MATERIALS AND METHODS

Data. The data used for fitting the baseline model were collected by the U.S. Public Health Service in Afro-American neurosyphilitic adults who were not previously exposed to malaria when malariatherapy was a recommended treatment for neurosyphilis.6 Several strains of P. falciparum were used and inoculated by the transfusion of blood from an infective gametocyte carrier or by the bites of infective vectors. We used discrete-time modeling with two-day steps; previous observations showed that parasitemias on odd and even days (defining the first day of patent parasitemia as day one) were strongly correlated, but odd day maximal densities were on average somewhat higher.5 We used observed parasite densities of each case over the course of the first wave of parasitemia as target points to be fitted. The maximum of the first wave was defined as the first density that is higher than all preceding ones, and not lower than any of the two following ones. The first wave ends with the monotone decrease after that maximum. A case was included in the fitting if its first wave satisfied the following criteria: 1) The maximum of the first wave is the absolute maximum of the case; 2) no treatment (curative or suppressive); 3) no missing parasitologic data on odd days; 4) identification of fever days; 5) a minimum of five data points; 6) first observed density less than 320/µL (conjecturing that otherwise the first patency was missed); 7) a log (maximum/last density) > 0.6 (to provide enough information about the downward slope); and 8) log (last/penultimate density) < 0.3 (to exclude cases in which the second wave follows too closely the first wave). Of 334 P. falciparum primary infections, 100 satisfied all criteria. Their distribution among strains was as follows: McLendon = 50, El Limon = 25, Santee Cooper = 21, Colombia = 3, and Costa = 1. Sixty-seven cases had been inoculated by blood and the remaining 33 were inoculated by vectors.

Our second selection criterion (no treatment) truncates the data: the cases it disqualifies must have been more severe than average. We imposed the criterion because we wanted a realistic model of the natural history of the first wave of asexual parasitemia. The truncation should be taken into account in interpreting simulations concerning severe disease.

Microscopic blood examination was performed almost daily, by the method of Earle and Perez, with a detection level of approximately 10 infected erythrocytes (IEs)/µL of blood.7 Temperature was checked every four hours routinely and every hour during paroxysms. Antimalarial treatments were used as follows. A curative treatment (usually chloroquine to which the parasites used were uniformly sensitive) was given on discharge or earlier if the clinician in charge decided that the severity of the malaria required its prompt termination; such emergency curative treatments were given in less than 5% of the cases, and nearly always during the first wave of parasitemia, which confirms the exceptional pathogenicity of the first wave. Small single doses of antimalarial drugs, most commonly quinine, whose action is fast and short, were given for partial suppression of the infection, again as decided by

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Abstract. We present a dynamic model of the highly pathogenic first wave of Plasmodium falciparum asexual parasitemia in non-immune persons. The model was successfully fitted to malaria therapy data. This required four case-specific parameters: the basic two-day multiplication factor, the time of onset of adaptive immunity, and the effective dose 50 densities for the innate and adaptive immune responses, respectively. All four parameters show large case-dependent variation that is mainly attributable to host factors. According to the model, the maximum value of the first wave is controlled mainly by the innate immune response. We used the model to explore the expected effects of vaccines targeting the parasite’s asexual blood stages on the basis of what we consider to be the biologically most plausible assumptions concerning the parameter modifications induced by vaccination. According to our simulations, the benefit of antiparasitic vaccination is strongly host dependent and vaccine efficacy at low immunogenicity is much larger against severe disease than against fever. This has implications for the early testing of the protective efficacy of a vaccine in humans.

INTRODUCTION

Plasmodium falciparum is a major cause of disease and death. Its control by currently available methods is not satisfactory, and the prospect of vaccination raises new hopes. A number of different vaccines that target the pre-erythrocytic stages of the parasite, the asexual blood stages (the parasite or its toxin[s]), or the sexual stages, alone or in various combinations, are at various stages of development.1-3 Plasmodium falciparum causes disease through its asexual parasitemia, which if not interrupted by death or treatment, usually displays an irregular wave-like periodicity with a median interval of approximately 20 days between successive local maxima, and an overall downward trend.4,5 The risk of severe (life-threatening) disease is strongly concentrated in non-immune persons, in particular during the first wave of parasitemia after a primary infection. Therefore, the first wave should be a major target of asexual blood stage antiparasitic vaccines (APVs). We present a mathematical model of that first wave, including options representing the possible biologic effects of such a vaccine; the fitting of the baseline model to malaria therapy data; and a selection of simulations exploring the expected effects of vaccination at the individual level.
the clinician in charge; approximately half of the patients received one or more such suppressive treatments.

**Basic model (without vaccination effects).** The model derives from our model of the full course of parasitemia in a non-immune individual. The central equation of that model includes antigenic variation and a basic two-day multiplication factor plus three immune responses (IRs) controlling the growth of the parasite population: 1) an innate IR representing the effect of the pro-inflammatory cytokine cascade (interferon-γ, tumor necrosis factor-α [TNF-α]); 2) an adaptive variant-specific IR representing the effect of variant-specific antibodies to *P. falciparum* erythrocyte membrane protein 1; and 3) an adaptive variant-transcending IR representing the effect of all other anti-sexual blood-stage antibodies.

We conjectured that if we restricted the model to the first wave of parasitemia in non-immune persons, we could simplify the description of naturally adaptive immunity by 1) reducing it to a single dimension, without distinction between variant-specific and variant-transcending IRs; and 2) ignoring its decay. The basic equation is

\[ P(t + 2) = P(t) m S_c(t) \frac{1 - P(t)}{P^*}, \]

where \( P(t) \) is the asexual parasite density (IEs/μL), \( t \) is the time (in days) elapsed since emergence from the liver, \( m \) is the two-day multiplication factor in the absence of any immunity, and \( S_c(t) \), \( S_m(t) \) are the probabilities to survive the interval \((t, t + 2)\), expressing, respectively, the effects of innate and adaptive IRs.

The effect of the innate IR is a function of the present parasite density

\[ S_i(t) = \frac{1}{1 + \frac{P(t)}{P^*}}, \]

where \( P^* \) is the critical parasite density at which the current multiplication factor is reduced by 50%.

The adaptive IR is a function of the cumulative parasite density; this function is determined by two host-specific parameters and one constant: 1) \( P_m^* \) (host-specific) is the critical cumulative parasite density at which the current multiplication factor is reduced by 50%; 2) \( \Delta_o \) (host-specific) is the delay required by adaptive immunity to become effective; for times before \( \Delta_o \) the cumulative density is set to zero, i.e., the adaptive IR has no effect and \( S_m(t) = 1 \); and 3) \( \Delta_1 = 8 \) days (constant for all cases) is the delay that determines for all cases the last term in the cumulative density for times \( t \geq \Delta_o \), i.e., for those times the cumulative density comprises all parasite densities from time zero until time \( t - 8 \):

\[ S_m(t) = \frac{1}{1 + \frac{P(t)}{P^*}} \frac{(P(0) + P(2) + \ldots + P(t - 8))}{P_m^*}. \]

Thus, the innate IR is a fast labile response to the current density. The adaptive IR is a delayed and lasting response to a cumulative density, involving the production of antigen-specific effectors. The two simple IRs in the model try to capture the succession of the two complex natural responses observed in malaria.

To simulate the impact of vaccines on fever, we need a fever threshold parasite density \( (P_f^*) \). Jeffery and others have shown, using the daily data from the same database, that the density on the first fever day varied among cases by more than four orders of magnitude and that the variation was much larger within than between parasite strains. All 100 cases had fever. We extracted for each case from the daily data the maximum asexual density up to the first fever day (temperature > 101°F = 38.3°C). We set the observed \( P_f^* \) equal to the highest odd day density up to the first fever day, odd or even. (If we would have restricted the determination of the fever threshold to the odd days, then there would have been seven patients without fever.) As we shall describe in the Results, the logarithm of the ratio of \( P_f^* \) divided by the maximum parasite density follows a uniform distribution. This finding will be used to simulate mild malaria when we explore the effects of an antiparasitic vaccine.

**Method of fitting.** We used as fitting targets the observed case-specific odd-day asexual parasite densities over the course of the first wave of asexual parasitemia. We assumed that the initial value (time zero) for the simulation is at a level of 0.003 IEs/μL, which corresponds to a parasite load of 15,000 IEs in 5 liters. The time interval between time zero and the onset of patent parasitemia is set equal to eight days for all patients; simulated and observed values are displayed on even days. Since the time for the onset of adaptive immunity is a discrete variable that is restricted to multiples of two days, this parameter cannot be estimated by the standard method of least squares. Therefore, we first fitted a simple piecewise linear statistical model by least squares for the logarithms of the observed parasitemia, where the inflection points could take on arbitrary values. This model assumed a linear increase followed by a period with a constant plateau and a subsequent linear decrease. The estimated time of the inflection point for the onset of the decrease was rounded to the nearest even integer and taken as the case-specific delay for the onset of adaptive immunity. We assumed that on this day the cumulative number of parasites up to eight days before the onset of the adaptive immunity is added to determine the effect of adaptive immunity. The remaining three parameters of the dynamic model, i.e., the basic multiplication factor and the two 50% effective dose 50 (ED50) values for the two IRs are fitted using a Java implementation of the Powell hill-climbing algorithm.

**Generation of a random sample of 2,000 virtual cases.** To simulate the effect of vaccination, we generated a random sample of 2,000 individuals from a five dimensional log-normal distribution that has the same location and correlation parameters as obtained from fitting. The standard deviations were reduced in such a way that the range of the simulated cases corresponds to the observed sample because in a larger sample with the same standard deviations the range would have been increased. The fever threshold \( P_f^* \) for each of the 2,000 individuals was calculated by multiplying the maximum parasite density with a factor between 0.0002 and 1 in such a way that the logarithm of this factor has a uniform distribution between \( \log_{10}(0.0002) \) and 0. This distribution gave an excellent fit for the distribution of the observed ratios of the fever thresholds divided by the observed maximum (see Results). This choice of the fever threshold ensured that at the baseline level all 2,000 individuals had fever during the first wave, as was also true for the cases whose parameters were used to generate the characteristics of the virtual population.

**Modeling the biologic effects of APVs.** On the basis of the
known characteristics of adaptive antibody-mediated immunity, APVs targeting the asexual blood-stages are expected to control parasite population growth through three effects: 1) production of relatively short-lived antibodies, 2) shortening the delay of onset of the adaptive IR to infection, and 3) strengthening of that IR (per unit of antigenic trigger). Effects 2) and 3) represent relatively long-lived immune memory, substituting a secondary for a primary IR. The three effects are simulated by the parameters $f_A$, $f_{ \Delta \alpha}$, and $f_{\alpha}$ respectively, which vary between zero and one. Since the individual delay for the onset of adaptive immunity can take on only even integer values, the product $f_{\Delta \alpha} \Delta_0$ is rounded accordingly. From this rounded delay onwards the innate IR is given by the formula

**Figure 1.** Course of observed (crosses) and fitted (continuous lines) parasitemia and immune response functions [$S_c(t)$ (broken lines) and $S_m(t)$ (hashed lines)] in five cases (A, S1018; B, S1102; C, S1235; D, S732; E, G408) selected for diversity. See Table 1 for the parameters and observed and computed maxima of these cases. IEs = infected erythrocytes.
\[
S_m(t) = \frac{1}{P(0) + P(2) + \ldots + P(t-8) + \left( \frac{1}{T_A - 1} \right) P_m^{t/7}}.
\]

As the immune parameter \( f_A \) decreases, the antibody effect increases; we assume that the antibodies are proportional to the power 5/7 of the \( ED_m \) value \( P_m^c \) of the adaptive IR. This ensures that individuals with low \( ED_m \) values have a larger vaccine efficacy with respect to the initial multiplication factor.

**Modeling the incidence of severe disease.** As indicator of the severity of a simulated case, we use its maximum circulating asexual parasite density on the basis of the following points. First, the question is not whether our indicator of severe malaria is equivalent to a valid diagnosis of severe malaria in a given individual, but whether it is valid for comparing populations with respect to their relative incidences of severe malaria. Second, we conjecture with most (but not all) experts that severe malaria in any of its clinical forms is very unlikely in the absence of a large total parasite biomass (circulating plus sequestered) at the time the severe malaria is diagnosed or in the recent past (e.g., in the previous two weeks). However, a critical review of the evidence for and against that conjecture would require a separate report. Third, methods currently being developed for the measurement of the total parasite biomass are not applicable in this context. We therefore need an acceptable proxy. Fourth, over a period of time, the circulating parasite biomass must be correlated with the sequestered parasite biomass. Fifth, the maximum observed density is a good indicator of the cumulative patent density: its correlation (after log transformation) with the cumulative remainder of the first wave in the 100 cases used for the present investigation is 0.89 (\( P < 0.0001 \)); its correlation (after log transformation) with the cumulative remainder of the full course of parasitemia in the previously studied set of 35 cases followed up to spontaneous cure is 0.72 (\( P < 0.0001 \)).

As common threshold density defining a severe case \( (P_c^*) \) we used the 95th percentile of the 2,000 simulated cases’ maximum density (i.e., \( 123,227 \) IEs/\( \mu L \)). Given the truncation of the underlying data (see above), that threshold is unrealistically low. Nevertheless, we assume that it is acceptable for identifying the most severe cases. There are three simulated cases among the 100 cases (i.e., 5% of 2,000) that satisfy the criterion of severe case whose fever threshold is larger than the common threshold for severe disease.

**RESULTS**

**Fitted model.** The model fitted the very diverse case-specific observations quite well, as illustrated for five cases in Figure 1, which also shows the two corresponding IR functions \( S_m(t) \) and \( S_m(t) \), and how they control the maximum density and the downward slope, respectively. The characteristics of these five patients are given in Table 1. The density curve mirrors the curve of \( S_m \). Innate immunity imposes a ceiling on the density and adaptive immunity (the \( S_m \) curve) determines the slope of the subsequent decrease. The time of onset of adaptive immunity is clearly identified when \( S_m \) is starting to be less than one.

<table>
<thead>
<tr>
<th>TABLE 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Characteristics of the five patients in Figure 1</td>
</tr>
<tr>
<td>Patient</td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>S1018</td>
</tr>
<tr>
<td>S1102</td>
</tr>
<tr>
<td>S1235</td>
</tr>
<tr>
<td>S732</td>
</tr>
<tr>
<td>G408</td>
</tr>
</tbody>
</table>

The overall fit of the 100 cases was remarkably good. The 712 data points had a standard deviation of the logarithmic residuals of 0.26, which corresponds to a multiplication factor of 1.82. The modeling explains 95% of the variance of the observations. It is based on 400 parameter estimates, i.e., four per patient.

In 97 of the 100 cases, the good fit required both IRs. In the remaining three, which were characterized by an exponential growth determined by four observations, followed by one more observation far below the maximum, a good fit was obtained without innate IR. These may represent a few exceptionally efficient adaptive IRs. We excluded them when we estimated the parameters for simulating vaccine effects.

Table 2 summarizes the distributions of the observed maximum densities and of the four fitted parameters. All have a wide range and a log-normal distribution. Since the data represent several parasite strains, two inoculation routes, and two U.S. Public Health Service institutions, we tested for possible effects of these factors on the parameter estimates. Table 3 compares the McLendon strain to the pool of the others. The \( P_c^* \), \( P_m^* \), and maximum \( P(t) \) values of the McLendon strain are significantly smaller than those of the pool. The factors are unfortunately confounded. The McLendon was used only in South Carolina, and comparison between places for the strains used in both places can account for the apparent effect of strain on \( P_c^* \) and max \( P(t) \). Table 4 compares the same parameters with respect to the mode of infection. We see no significant difference between infection by blood and by mosquito if one takes into account that six tests have been performed.

Table 5 shows the correlations among maximum \( P(t) \), \( m \), \( P_c^* \), \( P_m^* \), \( \Delta_0 \), and the ratio of the fever threshold and the maximum observed density. The maximum \( P(t) \) is highly correlated with \( P_c^* \). There is no correlation between \( m \) and \( P_c^* \), between \( m \) and \( P_m^* \), or between \( P_c^* \) and \( P_m^* \). There is an expected significant negative correlation between the fever ratio and maximum \( P(t) \), \( P_c^* \), and the delay. We tested the predictability of the observed first local maximum density by multiple linear regression using the four fitted parameters.

**TABLE 2**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Minimum</th>
<th>Median</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Observed maximum density</td>
<td>1,780</td>
<td>38,000</td>
<td>251,280</td>
</tr>
<tr>
<td>Multiplication factor (m)</td>
<td>6.4</td>
<td>12.5</td>
<td>24.3</td>
</tr>
<tr>
<td>Innate IR 50 ( (P_c^*) )</td>
<td>174</td>
<td>2,755</td>
<td>46,789</td>
</tr>
<tr>
<td>Adaptive IR 50 ( (P_m^*) )</td>
<td>0.008</td>
<td>20.4</td>
<td>21.386</td>
</tr>
<tr>
<td>Adaptive IR delay ( (\Delta_0) )</td>
<td>12</td>
<td>16</td>
<td>22</td>
</tr>
</tbody>
</table>

* IR = immune response.
The result is shown in Figure 2. The fit is remarkably good: 94% of the variability is explained by the four terms. The $P$ values for all four regression coefficients are at most 0.0005. The maximum parasitemia is predicted by the equation $max P = 0.039 m^{1.55} \Delta D^{0.12} P_{m}^{0.94} P_{m}^{0.033}$.

Distribution of the fever ratio. Figure 3 shows the observed and the theoretical cumulative distribution function of the fever ratio (fever threshold divided by the maximum parasitemia) on a logarithmic scale. We observe a good fit with a uniform distribution in the interval from 0.0002 to 1. This implies that the median ratio of $P_{f}^{*}$ divided by the maximum density is approximately 1.4%.

Simulating the effects of vaccines. Table 6 shows the values assigned to the model’s vaccination parameters. The lowest factors used were those found by trial and error to achieve 100% protection against both fever and severe disease in the virtual population of 2,000 individuals. The intermediate values are powers of the fourth root.

Figure 4 compares the course of parasitemia of cases S1018 (lowest $P_{m}^{*}$) and S732 (with a combination of high $P_{m}^{*}$ and $P_{m}^{*}$) to what it would have been if the cases had been immunized at increasing levels of immunogenicity (Table 6). The efficacy of the vaccine differs greatly between the two cases. At any particular level, the effect on initial slope and maximum density is much greater in case S1018 than in case S732. For case S1018, the initial slope is negative at levels three and four while for case S732 the initial slope always remains positive. For increasing levels of immunogenicity, the time of peak is shifted to the right until the initial slope becomes negative.

Figure 5 shows in the virtual population of 2,000 median and range of the initial multiplication factors by immunogenicity level (Table 6). The standard deviation (on a logarithmic scale) increases with the immunogenicity level.

Figure 6 shows in the virtual population of 2,000 vaccine efficacies (VEs) against fever and severe malaria for the vaccination parameters given in Table 6, given either all three vaccine effects, only the two memory effects, or only the effect on delay of onset of adaptive IR. The main outcomes are 1) in all comparisons VE increases much faster against severe malaria than against fever; 2) even after the decay of antibodies, memory (including shortening of the delay and strengthening the response per unit of antigen) can still give protection; 3) the shortening of the delay is the main single factor of VE against severe malaria; immunogenicity level 2, which only reduces the range of delays from 12–22 days to 8–16 days, already results in a VE of 80% against severe disease.

**DISCUSSION**

Adults with neurosyphilis are not the ideal population in which to study a problem that affects mainly African children. However, they are the only population in which a detailed follow-up of untreated *P. falciparum* infections will ever be available for obvious ethical reasons. The degree to which they are representative of African children cannot be estimated in any ethical way. Another limitation of the data is that we assume, like all other investigators that have used those data, that the infections are monoclonal. In spite of this, these data have made a unique contribution to our knowledge of *P. falciparum* infections in humans, and continue to be used by many investigators to address additional important questions concerning that infection.

The production of malaria models has been relatively abundant and is currently accelerating. Our 1999 review of intrahost models of malaria reviews 18 malaria models, as well as five related African trypanosomiasis models, all published from 1989 to 1998, except for the seminal trypanosomiasis model of Kosinski (1980). In May 2006, a Scopus search using the terms Math*, Model*, Malaria from 1999 until the present time yielded 81 reports among which six used the same data base as the present report. The most productive author is McKenzie, who provides 15 self-quotations on malaria modeling. Tom Smith called our attention to a book chapter by Saul (1994) that we had missed in our review article. Note that the number of genuinely different malaria models is smaller than the number of papers contributing to malaria modeling. For instance among the 18 malaria models reviewed in our 1999 paper, the first seven and the last nine are sets of variants of only two genuinely different models. We found only three published malaria modeling papers spe-

### Table 3
Comparison of the geometric means between the McLendon strain and other strains

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Maximum $P(t)$</th>
<th>$m$</th>
<th>$P_{m}^{*}$</th>
<th>$P_{m}^{*}$</th>
<th>$\Delta D$</th>
<th>$P_{m}^{*}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>McLendon</td>
<td>49</td>
<td>27,245</td>
<td>12.3</td>
<td>1,879</td>
<td>7.9</td>
<td>16.3</td>
<td>379</td>
</tr>
<tr>
<td>Others</td>
<td>48</td>
<td>55,773</td>
<td>12.9</td>
<td>3,863</td>
<td>37.6</td>
<td>16.9</td>
<td>720</td>
</tr>
<tr>
<td>$P$-value</td>
<td>0.0003</td>
<td>0.4</td>
<td>0.0003</td>
<td>0.025</td>
<td>0.18</td>
<td>0.18</td>
<td></td>
</tr>
<tr>
<td>% of total variation explained by strain</td>
<td>13</td>
<td>0.7</td>
<td>13</td>
<td>5</td>
<td>2</td>
<td>2</td>
<td></td>
</tr>
</tbody>
</table>

**DISCUSSION**

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### Table 4
Comparison of the geometric means between the infections by mosquito and blood

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Maximum $P(t)$</th>
<th>$m$</th>
<th>$P_{m}^{*}$</th>
<th>$P_{m}^{*}$</th>
<th>$\Delta D$</th>
<th>$P_{m}^{*}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>67</td>
<td>35,374</td>
<td>12.7</td>
<td>2,483</td>
<td>7.9</td>
<td>16.3</td>
<td>513</td>
</tr>
<tr>
<td>Mosquitoes</td>
<td>50</td>
<td>47,854</td>
<td>12.3</td>
<td>3,195</td>
<td>37.6</td>
<td>16.9</td>
<td>537</td>
</tr>
<tr>
<td>$P$-value</td>
<td>0.17</td>
<td>0.57</td>
<td>0.89</td>
<td>0.025</td>
<td>0.18</td>
<td>0.93</td>
<td></td>
</tr>
<tr>
<td>% of total variation explained by route of infection</td>
<td>2</td>
<td>0.3</td>
<td>0.02</td>
<td>5</td>
<td>2</td>
<td>0.0</td>
<td></td>
</tr>
</tbody>
</table>
specifically addressing the first wave of asexual parasitemia and its importance in pathogenicity: Saul (1994), Simpson and others (2002), and Haydon and others (2002). Simpson and others present a nonlinear mixed effects model and apply it to 244 cases from the data set also used for the present paper. They obtain good fits and estimate the two-day parasite multiplication factor (PMF) to be approximately 8 on average, with significant variation among parasite strains as well as among patients (90% prediction interval = 5.5–12.3). (Note that this factor is not a rate so that PMF would be a more proper name for it than parasite multiplication rate.) They conclude that PMF, host susceptibility, and immuno-competence all contribute to the expansion of the parasite biomass, and thus to disease severity, in *P. falciparum* malaria. They reached conclusions that are qualitatively similar to some of ours, with a different model. However, their estimates of the two-day multiplication factor are low. A downward bias could be expected from an actual error in solving $Y = bY$; the correct solution is not $\log_{10} Y = a + bt$ but $\log Y = a + bt$. A further complicating factor is their questionable post-fitting exclusion of 128 of 245 cases because they fail to fit the model. The more widely accepted procedure is to reject the model if it does not fit the data.

Haydon and others offer the promising statement “Given the importance of the early dynamics of infection for disease severity, it is critical to understand what factors limit parasite numbers during this [first acute] phase.” However, *P. chabaudi* in mice is not a good model of *P. falciparum* in humans. The assumption that the only problem concerning the use of predator-prey models as within-host models of *P. falciparum* is to decide whether the parasite is prey or predator misses the point that *P. falciparum* in humans never shows the behavior of predator-prey models (indefinitely sustained undamped oscillations for the good ones and damped oscillations towards an indefinitely stable positive equilibrium for the bad ones, a point stressed in our review). Dose is probably not an important factor of severity, unless one uses doses that are orders of magnitude larger than any natural inoculation. The question of dose is, like in much of the literature, addressed as if nature could vary dose of inoculum without varying jointly diversity of inoculum, and that increasing diversity is probably more pathogenic than increasing dose.

The paper by Saul is most relevant for the work presented here. The promised publication of the full details of the model has apparently not materialized, but several published models clearly derive from this model, starting with Paget-McNicol and others, whose report we also studied in detail.
In the present context, the most interesting features of the model of Saul are 1) concentration on the first wave of asexual parasitemia; 2) important roles for both innate and adaptive IRs (under other names); 3) his innate IR is in essence the same as ours, in both models IEs are both trigger and target of a fast and labile innate IR; his intercalation of an effector (TNF) between trigger and target is redundant for the study of APVs (but see below); and 4) his adaptive IR like ours has one dimension and is delayed; in his report it becomes effective 20 days after the parasite density reaches 0.2/μL (1 × 10^6/5 L). Its effect is to halve the two-day multiplication factor each day.

Some major differences between the approach of Saul and ours are 1) no explicit comparison is made between data and simulations; 2) his model ignores variation among parasite strains and among patients with respect to either the two-day multiplication factor or any immune response parameters; and 3) in his model the only effect of an APV is to reduce the two-day multiplication factor. That direct effect leads to a reduction of the initial slope and of the parasite biomass, effects that we also observe with our model. The disagreement between Saul and our group regarding individual variation is made more explicit in the report by Paget-McNicol and others.18

The report by Saul is interesting beyond its relevance for the present work because his simulations of the effect of antitoxic vaccines (for which the explicit effector is not redundant) are thought-provoking and should be taken into account in any discussion of that topic. His paper remains a useful overview of many important aspects of the evaluation of malaria vaccines.

Our baseline model produces realistic simulations of the highly pathogenic first wave of *P. falciparum* asexual parasitemia in non-immune persons on the basis of plausible biologic assumptions. This may justify the use of the model’s behavior to assess the importance of some of the risk factors for severe malaria listed in reviews of the problem.19–23

Several of the reviews discussed consider that a high multiplication factor is a probable risk factor for severe malaria. We find variation in *m* among cases and a significant effect of the multiplication factor on the maximum density. Possible underlying host factors might include red blood cell polymorphisms, e.g., hemoglobin S, which is relatively common in Afro-Americans, but on which no information is available for the data set used here.
Greater efficacy against severe versus mild *P. falciparum* malaria has also been found in field evaluation of the pre-erythrocytic vaccine RTS.S, as well as of insecticide-treated bed nets. A comprehensive explanation of those phenomena might include some of our findings, in addition to pre-erythrocytic IRs and the implication of the probable association, under natural exposure, between the number of clones inoculated into an individual and their antigenic diversity.

Marsh listed “polymorphism in genes governing acute phase response” in his table of risk factors for severe malaria. We found a large variation among cases with respect to *Pc*, the parameter of the innate IR. A large variation of *Pc* was also to be expected from the large variation found by Jeffery and others in the malarial therapy patients, with respect to maximum density (more than two orders of magnitude) and to the density associated with first fever (more than four orders of magnitude). The innate IR is probably a broad-spectrum host-specific response, as suggested by our earlier finding, in malaria therapy reinoculation data, the density at which an individual controls a first malaria inoculation using *P. ovale* (presumably controlled by a hypothetical *Pc* (*P. ovale*)) is positively and strongly correlated (r = 0.76) with the (much higher) level at which the same individual controls a second malaria inoculation using *P. falciparum*. Our assumption that a weak innate IR increases the risk of high parasitemia is compatible with the high circulating levels of pro-inflammatory cytokines associated with severe malaria: a weak innate IR per parasite is likely to allow the production per host of a large parasite biomass with anatomically diffuse sequestration, which is likely to produce a high systemic level of pro-inflammatory cytokines. Several field studies showed a positive correlation between intrinsic strength of proinflammatory cytokine responses and rate of clearance of parasites and fever; some also showed a negative correlation between systemic level and intrinsic strength of the response. Saul uses his model to simulate the impact of antitoxic vaccination. His simulations show that if the dominant direct effect of such a vaccine is to reduce the production of proinflammatory cytokines per parasite, this leads to an increase in parasite biomass, which in turn compensates for the de-

**Figure 6.** Vaccine efficacy (VE) with respect to fever (dotted lines) and severe malaria (solid lines) as a function of immunogenicity level as defined in Table 6. **A,** three vaccine effects combined; **B,** only the two memory effects; **C,** the effect on delay on its own.
creased production of proinflammatory cytokines per parasitome. The use of a high parasite density as an indicator of severe malaria has been discussed earlier in the report (see Materials and Methods). We expect no objection to the use of the fever threshold as a good marker for the diagnosis of uncomplicated (or mild) clinical malaria. We use a host-specific fever threshold because that is what the data suggest. Conversely, we use a host-independent density threshold for severe malaria. We do not expect that threshold to be invariant, but we conjecture that its variability is much smaller than that of the fever threshold. However, it cannot be ethically studied in humans. It could be studied in rodent malaria using death as an endpoint (not in mice because mortality due to P. berghei is 100%) but in a natural rodent-parasite pair. A good choice would be the Thamnomys rutilans-P.chabaudi pair because it shows a much lower host mortality, as well as some other scientifically attractive similarities with the pair humans-P. falciparum.\footnote{31}

Moving from a baseline model into simulations of the expected effects of a hypothetical APV involves two successive decisions: the selection of what model parameters will be affected by that vaccine, and the magnitude of the effect. Although the second decision is arbitrarily covering a wide range of hypothetical vaccine efficacies, the first decision should be evaluated in terms of plausibility on biologic grounds.

We have explained (see Materials and Methods) why we believe the factors $f_{\Delta t}$, $f_{\Delta P}$, $f_{\Delta m}$ capture three capital features of the adaptive IR. First, given the relative scarcity of pertinent data concerning effectors (e.g. antibodies), we conjecture that modeling the effectors explicitly would probably be redundant mathematically. Second, the finding of a positive correlation between log $\Delta t$ and log $P_m$ ($r = 0.45, P < 0.0001$) (Table 5) is rather reassuring because the two corresponding features of adaptation of the IR (acceleration and strengthening) rest on the same biologic process. Third, we have made the factors $f_{\Delta t}$, $f_{\Delta P}$, $f_{\Delta m}$ invariant among hosts. If as is probably more plausible biologically, we had allowed the immuno-competence of an individual to respond to vaccination to vary in the same direction as his or her basic immuno-competence to respond to natural infection, then variation among cases of a vaccine’s protective efficacy would be even greater than the one we obtain. Fourth, if in addition to $\Delta t$, we let a vaccine also shorten $\Delta P$, as is biologically plausible, the protective efficacy of the vaccine would be increased.

We found that protective efficacy is strongly dependent on the individual. The variability in our simulations must be an underestimate of the true variance because of the truncation of the underlying data with respect to severity. At low and intermediate levels of immunogenicity, protective efficacy is greater against severe disease than against fever. This has implications for vaccine development plans. Since the sample size required to assess protective efficacy against fever is much smaller than that required for assessing protective efficacy against severe disease, such plans tend to require proof of protective efficacy against fever as a criterion to pursue the (expensive) further development of a vaccine-candidate. If our finding is reliable (and we conjecture that particular outcome to be rather robust), then an APV that has protective efficacy against severe disease might be discarded because of a failure to show protective efficacy against fever.

We emphasize the following general points. First, to simulate possible effects of hypothetical blood-stage vaccines, it could be seriously misleading to ignore well-documented individual variations in biologically important parameters. Second, our model of the first wave of asexual parasitemia offers a biologically defensible way to take into account some important components of such individual variation.

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