RESEARCH ARTICLE

Predicting the Duration of Chloroquine, Mefloquine, Halfantrine and Artesunate for Blood Schizonticidal Effect using Mathematical Models of Malaria with Immune Response

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Abstract

This paper focuses on the mathematical model of the death rate of malaria parasite due to chloroquine, mefloquine, halfantrine and artesunate together with the immune response in order to determine the treatment period. The basic knowledge of probabilities, pharmacology, microbiology, medicines, physics and chemistry are used to support this model and hypotheses. One of the hypotheses is the convective theory used for calculating and converting the flow rate of drug molecules with respect to malaria parasite into the death rate of malaria parasite. Other instruments are five probability factors, which the authors use to determine the efficiency of the drugs parasites. Applied this model of death rate with normal malaria model and take into account of the immune response, the treatment durations result in the ranges between 1 to 15 days and are compatible with the real clinical data.

Keywords: Malaria, death rate, probability

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การทำนายระยะเวลายาคลอโรควิน เมโฟลควีน ฮาโลแฟนทรีน และอาร์เทซูเนท สำหรับผลจากการฆ่าเชื้อมาลาเรียในเลือด โดยใช้แบบจำลองคณิตศาสตร์ของมาลาเรียที่มีระบบภูมิคุ้มกัน

พนิต เสือวรรณศรี, ณัฐพันธ์ กิติสิน

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บทคัดย่อ

งานวิจัยนี้ได้ศึกษาแบบจำลองอัตราการตายของเชื้อมาลาเรียเนื่องจากยาคลอโรควิน เมโฟลควีน ฮาโลแฟนทรีน และอาร์เทซูเนทที่มีระบบภูมิคุ้มกัน เพื่อหาระยะเวลาของการรักษาแบบจำลองและสมมติฐานในงานชิ้นนี้ใช้ความรู้พื้นฐานทางด้านความน่าจะเป็น เภสัชวิทยา จุลชีววิทยา แพทย์ ฟิสิกส์และเคมี สมมติฐานข้อหนึ่งคืออัตราการไหลของโมเลกุลของยาเทียบกับเชื้อมาลาเรียโดยจะใช้ทฤษฎีการพาในการสร้างตัวแปรนี้ขึ้นมา เครื่องมือที่เหลือจะใช้ค่าความน่าจะเป็น 5 ค่าในการหาปริมาณยาในการฆ่าเชื้อมาลาเรีย ผลที่ได้หลังจากรวมอัตราการตายนี้กับแบบจำลองของเชื้อมาลาเรียที่มีระบบภูมิคุ้มกัน คือ ระยะเวลาในการรักษาจะอยู่ในช่วง 1 ถึง 15 วัน ซึ่งสอดคล้องและตรงกับข้อมูลจริงของผู้ป่วย

คำสำคัญ: มาลาเรีย, อัตราการตาย, ความน่าจะเป็น

Introduction

Malaria is a dangerous infectious disease due to transmission of *Plasmodium* parasites by biting of female *Anopheles* mosquitoes.[1] In 2015, there were malaria dissemination in 91 countries and WHO estimated that there were 212 million cases of malaria worldwide (within a range of 148 to 304 million). There are 429,000 people, who are died from malaria (within a range of 235,000 to 639,000).[2] Since the treatment of patients with malarial infection needs the advance methods for laboratory investigation that rarely exists in those developing countries, mathematical models can help to solve some of this problem. For example, the laboratory investigation needs to record the parasite density every day during the treatment period in order to determine when the treatment should be terminated, while the mathematical model in this study predicts the day that the parasite density will be cleared from bloodstream with just one blood sampling. Hence, the objective of this paper is to predict the treatment duration for the patient infected by malaria with immune response by constructing the mathematical model of the death rate of malaria. The numerical results are shown to be comparable to the actual treatment duration.

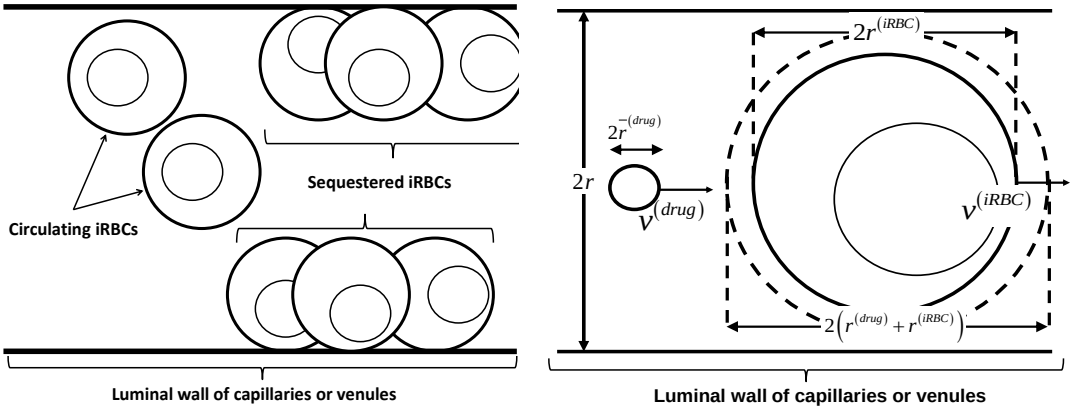
Before establishing the mathematical model, the pathogenesis of malaria and the mechanism of action of antimalarial drug are taken into account. The pathogenesis of malaria consists of two main cycles: sexual and asexual cycles. First, the sexual cycle needs to be reviewed. After a female *Anopheles* mosquito bites the patient infected malaria, a malarial agent called gametocyte is released from this patient and enters to the mosquito's stomach. When both male and female gametocytes (or microgametocytes and macrogametocytes, respectively) meet together, a zygote occurs by their reproductive process. Afterward, a zygote gradually develops into oocyst containing sporozoites. At the end of this sexual cycle, sporozoites are released from oocyst and enter into a human host bloodstream soon after this mosquito bites the human host. Second, the asexual cycle begins when the parasites are already in the human. After sporozoites come into a human host blood, they travel along blood circulation to invade liver cells. When sporozoites mature and convert into schizonts, liver cells are ruptured and release schizonts. Then, these schizonts will spread into blood system to infect healthy red blood cells (RBCs). They grow and develop into various stages within the infected red blood cells (iRBCs): early and late trophozoites, early and late schizonts, respectively. Finally, malaria parasites in the late schizonts, called merozoites, are mature, they are released into host blood and re-infect normal RBCs or normal liver cells again and again.[3]

The pathogenesis of malaria is different in each species. *P. falciparum* has two main phases in stage of intracellular parasites while *P. malariae, P. ovale, P. vivax* and *P. knowlesi* have only one phase. After extracellular parasites of *P. falciparum* infect normal RBCs, they can circulate in blood system freely. This situation and iRBCs in this phase are called circulating process and circulating iRBCs, respectively. If they survive from drugs or other factors, then they will fully grow and attach an intraluminal wall of capillaries and post-capillary venules of specific vital organs: brain, lungs, heart, kidneys and liver.[4,5] This situation and iRBCs in this phase are called sequestration process and sequestered iRBCs (Figure

1(a)). Thus, antimalarial drug has to spend time following the circulating iRBCs, but not the sequestered iRBCs which takes less time for the drug to kill.

During the entire lifetime of malaria parasites in the phase of iRBCs, they consume hemoglobin in the RBC's cytoplasm. However, they have to protect themselves from toxic and water-soluble hemes from digested hemoglobin by changing them into malarial pigment, known as non-toxic and water-insoluble hemozoin. Hence, the mechanism of antimalarial drugs killing parasites is to inhibit this conversion.[6,7] There are many antimalarial drugs involved with this inhibition such as chloroquine, artesunate, halofantrine and mefloquine. Remark that the route of mefloquine is oral. However, the authors will treat mefloquine via the intravenous route, which is also the same as chloroquine so that both of them can be correspondingly compared their results. Although the mechanism of artesunate is still unclear, the authors will treat artesunate as quinolines to generate numerical simulation.

The following assumptions and hypotheses were made: (i) the rate of collision between an iRBC and a drug molecule is determined by the relative velocity between both of them, based on the convective velocity in physics (see the subsection 1 in Material and Methods), (ii) probability of binding and capturing between both of them are calculated by five probability factors, based on the effective collision with the proper orientation of the collision theory in molecular chemistry, (iii) the drug is effective in the sense that for the right condition one drug molecule can annihilate or kill one iRBC, and (iv) the drug concentration in plasma is steady and time-independent since the rate of drug administration with normal saline solution via intravenous route is almost constant. This situation illustrates in Figure 1(b).



(a) (b)

Figure 1. (a) Circulating and sequestered iRBCs in *P. falciparum*. (b) The transportation of the drug molecule to an intracellular parasite within an iRBC by convection of bloodstream (see parameters and values in Table 1 and 2).

The assumption of equality between two volumes of prolate spheroid and sphere, represented as the structure of drug molecules, has to be concerned for calculating probability conveniently. To use spherical shape as drug structure, there is the principal conversion from prolate spheroid to sphere by calculating the geometric mean of the width and length of prolate spheroid, based on the equality of two volumes of prolate spheroid and sphere.[8]

First, define $r_w^{(drug)}$ and $r_l^{(drug)}$ to be the half width and half length of the structure of drug molecule, considered as prolate spheroid. Then, the volume of prolate spheroid, defined as $V^{(prolate)}$, is $V^{(prolate)} = \frac{4}{3}\pi\left(r_w^{(drug)}\right)^2 r_l^{(drug)}$. Similarly, the volume of sphere, defined as $V^{(sphere)}$, is $V^{(sphere)} = \frac{4}{3}\pi\left(r^{(drug)}\right)^3$. Thus, from the assumption of equality $V^{(prolate)} = V^{(sphere)}$, the average radius of drug molecule is called Stokes radius[8,9] and denoted by $r^{(stoke)} = \bar{r}^{(drug)} = \left(\left(r_w^{(drug)}\right)^2 r_l^{(drug)}\right)^{1/3}$.

Materials and Methods

In this section, the authors try to calculate the relative velocity when the drug molecules follow malaria parasites along blood circulation. The scope of this study is to process the values of various parameters under proper assumptions. For example, the authors treat the mechanism of drug action as the collision rate between a drug molecule and a parasite, which is derived from the relative velocity of the convection of bloodstream without involving the Brownian movement. Furthermore, the death rate of malaria parasite is derived from five probability factors and the flow rate of drug molecules by using this relative velocity. Remark that only the active drug molecules using with the relative velocity will be used to formulate the death rate of malarial parasite, which will be subtracted from the mathematical models of malaria. The reason is that after binding parasites, the active drug molecules are ready to kill parasites and afterward will be converted into an inactive form, which will be excreted out of host body. Therefore, pharmacokinetics and pharmaco-dynamics, which occur after binding between drug molecules and parasites, are not relevant to the mathematical models. However, if some drugs have complex mechanism of pharmacokinetics such as inhibiting of protein synthesis by binding at DNA or RNA of parasite, the probability of collision would be affected. First, the relative velocity between a drug molecule and an iRBC is used to determine the specific blood volume that contains the amount of particular drug. Second, the amount of this particular drug that can effectively kill a parasite has to be calculated by taking into account of the five probability factors of binding between a drug molecule and an iRBC. Five probability factors are formulated as follows (i) position probability factor determined by binding chance in the sense of position alignment between a drug molecule and an iRBC, (ii) binding probability factor of binding between a drug molecule and an intracellular parasite, but not the cytoplasmic iRBC, (iii) capture probability factor or chance of drug traveling that does not deviate from an iRBC, (iv) orientation probability factor or the probability of attachment between an active site of drug and iron molecules in heme, (v) population probability factor

is the ratio of the drug molecule to only one iRBC, but not the normal RBC or immune effectors. Then, the total probability factor can be approximately estimated by the multiplying all of them and will result in the effective of the drug molecules. Before using the total probability, drug mass is needed to be computed. First, the specific blood volume, derived from the relative velocity, has to be changed into drug mass by multiplying drug concentration in plasma and the parameters of chemistry, such as Avogadro's number and molecular mass, which are used to convert the amount drug mass into the number of drug molecule. Furthermore, the total of drug molecules in human host occurs in all capillaries and venules of human host. Remark that this paper will focus only on schizonticides with chloroquine, mefloquine, halofantrine and artesunate. Finally, after converting the flow rate of drug molecules into the death rate of malaria parasite, this death rate is subtracted from the dynamic population of iRBCs of malaria model in normal state and then simulates numerical results to predict treatment duration in cases of *P.* non-*falciparum* and *P. falciparum*, shown in Figure 2 and 3, respectively. Thus, all processes in these subsections are explained as follows.

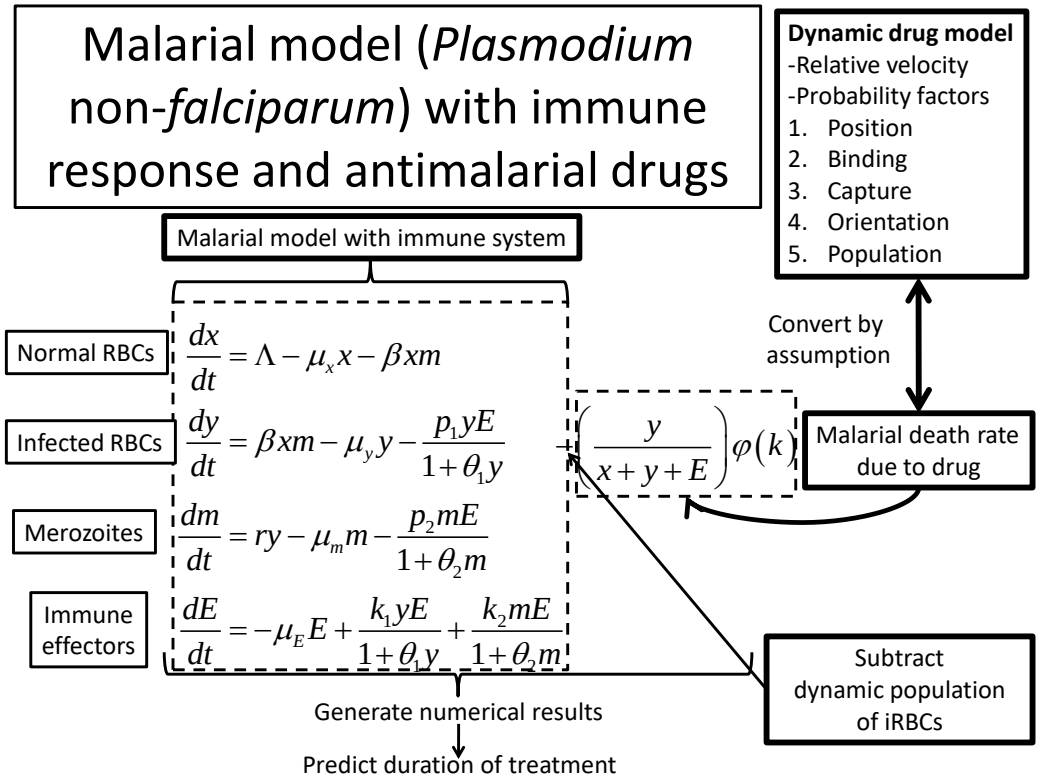


Figure 2. Algorithm for modeling the death rate of the malaria parasites in case of *P.* non-*falciparum* due to chloroquine, mefloquine, halofantrine and artesunate with immune response. Five probability factors and the relative velocity between a drug molecule and a parasite are used in order to predict the treatment duration.

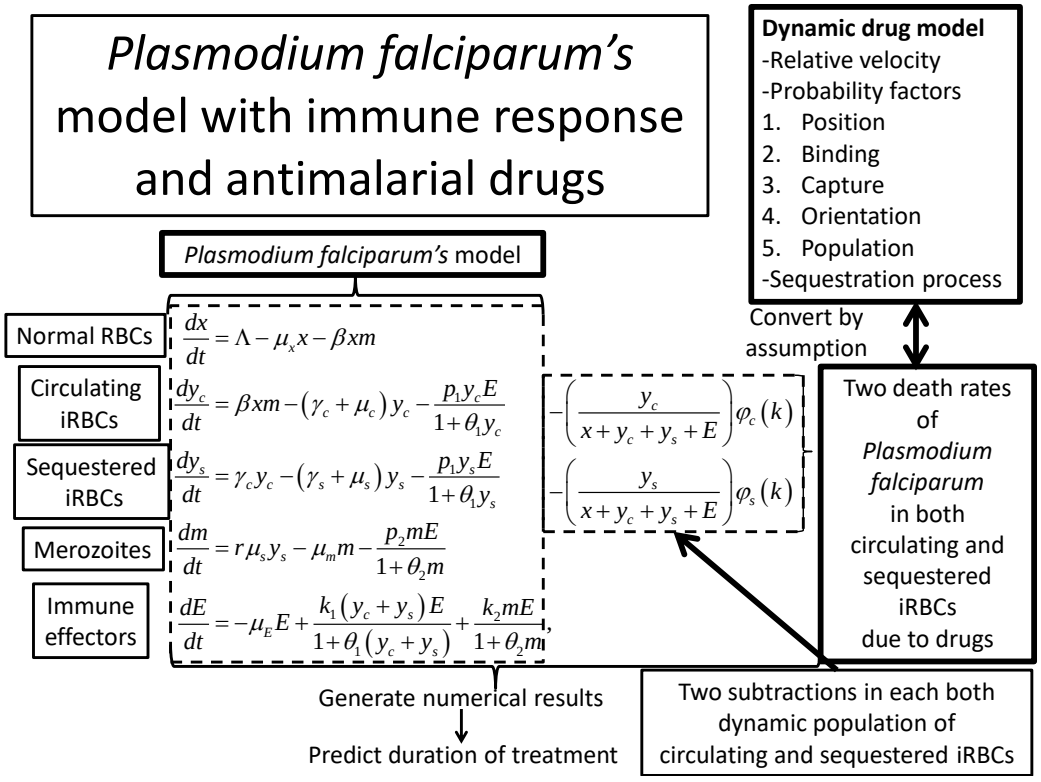


Figure 3. Algorithm for modeling the death rate of the malaria parasites in case of *P. falciparum* due to chloroquine, mefloquine, halofantrine and artesunate with the immune response. Five probability factors and the relative velocity between a drug molecule and a parasite are used in order to predict the treatment duration

1. Relative velocity between a drug molecule and an infected red blood cell

According to the fundamental of the relative velocity in physics, since a drug molecule is smaller than an iRBC, the velocity of drug molecule is faster than an iRBC, i.e. $v^{(drug)} > v^{(iRBC)}$, where $v^{(drug)}$ and $v^{(iRBC)}$ are two velocities of a drug molecule and an iRBC, respectively. Denote $v^{(relative)} = v^{(drug)} - v^{(iRBC)}$ as the relative velocity of drug with respect to the velocity of iRBC. Since $v^{(iRBC)}$ and $v^{(drug)}$ can be determined by multiplying between each local lag coefficient of them (denoted by $G^{(drug)}$ and $G^{(iRBC)}$ as two local lag coefficients of a drug and an iRBC, respectively) and the flow rate of blood (denoted by $v^{(blood)}$). Then, $v^{(iRBC)} = G^{(iRBC)} v^{(blood)}$ and $v^{(drug)} = G^{(drug)} v^{(blood)}$, where $G^{(drug)}$ and $G^{(iRBC)}$ are two local lag coefficients of a drug and an iRBC, respectively (see also Figure 1(b)).[10]

2. Position probability factor

Our situation in this paper that drug molecules move along blood vessels to bind with iRBCs is comparable to the straight-forward traveling of ships to collide with shoals or obstacles in water channels. The collision between drug molecules and iRBCs will occur if both of them are aligned in appropriate or correct position. Hence, the probabilistic model of ship grounding[11] is used to find the effective drug molecules which are potential to kill iRBCs. Nevertheless, since our situation is concerned in two-dimension (2D), the probabilistic model[11] is needed to be generalized from 1D to 2D before using in our model. Thus, the modified probability is defined to be the position probability factor in this paper by

$$p^{(position)} = \frac{\pi\left(r^{(iRBC)} + \bar{r}^{(drug)}\right)^2}{\pi r^2} = \left(\frac{r^{(iRBC)} + \bar{r}^{(drug)}}{r^{(vessel)}}\right)^2,$$

where $r^{(iRBC)}$ and $r^{(vessel)}$ are two radii of an iRBC and a human blood vessel in cross-sectional view, respectively.

3. Binding probability factor

The total volume of iRBCs, denoted by $V^{(iRBC)}$, contains two volumes. One of them is the volume of an intracellular parasite, denoted by $V^{(parasite)}$. The drug molecules must bind the volume $V^{(parasite)}$ to kill this parasite. The chance of binding between drug molecules and a parasite within iRBCs by using the modified probability,[12] called the binding probability factor and defined as

$$p^{(binding)} = \frac{V^{(parasite)}}{V^{(iRBC)}}.$$

Note that our binding probability factor is the parasite volume fraction.[13]

4. Capture probability factor

Before the events of position and binding between a drug molecule and an iRBC, a drug molecule can deviate from the iRBC during traveling. Based on physics, the longer distance between them, the less probability of binding together between them. Therefore, the capture probability[14] will be used to determine this probability in this situation, which is defined as

$$p^{(capture)} = \frac{r^{(iRBC)}}{r^{(iRBC)} + l},$$

where l is the length of blood vessel of human host.

5. Orientation probability factor

In view of chemistry, a product of chemical reaction occurs when each binding site of the substrates bind together with correct orientation. Thus, this probability is defined as the orientation probability factor,[15] which is the ratio of the binding area to the total surface area of a drug molecule and denoted by

$$p^{(orientation)} = \frac{\pi z^2}{4} / \left\{ 2\pi \left(r_w^{(drug)} \right)^2 \left(1 + \frac{r_w^{(drug)}}{e r_l^{(drug)}} \sin^{-1} e \right) \right\}.$$

Remark that the binding area of a drug molecule ($\frac{\pi z^2}{4}$) is circular with the diameter *z*. Since our hypothesis is that the structure of drug molecule is a prolate spheroid, the total surface area of a drug molecule is

$$2\pi \left(r_w^{(drug)} \right)^2 \left(1 + \frac{r_w^{(drug)}}{e r_l^{(drug)}} \sin^{-1} e \right),$$

where e is the eccentricity of the ellipse.

6. Population probability factor

Since a drug molecule can attach RBCs, iRBCs and immune effectors, drug molecules have to avoid normal RBCs and immune effectors. Thus, in case of *P.* non-*falciparum*, the probability in this situation, defined as the population probability factor, is

$$p^{(population)} = \frac{y(t)}{x(t) + y(t) + E(t)}.$$

In both cases of circulating and sequestration processes, there are two population probability factors of *P. falciparum*, which are

$$p_c^{(population)} = \frac{y_c(t)}{x(t) + y_c(t) + y_s(t) + E(t)} \text{ and}$$

$$p_s^{(population)} = \frac{y_s(t)}{x(t) + y_c(t) + y_s(t) + E(t)}, \text{ respectively.}$$

7. Total probability

To find the total probability of five probability factors, the authors will assume that they are independent in each other. Then, the total probability can be derived by multiplying them together, which is

$$p^{(total)} = p^{(position)} \times p^{(binding)} \times p^{(capture)} \times p^{(orientation)} \times p^{(population)}.$$

8. Mathematical model of malaria

In 2011, Yilong *et al*.[1] had constructed the mathematical model of the within-host dynamics of malaria infection with immune response as shown in Figure 4(a). Since this paper studies both *P. falciparum* and *P.* non-*falciparum*, the authors modify the following two models, which are derived from Yilong *et al*.[1] with Bichara *et al*.[16] and then adapt to the *P. falciparum*'s model with immune response as shown in Figure 4(a) and 4(b). Remark that the immune effectors, defined as *E*, represent the immune effect, which concerns only the white blood cells and does not include the RBCs (see Yilong *et al*.[1] in details). Thus, these two models are used together with our death rates to generate numerical results to predict treatment duration.

$$\frac{dx}{dt} = \Lambda - \mu_x x - \beta x m,$$
$$\frac{dy}{dt} = \beta x m - \mu_y y - \frac{p_1 y E}{1+\theta_1 y},$$
$$\frac{dm}{dt} = r y - \mu_m m - \frac{p_2 m E}{1+\theta_2 m},$$
$$\frac{dE}{dt} = -\mu_E E + \frac{k_1 y E}{1+\theta_1 y} + \frac{k_2 m E}{1+\theta_2 m},$$

(a)

$$\frac{dx}{dt} = \Lambda - \mu_x x - \beta x m,$$
$$\frac{dy_c}{dt} = \beta x m - \left(\gamma_c + \mu_c\right) y_c - \frac{p_1 y_c E}{1+\theta_1 y_c},$$
$$\frac{dy_s}{dt} = \gamma_c y_c - \left(\gamma_s + \mu_s\right) y_s - \frac{p_1 y_s E}{1+\theta_1 y_s},$$
$$\frac{dm}{dt} = r\gamma_s y_s - \mu_m m - \frac{p_2 m E}{1+\theta_2 m},$$
$$\frac{dE}{dt} = -\mu_E E + \frac{k_1\left(y_c + y_s\right) E}{1+\theta_1\left(y_c + y_s\right)} + \frac{k_2 m E}{1+\theta_2 m},$$

(b)

Figure 4. The mathematical model of within-host malaria parasite with immune response[1] in cases of (a) *P.* non-*falciparum* and (b) *P. falciparum* from Yilong *et al*.[1] with Bichara *et al*.[16] with our modification, respectively. All variables, parameters and values can be seen in Table 1 and 2.

9. Dynamic drug model

The steps for constructing the death rate of malaria parasite are the following (i) find the number of drug molecules binding parasites per unit time (ii) assume that the effective drug killing parasites satisfied the correct condition in the sense that one iRBC can be annihilated per one drug molecule. First, in the scope of one blood vessel, if the length that blood volume sweeps with time t is $v^{(relative)}t$ with its the cross-sectional area of a vessel is $\pi\left(r^{(vessel)}\right)^2$, then the specific blood volume in one vessel is $\pi\left(r^{(vessel)}\right)^2 v^{(relative)}t$. Second, change this specific blood volume into drug mass by multiplying plasma drug concentration $C(t)$ and convert the amount of this drug mass into the number of drug molecules again by multiplying Avocado's

number *A* and dividing by the molecular mass of this drug *M*. Afterward, the authors interpolate this flow rate of drug molecules into all blood vessels of the whole human body. Thus, the flow rate of drug molecules, denoted by $N^{(drug)}$, is $N^{(drug)} = \pi\left(r^{(vessel)}\right)^2 C(t)Av^{(relative)}t/M$. Since there are a few effects with hemodynamics of human host, two probabilistic chances of the binding of drug molecules and parasites in view of blood circulation have to be taken into account as follows: (a) find the period of time that drug molecules can follow parasites by determining the ratio of time in blood vessels, called transit time and denoted by τ, and the circulatory time, denoted by ρ. (b) use the ratio of only unbounded drug molecules to all drug molecules, i.e. the free fraction of drug, denoted by α. (c) multiply both of them with the total probability factor as before. (d) divide by the total blood volume of patient, denoted by $V^{(blood)}$. Finally, change $N^{(drug)}$ into the rate by dividing by *t*. Then, the death rate of the malaria parasites is defined as

$$\Psi(t) = \frac{\pi\alpha N^{(vessel)}\tau\left(r^{(vessel)}\right)^2 C(t)Ap^{(total)}v^{(relative)}}{\rho MV^{(blood)}}$$

(see also Figure 5).

Since this death rate is considered in both capillaries and venules, the two death rates in cases of both capillaries and venules are needed to be constructed separately. Therefore, the total death rate of the malaria parasites is defined as $\Psi^{(total)}(t) = \Psi^{(capillary)}(t) + \Psi^{(venule)}(t)$.

By one of the assumptions satisfied in our situation, the antimalarial treatment by intravenous route of drug administration maintains the constant level of drug concentration in plasma. First, if drug concentration does not depend on time, then the drug concentration constant k will be used instead of $C(t)$. Second, the variables of the population of $x(t)$, $y(t)$ and $E(t)$ in population probability factor have to be extracted from the death rate before simulating the results in order to determine the dynamic of malaria parasites conveniently. Hence, define

$$\varphi(k) = \Psi^{(total)}(k)/\,p^{(population)} = \left(\frac{x+y+E}{y}\right)\Psi^{(total)}(k),$$

where the death rate of malaria parasite in other form is given by

$$\Psi^{(total)}(k) = \left(\frac{y}{x+y+E}\right)\varphi(k).$$

Thus, when iRBCs are totally killed, i.e. $y \to 0$, the death rate tends to zero, i.e. $\Psi^{(total)}(k) \to 0$. This statement conforms the real situation. Hence, *P.* non-*falciparum*'s model with antimalarial drug and immune response is

Relative velocity	$v^{(relative)}$	
	↓	$\times\pi(r^{(vessel)})^2$
Blood flow with drug in one vessel	$\pi(r^{(vessel)})^2 v^{(relative)}$	
	↓	$\times N^{(vessel)}$
Blood flow with drug in all vessels	$\pi(r^{(vessel)})^2 N^{(vessel)} v^{(relative)}$	
	↓	$\times(\tau/\rho)$
Blood flow with drug in capillaries/venules	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} v^{(relative)}/\rho$	
	↓	$\times C(t)$
Flow rate of time-dependent drug concentration	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} C(t) v^{(relative)}/\rho$	
	↓	$C(t) \longrightarrow k$
Flow rate of constant drug concentration	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} k v^{(relative)}/\rho$	
	↓	$\times(A/M)$
Flow rate of drug molecule	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} A k v^{(relative)}/\rho M$	
	↓	$\times \alpha$
Flow rate of drug molecule in free form	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A k v^{(relative)}/\rho M$	
	↓	$\times p^{(total)}$
Effective flow rate of drug molecule	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M$	
	↓	convert drug to parasite
Death rate of malaria parasite (*cell/day*)	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M$	
	↓	$\div V^{(blood)}$
Death rate of malaria parasite density (*cells/(μl.day)*)	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M V^{(blood)}$	

Figure 5. This diagram shows formulation of the death rate of malaria parasite density, where $p^{(population)} = \frac{y}{x+y+E}$ in $p^{(total)}$.

$$\frac{dx}{dt} = \Lambda - \mu_x x - \beta xm,$$
$$\frac{dy}{dt} = \beta xm - \mu_y y - \frac{p_1 yE}{1+\theta_1 y} - \left(\frac{y}{x+y+E}\right)\varphi(k),$$
$$\frac{dm}{dt} = ry - \mu_m m - \frac{p_2 mE}{1+\theta_2 m},$$
$$\frac{dE}{dt} = -\mu_E E + \frac{k_1 yE}{1+\theta_1 y} + \frac{k_2 mE}{1+\theta_2 m},$$

In case of *P. falciparum*, there are two phases of iRBCs in *P. falciparum*. The authors will illustrate the formulation of death rate of malaria parasite in case of *P. falciparum* in circulating process and *P.* non-*falciparum* at once because both of them have almost the same pathophysiology. The algorithm of modeling this death rate in both cases can be concluded as follows: (i) find the quantity of drug molecules attaching circulating iRBCs in case of *P. falciparum* or total iRBCs in case of *P.* non-*falciparum* per unit time, (ii) assume that drug molecules in the sense for the correct condition that one iRBC can be annihilated by only one drug molecule (see also Figure 6).

For the population probability factor in case of *P. falciparum*, since the circulating and sequestered processes in the phase of intracellular parasites of *P. falciparum* are different in the sense of pathophysiology, the formulation of death rate of malaria parasite is separated into two parts. One death rate in the circulating process is almost the same as the case of *P.* non-*falciparum*, i.e. two population probability factors of them are different as follows:

$$p_c^{(population)} = \frac{y_c}{x + y_c + y_s + E},$$
$$p_s^{(population)} = \frac{y_s}{x + y_c + y_s + E} \quad \text{and}$$
$$p^{(population)} = \frac{y}{x + y + E}$$

are three population probability factors in case of circulating and sequestered processes in *P. falciparum*, and *P.* non-*falciparum*, respectively.

The first difference between the circulating and sequestered processes is that sequestered iRBCs attach with the luminal wall of blood vessels, i.e. $v_s^{(iRBC)} = 0$. Thus, the relative velocity between drug molecules and sequestered iRBCs is $v_s^{(relative)} = v_s^{(drug)}$.

The second difference is that sequestration process occurs only in specific vital organs of human host, which are brain, heart, left and right lungs, liver and left and right kidneys.[4,5] Therefore, the death rate of malaria parasite in sequestration process is determined only in blood vessels of specific vital organs.

Relative velocity	$v^{(relative)}$	
	↓	$\times\pi(r^{(vessel)})^2$
Blood flow with drug in one vessel	$\pi(r^{(vessel)})^2 v^{(relative)}$	
	↓	$\times N^{(vessel)}$
Blood flow with drug in all vessels	$\pi(r^{(vessel)})^2 N^{(vessel)} v^{(relative)}$	
	↓	$\times(\tau/\rho)$
Blood flow with drug in capillaries/venules	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} v^{(relative)}/\rho$	
	↓	$\times k$
Flow rate of drug mass	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} k v^{(relative)}/\rho$	
	↓	$\times(A/M)$
Flow rate of drug molecule	$\pi\tau(r^{(vessel)})^2 N^{(vessel)} A k v^{(relative)}/\rho M$	
	↓	$\times\sigma$
Flow rate of drug molecule in free form	$\pi\sigma\tau(r^{(vessel)})^2 N^{(vessel)} A k v^{(relative)}/\rho M$	
	↓	$\times p^{(total)}$
Effective flow rate of drug molecule	$\pi\sigma\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M$	
	↓	convert drug to parasite
Death rate of malaria parasite (*cell/day*)	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M$	
	↓	$\div V^{(blood)}$
Death rate of malaria parasite density (*cells/(μl.day)*)	$\pi\alpha\tau(r^{(vessel)})^2 N^{(vessel)} A p^{(total)} k v^{(relative)}/\rho M V^{(blood)}$	

Figure 6. This diagram shows the first step formulation of the death rate of circulating iRBCs, where $p_c^{(population)} = \frac{y_c}{x + y_c + y_s + E}$ in $p_c^{(total)}$.

Define $N_{(vital)}^{(capillary)}$ and $N_{(vital)}^{(venule)}$ as the number of total capillaries and total venules of all specific vital organs, respectively. To determine both $N_{(vital)}^{(capillary)}$ and $N_{(vital)}^{(venule)}$, the authors assume that the number of blood vessels of an organ is directly proportional to this organ's weight. Thus, if $W^{(vital)}$ and $W^{(body)}$ are defined as the two weights of all specific vital organs and whole body, respectively, then $W^{(vital)}/W^{(body)}$, $N_{(vital)}^{(capillary)}/N^{(capillary)}$ and $N_{(vital)}^{(venule)}/N^{(venule)}$ are equal, i.e.

$$\frac{W^{(vital)}}{W^{(body)}} = \frac{N_{(vital)}^{(capillary)}}{N^{(capillary)}} = \frac{N_{(vital)}^{(venule)}}{N^{(venule)}}.$$

Since the percentage of kidneys, lungs, liver, heart and brain to whole body weight 0.49%, 1.72%, 2.09%, 0.51% and 1.98%, respectively[17], the total percentage of all specific vital organs, defined as λ, is

$$\lambda = \frac{W^{(vital)}}{W^{(body)}} = 1.98 + 0.51 + 2.09 + 1.72 + 0.49\% = 6.79\%$$

Incorporating λ with $\Psi_s^{(total)}(t)$, then two death rates of *P. falciparum* in circulating and sequestration process are

$$\Psi_c(t) = \frac{\pi\alpha N^{(vessel)}\tau r^2 C(t) Ap^{(total)}}{\rho M}\left(G^{(drug)} - G^{(iRBC)}\right)v^{(blood)},$$
$$\Psi_s(t) = \frac{\pi\alpha N^{(vessel)}\tau r^2 C(t) Ap^{(total)}}{\rho M}\lambda G^{(drug)}v^{(blood)}.$$

Both death rates are considered in only capillaries and venules, then

$$\Psi_c^{(total)}(k) = \Psi_c^{(capillary)}(k) + \Psi_c^{(venule)}(k)$$
$$\Psi_s^{(total)}(k) = \Psi_s^{(capillary)}(k) + \Psi_s^{(venule)}(k)$$

Before coupling these two death rates in the mathematical model of *P. falciparum*, both of them have to be formulated in terms of the standard variables such that x, y_c, y_s and E in the dynamic system as the following steps: (i) since both two population probability factors in cases of circulating and sequestered iRBCs are

$$p_c^{(population)} = \frac{y_c}{x + y_c + y_s + E} \text{ and } p_s^{(population)} = \frac{y_s}{x + y_c + y_s + E},$$

(ii) define

$$\varphi_c(k) = \Psi_c^{(total)}(k) / p_c^{(population)} = \left(\frac{x + y_c + y_s + E}{y_c}\right)\Psi_c^{(total)}(k),$$

$$\varphi_s(k) = \Psi_s^{(total)}(k)(k) / p_s^{(population)} = \left(\frac{x + y_c + y_s + E}{y_s}\right)\Psi_s^{(total)}(k),$$

(iii) subtract both $\Psi_c^{(total)}(k) = \left(\frac{y_c}{x+y_c+y_s+E}\right)\varphi_c(k)$ and $\Psi_s^{(total)}(k) = \left(\frac{y_s}{x+y_c+y_s+E}\right)\varphi_s(k)$ from the dynamic variables y_c and y_s in the mathematical model of *P. falciparum.* Thus, the mathematical model of *P. falciparum* with antimalarial drugs and immune response is

$$\frac{dx}{dt} = \Lambda - \mu_x x - \beta x m,$$

$$\frac{dy_c}{dt} = \beta x m - (\gamma_c + \mu_c) y_c - \frac{p_1 y_c E}{1 + \theta_1 y_c} - \left(\frac{y_c}{x + y_c + y_s + E}\right)\varphi_c(k),$$

$$\frac{dy_s}{dt} = \gamma_c y_c - (\gamma_s + \mu_s) y_s - \frac{p_1 y_s E}{1 + \theta_1 y_s} - \left(\frac{y_s}{x + y_c + y_s + E}\right)\varphi_s(k),$$

$$\frac{dm}{dt} = r\mu_s y_s - \mu_m m - \frac{p_2 m E}{1 + \theta_2 m},$$

$$\frac{dE}{dt} = -\mu_E E + \frac{k_1 (y_c + y_s) E}{1 + \theta_1 (y_c + y_s)} + \frac{k_2 m E}{1 + \theta_2 m},$$

where all parameters and values are in Table 1 and 2.

Table 1. Parameters for numerical simulations in case of malarial infection.

Symbols	Value	Variables	Ref.
$G^{(drug)}$	1	lag coefficient of a drug in capillary and venule	10
$G^{(iRBC)}$	0.6,0.9	lag coefficient of an iRBC in capillary and venule, respectively.	10
x(0)	5×10^6	initial normal density of normal RBCs (*cells/*μ*l*)	1
Λ	4.15×10^4	production rate of RBCs (*cells/*μ*l/day*)	1
$r^{(RBC)}$	3.9	radius of RBC (μm)	13
$r^{(iRBC)}$	3.9	radius of iRBC (μm)	13
$V^{(iRBC)}$	86-116	volume of iRBC (μm^3)	13
$\delta^{(iRBC)}$	0.03-0.8	parasite volume fraction	13

Table 1 -- Continued. Parameters for numerical simulations in case of malarial infection.

Symbols	Value	Variables	Ref.
g	12	product rate of merozoites (/*day*)	1
β	2×10^{-9}	infective rate (μl/*cell.day*)	1
μ_x	8.3×10^{-3}	decay rate of RBCs (/*day*)	1
μ_y	1	decay rate of iRBCs (/*day*)	1
μ_c	0.42	decay rate of circulating iRBCs (/*day*)	16
μ_s	0.08	decay rate of sequestered iRBCs (/*day*)	16
γ_c	1.03	transition rate from circulating to sequestered iRBCs (/*day*)	16
γ_s	0.74	transition rate from sequestered iRBCs to merozoites (/*day*)	16
p_1	10^{-8}	removal rate of all iRBCs by immune system	1
p_2	10^{-8}	removal rate of merozoites by immune system	1
k_1	2.5×10^{-5}	proliferation rate of immune effectors by all iRBCs	1
k_2	4.69×10^{-5}	proliferation rate of immune effectors by merozoites	1
θ_1	5×10^{-4}	$1/\theta_1$half saturation constant for y_c(t) and y_s(t)	1
θ_2	6.67×10^{-4}	$1/\theta_2$half saturation constant for m(t)	1
μ_m	48	decay rate of merozoites (/*day*)	1
$m(0)$	10^4	initial density of merozoites (*cells*/μl)	1
$E(0)$	10^{-4}	initial density of immune effectors (*cells*/μl)	1
ρ	60	blood circulatory time (*s*)	18
$r^{(capillary)}$	3	radius of capillary (μm)	19
$r^{(venule)}$	10	radius of venule(μm)	19
τ	1	transit time (*s*) of capillary and venule	19
l	200,50-500	Length (μm) of capillary and venule, respectively.	20
$v^{(blood)}$	0.3,4	blood velocity (*mm*/*s*) in capillary and venule, respectively.	19,21
N	10^9,10^7	number of capillaries and venule, respectively.	22
$V^{(blood)}$	5	whole blood volume (liters)	23
$X(t_0)$	10^5	parasite density in blood a tt_0	24,25
A	6.02×10^{23}	Avogadro's number	26
	40-50	Ratio of drug efflux in case of resistant parasite to susceptible parasite	27

Table 2. Parameters of chloroquine, mefloquine, halofantrine and artesunate (see all parameters in Table 1).

Notations	r_l(*nm*)	r_w(*nm*)	*M*	α	*k* (*ng/ml*)	Ref.
Chloroquine	0.7856	0.35525	319.8721	0.45	120	28
Mefloquine	0.6333	0.33765	378.3122	0.02	500-638	29
Halofantrine	0.86655	0.427	500.4237	0.02	1000	29,30
Artesunate	0.7619	0.37065	384.4208	0.38	110-310	31,32

Results and Discussion

This section describes the results of the numerical simulation from using artesunate, chloroquine, mefloquine and halofantrine. They represent the antimalarial drugs for three cases: single drug treatment or monotherapy, drug resistance and artemisinin-based combination therapy or ACT.

In case of single drug treatment, the treatment durations for clearing parasite of all chloroquine, mefloquine, halofantrine and artesunate in case of *P.* non-*falciparum* are the same pattern (straight line in log-linear axis). These patterns and treatment durations in Figure 7(a), 7(b), 7(c) and 7(d) are also similar to Figure 1 and 4 in White[25] Furthermore, in case of *P. falciparum*, the numerical results from artesunate, chloroquine, mefloquine and halofantrine in Figure 8(a), 8(b), 8(c) and 8(d), respectively, also show that their patterns are also straight line in log-linear axis as well.

In case of drug resistance, *Plasmodium sp.* occurs only for some certain drugs. Chloroquine-resistant *P. vivax* are found in Papua New Guinea, Indonesia, Burma (Myanmar), India, and Central and South America but not *P. ovale*, *P. malariae* and *P. knowlesi*.[3] *P. falciparum* also resists other non-quinoline groups, such as mefloquine and halofantrine.[33] Consequently, in order to obtain our numerical results, the only drug efflux will be considered for our mechanism of drug resistance (see Sinha *et al*.[34] in details), which can be formulated in mathematical formula. Thus, an assumption that all *Plasmodium sp.* resist all drugs except artemisinin is added in this study for numerical analysis. In the process of calculation, the efficacy of drug resistance is defined as the ratio of drug efflux in case of resistant parasite to susceptible parasite, denoted by ε (see this value in Table 1), and applied by dividing with the death rate of malaria parasite. Thus, the death rates of malaria parasite with drug resistance are $\left(\frac{y_c}{x+y_c+y_s+E}\right)\frac{\varphi_c(k)}{\varepsilon}$, $\left(\frac{y_s}{x+y_c+y_s+E}\right)\frac{\varphi_s(k)}{\varepsilon}$, and $\left(\frac{y}{x+y+E}\right)\frac{\varphi(k)}{\varepsilon}$ in cases of *P. falciparum* with circulating and sequestration process, and other *Plasmodium sp.*, respectively (see also Table 3). According to the simulation, those patterns of results from chloroquine, mefloquine and halofantrine

are unchanged but their durations are longer than the results without drug resistance, as shown in Figure 9(a), 9(c) and 9(e) in case of *P.* non-*falciparum*, and Figure 10(a), 10(c) and 10(e) in case of *P. falciparum*, respectively. Thus, this parameter of resistance conforms with the real patient's data that it can affect treatment duration.

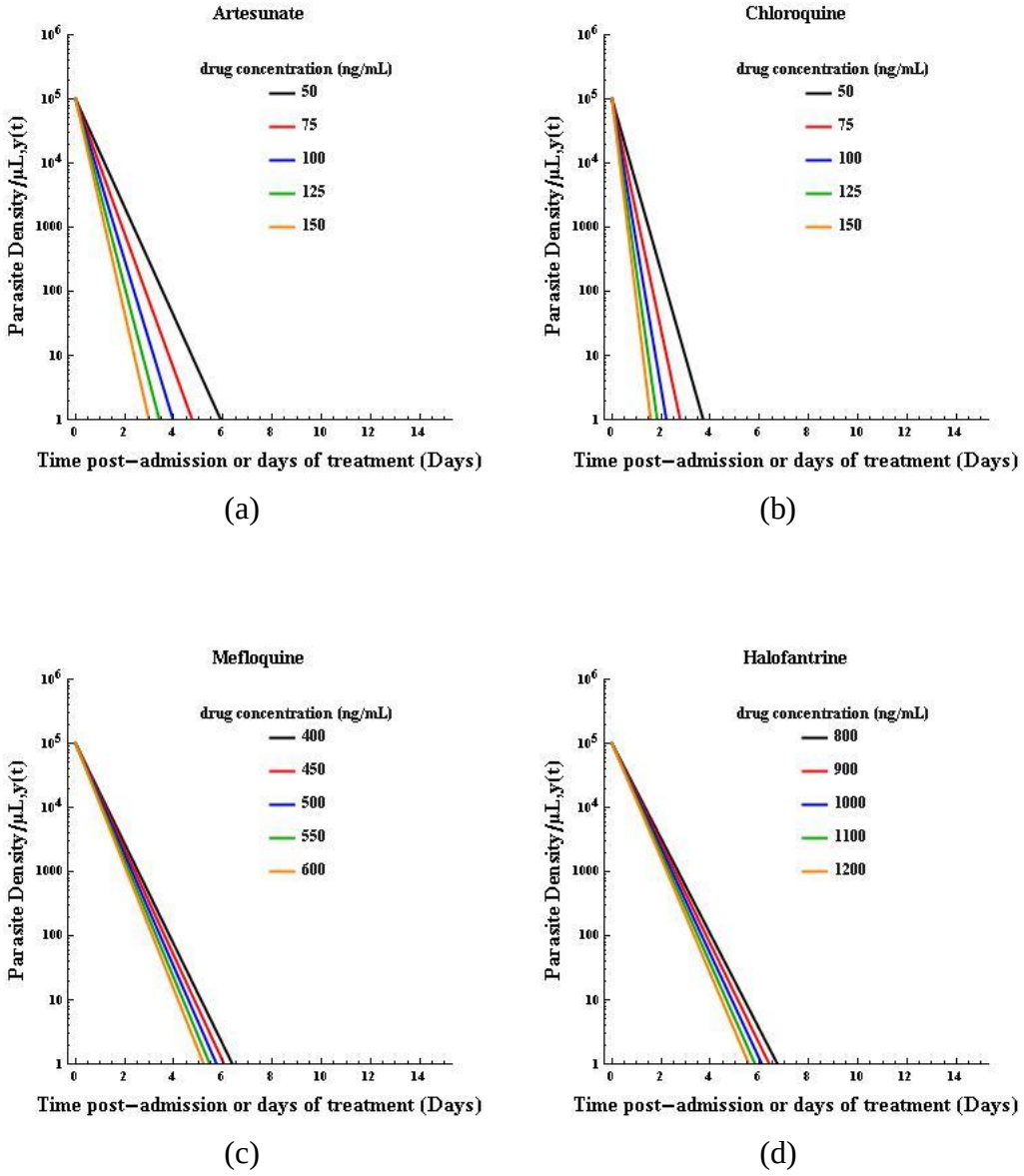


Figure 7. Comparison between real patient's data of Figure 1 and 4 in White[25] and our numerical results from (a) artesunate, (b) chloroquine, (c) mefloquine and (d) halofantrine in case of *P.* non-*falciparum*.

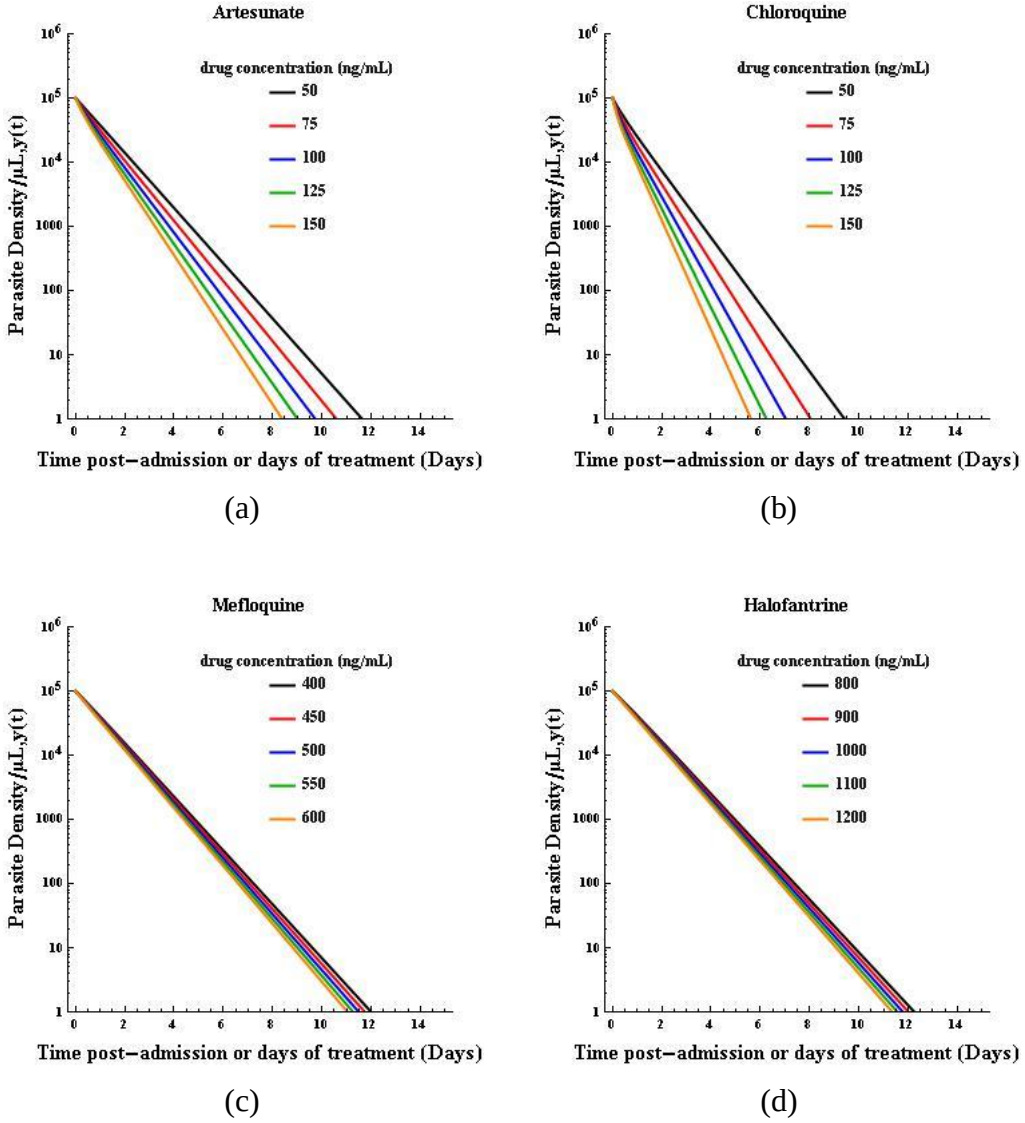


Figure 8. Our numerical results from artesunate, chloroquine, mefloquine and halofantrine in case of *P. falciparum*, shown in Figure 8(a), 8(b), 8(c) and 8(d), respectively.

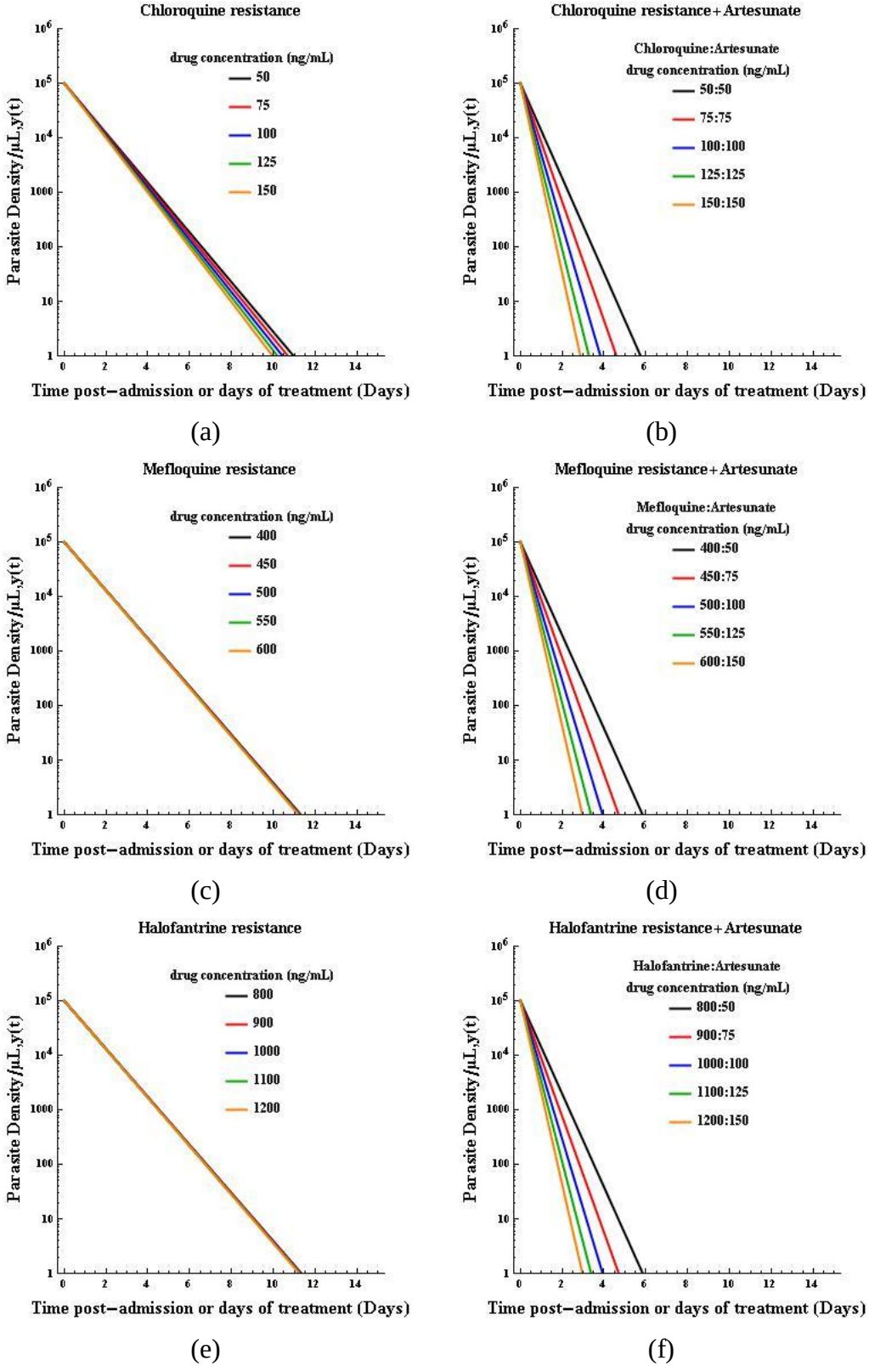


Figure 9. Numerical results of chloroquine, mefloquine and halofantrine with drug resistance and artesunate-based combination therapy (ACT) in case of *P.* non-*falciparum*.

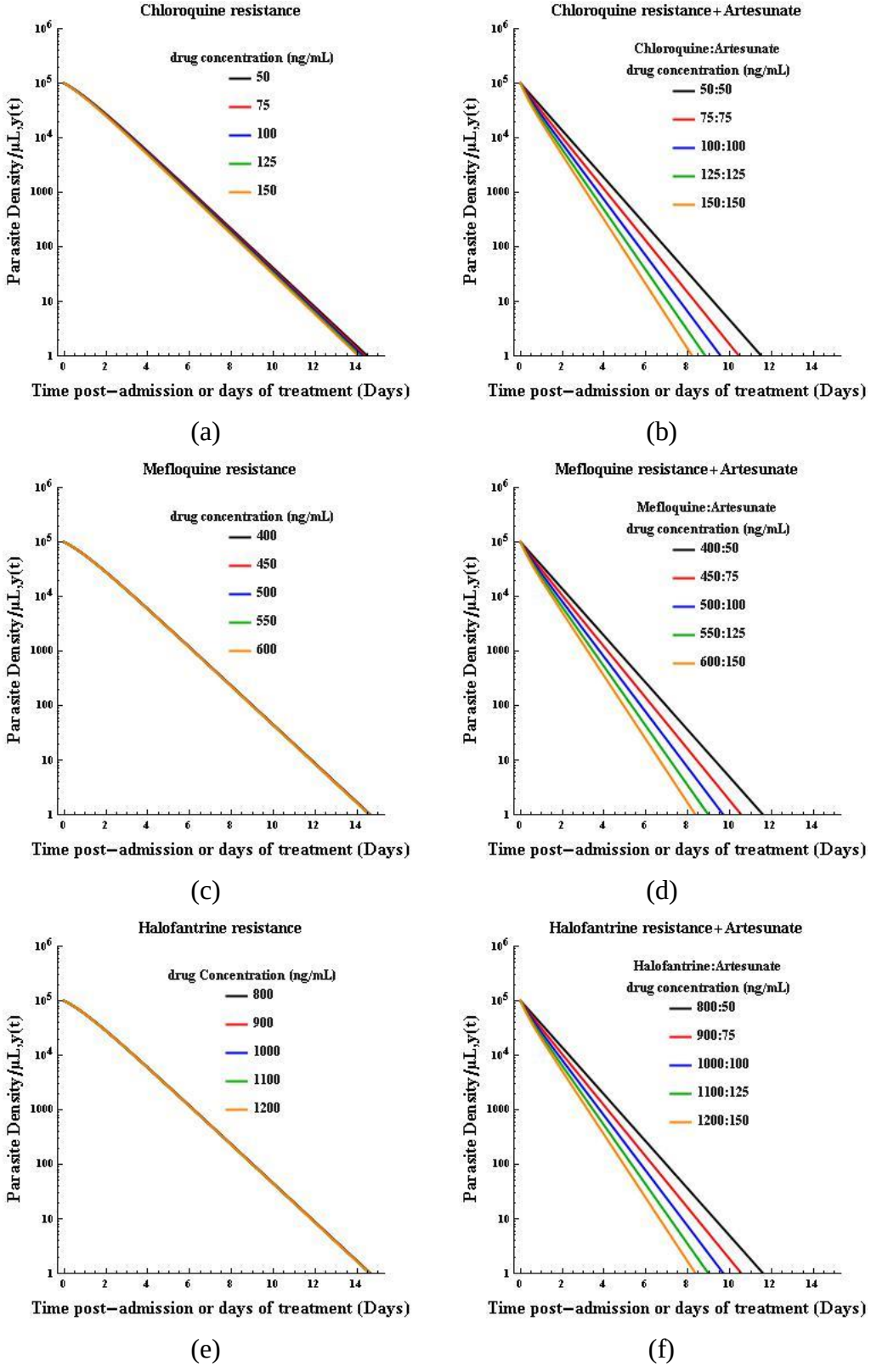


Figure 10. Numerical results of chloroquine, mefloquine and halofantrine with drug resistance and artesunate-based combination therapy (ACT) in case of *P. falciparum*.

In case of monotherapy with resistance and combination with artemisinin (or ACT), the death rate of the malaria parasites in this case is modeled by the partial summation of the death rates in each drug and artesunate with its resistance. In view of calculation, the death rates of the malaria parasite are

$$\left(\frac{y_c}{x+y_c+y_s+E}\right)\left(\frac{\varphi_c^{(drug)}(k)}{\varepsilon}+\varphi_c^{(artesunate)}(k)\right),$$
$$\left(\frac{y_s}{x+y_c+y_s+E}\right)\left(\frac{\varphi_s^{(drug)}(k)}{\varepsilon}+\varphi_s^{(artesunate)}(k)\right) \quad \text{and}$$
$$\left(\frac{y}{x+y+E}\right)\left(\frac{\varphi^{(drug)}(k)}{\varepsilon}+\varphi^{(artesunate)}(k)\right)$$

in cases of *P. falciparum* with circulating and sequestration process, and *P.* non-*falciparum*, respectively (see also Table 3). After generating the results, all treatment durations in case of *P.* non-*falciparum* in Figure 9(b), 9(d) and 9(f), and *P. falciparum* in Figure 10(b), 10(d) and 10(f), are shortened with respect to monotherapy. This confirms that ACT is more effective than monotherapy. Thus, the suggestion is that artesunate should be used in case of drug resistance.

Table 3. The death rates of malaria parasite in cases of general drug, drug with resistance, artesunate and artemisinin-based combination therapy, and population probability factors.

Species	***Plasmodium falciparum***		**Other *P. species***
Process	Circulating	Sequestration	-
Population probability factor	$p_c^{(population)}$ or $\frac{y_c}{x+y_c+y_s+E}$	$p_s^{(population)}$ or $\frac{y_s}{x+y_c+y_s+E}$	$p^{(population)}$ or $\frac{y}{x+y+E}$
Death rates of the malaria parasites			
Drug	$p_c^{(population)}\varphi_c^{(drug)}(k)$	$p_s^{(population)}\varphi_s^{(drug)}(k)$	$p^{(population)}\varphi^{(drug)}(k)$
Drug with resistance	$\frac{p_c^{(population)}\varphi_c^{(drug)}(k)}{\varepsilon}$	$\frac{p_s^{(population)}\varphi_s^{(drug)}(k)}{\varepsilon}$	$\frac{p^{(population)}\varphi^{(drug)}(k)}{\varepsilon}$
Artesunate	$p_c^{(population)}\varphi_c^{(artesunate)}$	$p_s^{(population)}\varphi_s^{(artesunate)}$	$p^{(population)}\varphi^{(artesunate)}(k)$
Artemisinin-based combination therapy (ACT)	$p_c^{(population)}\times\left(\frac{\varphi_c^{(drug)}(k)}{\varepsilon}+\varphi_c^{(artesunate)}\right)$	$p_s^{(population)}\times\left(\frac{\varphi_s^{(drug)}(k)}{\varepsilon}+\varphi_s^{(artesunate)}\right)$	$p^{(population)}\times\left(\frac{\varphi^{(drug)}(k)}{\varepsilon}+\varphi^{(artesunate)}(k)\right)$

Conclusion

The numerical results from the mathematical model of the within-host malaria parasite with antimalarial drugs taking with the immune response of both *Plasmodium falciparum* and other *Plasmodium species* illustrate that all treatment durations are within 1-15 days in cases of all artesunate, chloroquine, mefloquine and halofantrine. Furthermore, treatment duration or the period time to kill both circulating and sequestered iRBCs in these results are 2-12 days, which conform to the patients' clinical data. However, in order to use this research, all of above conditions in this study must be met. The weak points in this study are concerning too many assumptions are needed. Therefore, the improvement for this research is to reduce these conditions or assumptions to make the model conforming to the actual results. Finally, our mathematical models of malaria parasite with antimalarial drugs together with the immune response under hypotheses used with pharmacology, theoretical physics and chemistry are reasonably comparable to the real clinical data from laboratory results and therefore meet our objective to mathematically predict the duration of treatment.

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