plasmodium falciparum* infection dynamics and transmission potential following treatment with sulfadoxine-pyrimethamine. *J Antimicrob Chemother* 2006, 58: 47-51

34. Paul R, Bonnet S, Boudin C, Tchuinkam T, Robert V. Aggregation in malaria parasites places limits on mosquito infection rates. *Inf Genet Evol* 2007, 7: 577-586

35. Koella JC. On the use of mathematical models of malaria transmission. *Acta Trop* 1991, 49: 1-25

36. McKenzie FE. Why model malaria? *Parasitol Today* 2000, 16: 511-516

37. Grimm V. Ten years of individual-based modelling in ecology: what have we learned and what could we learn in future? *Ecolog Modelling* 1999, 115: 129-148

38. Struchiner CJ, Halloran ME, Spielman A. Modeling malaria vaccines. I: New uses for old ideas. *Math Biosci* 1989; 94: 87-113

39. Cancre N, Tall A, Rogier C, Faye J, Sarr O, Trape JF, Spiegel A, Bois F. Bayesian analysis of an epidemiologic model of *Plasmodium falciparum* malaria infection in Ndiop, Senegal. *Am J Epidemiol* 2000, 152: 760-770

40. Carneiro I, Smith T, Lusingu J, Malima R, Utzinger J, Drakeley C. Modeling the relationship between the population prevalence of *Plasmodium falciparum* malaria and anemia. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 82-89

41. Maire N. Using volunteer computing to simulate the epidemiology and control of malaria: malariacontrol.net. Distributed & Grid Computing - Science made transparent for everyone. Principles, applications and supporting communities. Rechenkraft.net, in press

42. Taylor L, Begley J, Kupca V, Stefansson G. A simple implementation of the statistical modelling framework Gadget for cod in Icelandic waters. *Afr J Marine Sci* 2007, 29: 223-245

43. Cooper B, Lipsitch M. The analysis of hospital infection data using hidden Markov models. *Biostat* 2004, 5: 223-237

44. Smith T. Estimation of heterogeneity in malaria transmission by stochastic modelling of apparent deviation from mass action kinetics. *Malar J* 2008, 7: 12

45. Woolhouse ME, Dye C, Etard JF, Smith T, Charlwood JD, Garnett GP, Hagan P, Hii J, Ndhlovu PD, Quinnell RJ, Watts CH, Chandiwana SK, Anderson RM. Heterogeneities in the transmission of infectious agents: implications for the design of control programs. *Proc Natl Acad Sci USA* 1997, 94: 338-342

46. Schmid-Hempel P, Koella J. Variability and its implications for host-parasite interactions. *Parasitol Today* 1994, 10: 98-102

47. Dye C, Hasibeder G. Population dynamics of mosquito-borne disease: effects of flies which bite some people more frequently than others. *Trans R Soc Trop Med Hyg* 1986, 80: 69-77

48. Smith DL, McKenzie FE, Snow RW, Hay SI. Revisiting the basic reproductive number for malaria and its implications for malaria control. *PLoS Biol* 2007, 5: e42

49. Hill AV, Allsopp CE, Kwiatkowski D, Anstey NM, Twumasi P, Rowe P, Bennett S, Brewster D, McMichael AJ, Greenwood BM. Common west African HLA antigens are associated with protection from severe malaria. *Nature* 1991, 352: 595-600

50. Mackinnon M, Mwangi T, Snow R, Marsh K, Williams T. Heritability of malaria in Africa. *PLoS Med* 2005, 2: 340

51. Modiano D, Petrarca V, Sirima BS, Nebie I, Diallo D, Esposito F, Coluzzi M. Different response to *Plasmodium falciparum* malaria in West African sympatric ethnic groups. *Proc Natl Acad Sci USA* 1996, 93: 13206-13211

52. Williams TN, Mwangi TW, Wambua S, Alexander N, Kortok M, Snow RW, Marsh K. Sickle cell trait and the risk of *Plasmodium falciparum* malaria and other childhood diseases. *J Inf Dis* 2005, 192: 178-186

53. Nathan R, Masanja H, Mshinda H, Schellenberg JA, de Savigny D, Lengeler C, Tanner M, Victora CG. Mosquito nets and the poor: can social marketing redress inequities in access? *Trop Med Int Health* 2004, 9: 1121-1126

54. Keating J, Macintyre K, Mbogo C, Githure J, Beier JC. Self-reported malaria and mosquito avoidance in relation to household risk factors in a Kenyan coastal city. *J Biosoc Sci* 2005, 37: 767-771

55. Masanja H, Schellenberg J, de Savigny D, Mshinda H, Victora C. Impact of Integrated Management of Childhood Illness on inequalities in child health in rural Tanzania. *Health Policy Plan* 2005, Suppl 1: i77-i84

56. Armstrong Schellenberg J, Victora C, Mushi A, de Savigny D, Schellenberg D, Mshinda H, Bryce J, for the Tanzania IMCI MCE baseline household survey study group. Inequalities among the very poor: health care for children in rural southern Tanzania. *Lancet* 2003, 361: 561-566

57. Power C, Matthews S. Origins of health inequalities in a national populaiton sample. *Lancet* 1997, 350: 1584-1589

58. Mbaye A, Richardson K, Balajo B, Dunyo S, Shulman C, Milligan P, Greenwood B, Walraven G. A randomised, placebo-controlled trial of intermittent preventive treatment with sulphadoxine-pyrimethamine in Gambian multigravidae. *Trop Med Int Health* 2006, 11: 992-1002.

59. Maire N, Aponte JJ, Ross A, Thompson R, Alonso P, Utzinger J, Tanner M, Smith T. Modeling a field trial of the RTS,S/AS02A malaria vaccine. *Am J Trop Med Hyg* 2006, 75: 104-110

60. Rykiel E. Testing ecological models: the meaning of validation. *Ecolog Model* 1996, 90: 229-244.

61. Bailey N. The biomathematics of malaria. London: Charles Griffin and Co Ltd, 1982

62. Nedelman J. Inoculation and recovery rates in the malaria model of Dietz, Molineaux and Thomas. *Math Biosci* 1984, 69: 209-233.

63. Hodges J, Dewar J. Is it you or your model talking? A framework for model validation. 1992 Santa Monica, CA, RAND

64. Najera J. A critical review of the field application of a mathematical model of malaria eradication. *Bull WHO* 1974, 50: 449-457

65. Maire N, Tediosi F, Ross A, Smith T. Predictions of the epidemiologic impact of introducing a pre-erythrocytic vaccine into the expanded program on immunization in sub-Saharan Africa. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 111-118

66. Tediosi F, Hutton G, Maire N, Smith TA, Ross A, Tanner M. Predicting the cost-effectiveness of introducing a pre-erythrocytic malaria vaccine into the expanded program on immunization in Tanzania. *Am J Trop Med Hyg* 2006, 75 (Suppl 2): 131-143.

Appendix A.1

Equations and parameter estimates of the integrated model of malaria epidemiology

Infection of the human host[1]

$E_a(i,t)$, the age-adjusted EIR for individual *i* at time *t*, is given by

$$E_a(i,t) = E_{max}(t)\frac{A(a(i,t))}{A_{\max}} \qquad \text{(A.1)}$$

where, $A(a(i,t))$ is the average body surface area estimated for an individual of age $a(i,t)$ and $A_{\max}$ is the average surface area of people aged 20 years or more in the same population. E_{max} (t) refers to the usual measure of the EIR computed from human bait collections. The force of infection is then

$$\lambda(i,t) = E_a(i,t)(S_\infty + \frac{1-S_\infty}{1+\frac{E_a(i,t)}{E^*}})(S_{imm} + \frac{1-S_{imm}}{1+\left(\frac{X_p(i,t)}{X_p^*}\right)^{\gamma_p}}) \qquad \text{(A.2)}$$

where $S_{imm}, X_p^*, E^*, \gamma_p, S_\infty$ are constants (Table A.1) and

$$X_p(i,t) = \int_{t-a(i,t)}^{t} E_a(i,\tau)d\tau\,. \qquad \text{(A.3)}$$

The number of infections $h(i,t)$ introduced in time step *t*, is distributed as

$$h(i,t) \sim Poisson(\lambda(i,t)) \qquad \text{(A.4)}$$

Characteristics of the simulated infections[2]

Each new infection *j*, initiated in individual *i* at time t_0 is assigned a duration of $t_{\max}$, sampled from

$$\ln\left(t_{\max}(i,j)\right) \sim Normal(5.13, 0.80) \qquad \text{(A.5)}$$

The log density in the absence of previous exposure at each time point, $\tau = 0,1,...,t_{\max}(i,j)$ of the infection j in host i is then normally distributed with expectation

$$\ln(y_0(i,j,\tau)) = \ln d(i) + \ln\left(y_G\left(\tau,\tau_{\max}\right)\right) \qquad \text{(A.6)}$$

where $y_G\left(\tau,\tau_{\max}\right)$ is an empirical description of malariatherapy patients from the Georgia hospital and *d(i)* represents between-host variation drawn from a log-normal distribution with variance σ_i^2.

We measure exposure to asexual blood stages with

$$X_y(i,j,t) = \int_{t-a}^{t} Y(i,\tau) \quad d\tau \quad - \quad \int_{t_{0,j}}^{t} y(i,j,\tau) \quad d\tau \qquad \text{(A.7)}$$

where $Y(i,\tau)$ is the total parasite density of individual *i* at time τ and $y(i,j,\tau)$ is the density in individual *i* for infection *j* at time τ, and

$$X_h(i,t) = \int_{t-a}^{t} h(i,\tau) \quad d\tau \quad -1. \qquad \text{(A.8)}$$

The expected log density for each concurrent infection is then

$$E\left(\ln\left(y\left(i,j,\tau\right)\right)\right) = D_y D_h D_m \cdot \ln(y_0(i,j,\tau)) + \ln\left(\frac{D_x}{M(t)} + 1 - D_x\right) \qquad \text{(A.9)}$$

where *M(t)* is the total multiplicity of infection and

$$D_y = \frac{1}{1+\frac{X_y(i,j,t)}{X_y^*}}, \qquad \text{(A.10)}$$

$$D_h = \frac{1}{1+\frac{X_h(i,t)}{X_h^*}}, \qquad \text{(A.11)}$$

$$D_m = 1 - \alpha_m \exp\left(-\frac{0.693a}{a_m^*}\right) \qquad \text{(A.12)}$$

and X_y^*, X_h^*, D_x, a_m^*, and α_m, are further constants.

Variation within individual hosts is quantified by a term $\sigma_y^2(i,j,\tau)$, where

$$\sigma_y^2(i,j,\tau) = \frac{\sigma_0^2}{1+\frac{X_h(i,t)}{X_v^*}} \qquad \text{(A.13)}$$

and σ_0^2 and X_v^* are constants (Table A.1). The simulated densities are specified using

$$\ln(y(i,j,\tau)) \sim Normal(E(\ln(y(i,j,\tau))), \sigma_y^2(i,j,\tau)) \qquad \text{(A.14)}$$

The total density at time *t* in host *i* is then the sum of the densities of the various co-infections *j* i.e.

$$Y(i,t) = \sum_j y(i,j,\tau(i,j)) \qquad \text{(A.15)}$$

Model for infectivity of the human host (Chapter 5 and Killeen and others[3])

Let

$$\Upsilon(i,t) = \beta_1 Y(i,t-2) + \beta_2 Y(i,t-3) + \beta_3 Y(i,t-4) \qquad \text{(A.16)}$$

where *t* is in five-day units, and

$$\ln(y_g(i,t)) \sim \text{Normal}(\ln(\rho\Upsilon(i,t))), \sigma_g{}^2) \qquad \text{(A.17)}$$

where $\beta_1, \beta_2, \beta_3, \rho, \sigma_g{}^2$ are constants (Table A.1). Define

$$\begin{aligned} \Pr(y_g(i,t) > y_g^*) &= \Phi\left[\frac{\ln\left(\rho\Upsilon(i,t)\right) - \ln(y_g^*)}{\sigma_g}\right] \\ &= \Phi\left[\frac{\ln\left(\Upsilon(i,t)\right)}{\sigma_g} - \rho^*\right] \end{aligned} \qquad \text{(A.18)}$$

where Φ is the cumulative normal distribution. Then the proportion of mosquitoes that are infected feeding on individual *i* at time *t* is

$$I_m(i,t) = \left[\Pr(y_g(i,t) > y_g^*)\right]^2 \qquad \text{(A.19)}$$

and the probability that a mosquito becomes infected at any feed is

$$\kappa_u(t) = \eta \frac{\sum_i \left(A(a(i,t)) I_m(i,t) \right)}{\sum_i A(a(i,t))} \quad \text{(A.20)}$$

where η is a constant scale factor.

Define $\kappa_u^{(0)}(t)$ as the value of $\kappa_u(t)$ in the simulation of an equilibrium scenario to which an intervention has been applied. Let $E_{\max}^{(0)}(t+l_v)$ be the corresponding entomological inoculation rate. $\kappa_u^{(1)}(t)$ and $E_{\max}^{(1)}(t+l_v)$ are the corresponding values for the intervention scenario. Then

$$E_{\max}^{(1)}(t+l_v) = \frac{E_{\max}^{(0)}(t+l_v)\ \kappa_u^{(1)}(t)}{\kappa_u^{(0)}(t)} \quad \text{(A.21)}$$

where l_v corresponds to the duration of the sporogonic cycle in the vector, which we approximate with ten days. ($E_{\max}^{(0)}(t+l_v)/\kappa_u^{(0)}(t)$ is the total vectorial capacity).

Acute morbidity (Chapter 2)

An episode of acute morbidity occurs in individual *i*, at time *t*, with probability

$$P_m(i,t) = \frac{Y_{\max}(i,t)}{Y^*(i,t) + Y_{\max}(i,t)} \quad \text{(A.22)}$$

where Y^* is the pyrogenic threshold and $Y_{\max}$ is the maximum density of 5 daily densities sampled during the 5-day time interval *t*. The pyrogenic threshold evolves over time via

$$\frac{dY^*(i,t)}{dt} = \frac{\alpha Y(i,t)}{\left(Y_1^* + Y(i,t)\right)\left(Y_2^* + Y^*(i,t)\right)} - \varpi Y^*(i,t) \quad \text{(A.23)}$$

with the initial condition $Y^*(i,0) = Y_0^*$ at the birth of the host and α, ϖ, Y_1^*, and Y_2^* are constants.

We consider two different classes of severe episodes, B_1 and B_2. $P_{B_1}(i,t)$ is the probability that an acute episode is a class B_1 severe episode and is specified using

$$P_{B_1}(i,t) = \Pr\left(\mathrm{H}(i,t) \in \mathrm{B}_1 \mid \mathrm{H}(i,t) \in \mathrm{A}\right) = \frac{Y_{max}(i,t)}{Y_{B_1}^* + Y_{max}(i,t)} \quad \text{(A.24)}$$

where $Y_{B_1}^*$ is a constant and $\mathrm{H}(i,t)$ is the clinical status.

The second, non-intersecting, subset of severe malaria episodes (B_2) occur when an otherwise uncomplicated malaria episode happens to coincide with some other insult, which occurs with risk

$$F(a(i,t)) = \frac{F_0}{1+\left(\frac{a(i,t)}{a_F^*}\right)} \qquad \text{(A.25)}$$

where F_0 is the limiting value of $F(a(i,t))$ at birth, and a_F^* is the age at which it is halved.

The probability that an episode belonging to class B_2 occurs at time t, conditional on there being a clinical episode at that time is $P_{B_2}(i,t)$ where

$$P_{B_2}(i,t) = \Pr(\mathrm{H}(i,t) \in \mathrm{B}_2 | \ \mathrm{H}(i,t) \in \mathrm{A}) = F(a(i,t)) \qquad \text{(A.26)}$$

The age and time specific risk of severe malaria morbidity conditional on a clinical episode is then given by

$$P_B(i,t) = P_{B_1}(i,t) + P_{B_2}(i,t) - P_{B_1}(i,t)P_{B_2}(i,t), \qquad \text{(A.27)}$$

Mortality (Chapters 3 and 4)

Malaria deaths in hospital are a random sample of those severe malaria cases deemed to be admitted, with age-dependent sampling fraction $Q_h(a)$, the hospital case fatality rate, derived from the data of Reyburn and others.[4]

We estimate the severe malaria case fatality in the community, $Q_c(a)$ for agegroup a with

$$Q_c(a) = \frac{Q_h(a)\varphi_1}{1-Q_h(a)+Q_h(a)\varphi_1}, \qquad \text{(A.28)}$$

where φ_1, the estimated odds ratio for death in the community compared to death in in-patients, is an age-independent constant and $Q_h(a)$ is the hospital case fatality rate. Malaria mortality is the sum of the hospital and community malaria deaths. The risk of neonatal mortality attributable to malaria (death in class D_1) in first pregnancies is set equal to $0.3\mu_{PG}$ where μ_{PG} is given by

$$\mu_{PG} = \mu_{\max}\left[1-\exp\left(-\frac{x_{PG}}{x^*_{PG}}\right)\right], \qquad \text{(A.29)}$$

where x_{PG} is related to x_{MG}, the prevalence in simulated individuals of age 20-24 years via

$$x_{PG} = 1-\frac{1}{1+\left(\frac{x_{MG}}{x^*_{MG}}\right)} \qquad \text{(A.30)}$$

and x^*_{MG} and x^*_{PG} are constants (Table A.1).

An indirect death in class D_2 is provoked at time *t*, conditional on there being a clinical episode at that time, with probability $P_{D_2}(i,t)$ where

$$P_{D_2}(i,t) = \Pr(\mathrm{H}(i,t) \in \mathrm{D}_2 \mid \mathrm{H}(i,t) \in \mathrm{A}) \text{ and}$$
$$P_{D_2}(i,t) = \frac{Q_D}{1+\left(\frac{a(i,t)}{a^*_F}\right)} \qquad \text{(A.31)}$$

where Q_D is limiting value of $P_{D_2}(i,t)$ at birth and a^*_F is a constant. Deaths in class D_2 occur 30 days (6 time steps) after the provoking episodes.

Anaemia[75]

The prevalence of anaemia, $p_A(a,t)$, in age group with mid-age *a*, at time *t* is specified by

$$\mathrm{logit}\left(p_A(a,t)\right) = \beta_{a0} + \frac{\beta_{a2}a^*}{a^*+a} + \frac{\beta_P p_P(a,t)}{p^*+p_P(a,t)} + \beta_I p_P(a,t)\log(a) \qquad \text{(A.32)}$$

where $p_P(a,t)$ is the prevalence of patent parasitaemia in the age group and $\beta_0, \beta_P, p^*, \beta_{a1}, a^*, \beta_I$ are constants.

Table A.1. Model parameter values

Parameter	Description	Units/ dimension	Published values, 2006‡	Values Ch. 6* model 1	Values Ch 6* model 4
S_∞	Lower limit of success probability of inoculations at high $E_a(i,t)$	Proportion	0.049	0.049	0.049
E^*	Critical value of $E_a(i,t)$	Inoculations/ person-night	0.032	0.032	0.032
S_{imm}	Lower limit of success probability of inoculations in immune individuals	Proportion	0.14	0.14	0.14
γ_p	Steepness of relationship between success of inoculation and $X_p(i,t)$	Dimensionless constant	2.04	2.06	2.04
X_p^*	Critical value of cumulative number of entomological inoculations	Inoculations	1514.4	2801.5	2116.3
X_h^*	Critical value of cumulative number of infections	Infections	97.3	97.8	94.8
X_y^*	Critical value of cumulative number of parasite days	Parasite-days/µl x 10^{-7}	3.5	13.8	6.5
X_v^*	Critical value of cumulative number of infections for variance in parasite densities	Infections	0.92	0.92	0.92
a_m^*	Decay of maternal protection	Per year	2.53	2.59	2.51
D_x	Effect of concurrent co-infections	Infections	0	0	0
β_1	Effect of asexual density (lag 10 days) on expected gametocytaemia (fixed)	Dimensionless	1	1	1
β_2	Effect of asexual density (lag 15 days) on expected gametocytaemia	Dimensionless	0.46	0.46	0.46
β_3	Effect of asexual density (lag 20 days) on expected gametocytaemia	Dimensionless	0.17	0.17	0.17
ρ	Location parameter for the distribution of the ratio of gametocytes to asexual parasites	Dimensionless	0.00031	0.00031	0.00031
η	Scale factor for probability that a mosquito becomes infected at any feed	Dimensionless	0.56	0.56	0.56

Parameter	Description	Units/ dimension	Published values, 2006‡	Values Ch. 6* model 1	Values Ch 6* model 4
σ_g	Standard deviation of the distribution of the ratio of gametocytes to asexual parasites	Dimensionless	3.91	3.91	3.91
α	Factor determining increase in $Y^*(i,t)$	Parasites^{2}µl^{-2}day^{-1}	143,000	157,000	179,000
ϖ	Decay rate of pyrogenic threshold	Year^{-1}	2.5	2.5	2.5
Y_0^*	Pyrogenic threshold at birth	Parasites/µl	296.3	328.1	244.3
Y_1^*	Critical value of parasite density in determining increase in Y^*	Parasites/µl	0.60	0.60	0.59
Y_2^*	Critical value of $Y^*(i,t)$ in determining increase in $Y^*(i,t)$	Parasites/µl	6502.3	6502.3	6502.3
β_f	Contribution of five day parasitaemia to acquired immunity in the presence of a fever	Proportion	-	-	0.80
$Y_{B_1}^*$	Parasitemia threshold for severe episodes type B_1	Parasites/µl	784,000	347,000	258,000
F_0	Prevalence of co-morbidity/susceptibility at birth relevant to severe episodes (B_2)	Proportion	0.092	0.099	0.094
a_F^*	Critical age for co-morbidity	Years	0.117	0.116	0.119
φ_1	Case fatality for severe episodes in the community compared to hospital	Odds ratio	2.09	2.07	2.09
Q_n	Non-malaria intercept for infant mortality rate	Deaths/1000 livebirths	49.5	50.6	52.0
Q_D	Co-morbidity intercept relevant to indirect mortality	Proportion	0.019	0.018	0.017
x_{MG}^*	Critical value of the simulated prevalence for ages 20-25 years	Proportion	0.19	0.19	0.19
$\mu_{\max}$	Upper limit of risk of neonatal mortality in primigravidae	Proportion	0.011	0.011	0.011
x_{PG}^*	Critical value of prevalence for neonatal mortality risk	Proportion	0.25	0.25	0.25
β_0	Intercept	Log odds	-6.13	-6.13	-6.13

Parameter	Description	Units/ dimension	Published values, 2006‡	Values Ch. 6* model 1	Values Ch 6* model 4
β_P	Effect of parasite prevalence	Log odds	12.5	12.5	12.5
p^*	Critical value of parasite prevalence	Proportion	2.84	2.84	2.84
β_{a1}	Magnitude of age effect	Per year	3.14	3.14	3.14
a^*	Critical age	Years	3.66	3.66	3.66
β_I	Age-prevalence interaction effect	Log odds	-0.75	-0.75	-0.75

‡ Parameter values were estimated separately for each outcome, and in some cases were conditional on the values of parameters in model components (Am J Top Med Hyg 2006, Suppl 2).

* Parameter values for variables marked with † were estimated simultaneously using distributed computing via the internet. They were harvested on 8 August 2007 from malariacontrol.net

References for Appendix A.1

1. Smith T, Maire N, Dietz K, Killeen GF, Vounatsou P, Molineaux L, Tanner M. Relationship between the entomologic inoculation rate and the force of infection for *Plasmodium falciparum* malaria. *Am J Trop Med Hyg* 2006, 75: 11-18

2. Maire N, Smith T, Ross A, Owusu-Agyei S, Dietz K, Molineaux L. A model for natural immunity to asexual blood stages of *Plasmodium falciparum* malaria in endemic areas. *Am J Trop Med Hyg* 2006, 75: 19-31

3. Killeen GF, Ross A, Smith T. Infectiousness of malaria-endemic human populations to vectors. *Am J Trop Med Hyg* 2006, 76 (Suppl 2): 38-45

4. Reyburn H, Drakeley C, Carneiro I *et al* The epidemiology of severe malaria due to Plasmodium falciparum at different transmission intensities in NE Tanzania. LSHTM Malaria Centre Report 2002-2003. 2004:6-7