AB Journals

African Journal of Mathematics and Statistics Studies
ISSN 2689-5323 • Volume 9, Issue 3, 2026 • pp. 28–44

RESEARCH ARTICLE Received: 12 Aug 2026, Accepted: 25 Aug 2026, Published: 9 Sep 2026

Mathematical Modelling of PfEMP1 Gene Regulation: GF(2) Insights into Chromatin-Mediated Virulence Switching in Plasmodium Falciparum

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Abstract

Plasmodium falciparum possesses complex regulatory mechanisms that enable it to adapt to the host environment and regulate virulence-associated gene expression. PfEMP1, encoded by the var gene family, plays a central role in parasite pathogenicity and immune evasion. However, the mathematical relationship between PfEMP1 expression and epigenetic regulatory factors remains

an important area for understanding parasite transcriptional control. In this study, a finite-field modeling approach was applied to investigate the relationship between PfEMP1 expression and key epigenetic regulators, including PfSIR2A, HP1, and H3K9me3. The parasite regulatory network was represented as a discrete dynamical system and fixed points were identified. Analysis of the fixed-points demonstrated a consistent inverse relationship between PfEMP1 expression and the epigenetic repression-associated state represented by PfSIR2A, HP1, and H3K9me3. PfEMP1 repression was associated with an active epigenetic state (PfSIR2A, HP1, H3K9me3) = (1,1,1), while PfEMP1 activation corresponded with a repressed epigenetic state (PfSIR2A, HP1, H3K9me3) = (0,0,0). These findings suggest that PfEMP1 regulation is controlled through an epigenetic switching mechanism.

Keywords: Plasmodium falciparum, Gene regulation, Epigenetic regulation, Finite field, Fixed points.

How to Cite: Monday, O., Abednego, I. O., & Abraham, E. O. (2026). Mathematical Modelling of PfEMP1 Gene Regulation: GF(2) Insights into Chromatin-Mediated Virulence Switching in Plasmodium Falciparum. African Journal of Mathematics and Statistics Studies, 9(3), 28–44. https://doi.org/10.52589/AJMSS-8KGONOAO

INTRODUCTION

Background to the Study

Malaria is a disease caused by unicellular parasites of the Plasmodium genus, which are transmitted to humans through the bites of infected female Anopheles mosquitoes (World Health Organization, 2022). Six different Plasmodium species can infect humans: *P. vivax, P. falciparum, P. ovale, P. malariae, P. knowlesi*, and *P. cynomolgi*, with falciparum malaria being the most virulent species responsible for the majority of severe and fatal cases of malaria (David et al., 2019; Jensen et al., 2020).

During its complex life cycle, *Plasmodium* parasites differentiate into multiple developmental stages, switching between sexual and asexual forms in their primary (mosquito) and intermediate (human) hosts, respectively (Tripathi et al., 2022; Rajneesh et al., 2023). The transmission of malaria begins when an infected mosquito bites a human, injecting sporozoites into the bloodstream. These sporozoites quickly travel to the liver, where they invade hepatocytes and undergo asexual replication, producing thousands of merozoites. The merozoites are released into the bloodstream, where they invade red blood cells (RBCs). Inside the RBCs, P. falciparum undergoes a 48-hour asexual replication cycle, leading to the destruction of the RBCs and the release of more merozoites, which can then infect other RBCs (Rajneesh et al., 2023). This cycle of invasion, replication, and RBC lysis is responsible for the clinical manifestations of malaria, including fever, chills, anaemia, and, in severe cases, cerebral malaria, which can be fatal (Bakary et al., 2018; Balau et al., 2023).

The parasite's molecular structure is characterized by its intricate genome, which is rich in adenine-thymine (AT) pairs, accounting for about 80% of the DNA, contributing to the unique structural features of P. falciparum. This AT-rich genome encodes a variety of proteins, including those involved in immune evasion, such as the Plasmodium falciparum erythrocyte membrane protein 1 (PfEMP1), which is responsible for antigenic variation (Matthews et al., 2020; Rovira-Graells et al., 2022).

The genetic diversity of Plasmodium parasites poses a significant challenge to malaria control. The rapid mutation allows the parasite to produce variants that evade immune detection, reducing vaccine effectiveness, especially in regions with diverse strains. This genetic variability limits the long-term efficacy of existing vaccines and complicates their application in different geographical areas (Olivier et al., 2025). Having an understanding of the expression mechanisms underlying gene expression in P. falciparum and disrupting such mechanisms are potential strategies for developing new antimalarial therapies and vaccines for malaria control and prevention (Barbuti et al., 2025). Also, having the knowledge and understanding of how these genetic factors interact to activate or suppress genes is essential for developing new antimalarial therapies and vaccines for malaria control and prevention (Olivier et al., 2025).

This study proposes a finite-field modeling approach to investigate the association between PfEMP1 expression and key epigenetic regulators, including PfSIR2A, HP1, and H3K9me3. The parasite regulatory network was represented as a discrete dynamical system $F: K^4 \rightarrow K^4$, where K represents $GF(2)$, describing binary gene expression states. For a proper understanding of gene interactions and regulatory relationships, the biological gene interactions (activation/repression) were modeled into a logical network model and expressed in logic algebraic rules (functions). The model was translated into a formal mathematical framework of polynomial equations to uncover stable gene expression pathways and identify key control nodes within the network. This algebraic framework provides a rigorous and flexible

foundation for integrating biological data, predicting the system behaviour, and identifying potential molecular targets for therapeutic intervention.

PRELIMINARY

Genes

A gene is a fundamental unit of heredity in living organisms. It is a specific sequence of nucleotides within deoxyribonucleic acid (DNA) that contains the instructions for building and maintaining an organism's cells and passing genetic traits to offspring. Genes store the information required to produce proteins, which are essential for the structure, function, and regulation of the body's tissues and organs (Harvey et al., 2021).

Gene Expressions and Regulations

Gene expression refers to the process by which the information in the DNA is transformed into cellular function. Towards this end, a series of steps takes place that starts with the transcription of the gene from the DNA into another form known as the ribonucleic acid (RNA), followed by the translation of the RNA into a protein that is then assembled and processed in the necessary way to carry out its ultimate function. This flow of information is commonly referred to as the central dogma of molecular biology, which stipulates that information flows in one direction only (Aleem, 2010). For example: $X_i(t+1) = X_1(t) \vee \neg X_2(t)$ means gene X_i is activated if X_1 is ON OR X_2 is OFF (Thakar (2024).

Gene expression in Plasmodium falciparum (P. falciparum) is a highly regulated and complex process that plays a critical role in the parasite's ability to adapt to different environments and evade the host immune system (Real et al., 2021).

Finite Field

Finite fields involve algebraic structures with a finite number of elements. This approach can model the effects of genetic variations and mutations on gene regulatory networks. Finite field models are useful for capturing non-linear dynamics and complex interactions in genetic networks. They provide a precise mathematical framework for studying how different genetic configurations affect gene expression and contribute to phenotypic diversity. In a finite field, each gene (or regulator) is represented by a discrete state in a finite field GF(q), where q encodes biologically meaningful levels (OFF/HIGH ⇒ q = 2; OFF/LOW/HIGH ⇒ q = 3). The update of an n-gene network is a polynomial map F: $GF(q)^n \rightarrow GF(q)^n$, with one coordinate polynomial per gene, such that $x(t+1) = F(x(t))$. (Raina and Macaulay, 2019).

Fixed point

A fixed-point $x \in \mathbb{F}_2^n$ is a state satisfying $F(x) = x$. Such a state remains unchanged over time and typically represents a stable phenotype or gene expression pattern, which the system remains in indefinitely once reached. Fixed points often correspond to stable gene expression patterns, such as cell types or steady biological states. (Raina and Macaulay, 2019).

THE MODEL AND RESULTS

Variables for the Full Network Model

Variables for the model are given in Table 3.1 below:

Table 3.1: Core genes/regulators of *Plasmodium falciparum* DNA.

Gene / Regulator	Full name	Function
PfEMP1 (var genes)	Plasmodium falciparum erythrocyte membrane protein 1	Immune evasion and survival strategies gene
PfCRT	Plasmodium falciparum chloroquine resistance transporter	Chloroquine resistance gene
PfMDR1	Plasmodium falciparum multidrug resistance protein 1	Multidrug resistance gene
PfDHFR	Plasmodium falciparum dihydrofolate reductase	Antifolate drug resistance gene
AP2-G	ApiAP2/AP2-domain transcription factor G	Gametocyte commitment/transmission gene
PfSIR2A	Plasmodium falciparum Silent Information Regulator 2A;	Var genes repressor gene
PfK13	Plasmodium falciparum Kelch 13 protein	Artemisinin resistance gene
HP1	Heterochromatin Protein 1	Genome stability maintenance gene
H3K9me3	Histone H3 lysine 9 trimethylation	Epigenetic memory (pattern of gene activity) gene

Parameters for the Full Network Model

Table 3.2: (Core stress/drug pressures) and selectors of Plasmodium falciparum in humans.

Stress signal	Symbol	Selectors
Stress (general host/drug pressure)	ω	Immune system, drug pressure, etc. response to attack
CQ	λ	Chloroquine gene selector
s-Partners	ρ	Partner drug gene selector (amodiaquine/lumefantrine/mefloquine).
AF	α	Antifolate gene selector
ART	β	Artemisinin gene selector.

Sources of Table 3.1 and Table 3.2: Adegbola et al. (2023); *Bui et al. (2021); Schmedes et al.*

(2021); Maiga et al. (2021); Kayode et al. (2021); Stewart et al. (2022); Adegbola et al. (2023);

Wyss et al. (2024); Melong et al. (2024); Voss et al. (2024); Hviid et al. (2024); Zhou et al. (2024); Walker et al. (2025); Fançony et al. (2025); World Malaria Report (2024, 2025); *WHO (2025);* etc.

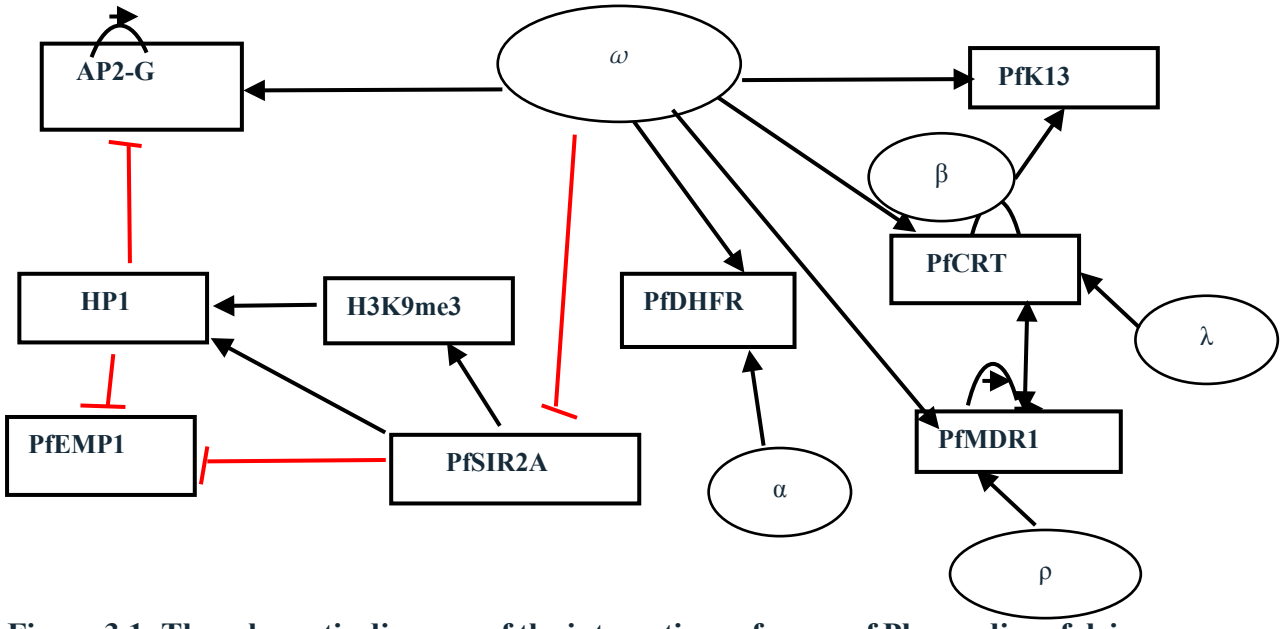

Figure 3.1: The schematic diagram of the interactions of genes of Plasmodium falciparum associated with DNA complexity in the human host.

The networks shown above indicate that the parasite's complexity is not just in its gene count but also in the regulatory logic rules by which genes or regulators control each other's activities encoded in its DNA. This allows it to dynamically switch or toggle gene states to balance growth, defence, and transmission, making it an incredibly adaptable pathogen. This Boolean network model helps illustrate this complexity, providing a computational method to simulate, predict, and possibly intervene in these decision-making processes for therapeutic purposes.

In this paper, we are mathematically projecting a 4-node variable sub-network and one parameter (Stress ω) to investigate the association between PfEMP1 expression and key epigenetic regulators, including PfSIR2A, HP1, and H3K9me3, as shown in the table below. This is due to the fact that these genes/regulators are connected in gene regulation as shown in the schematic diagram above.

Gene / Regulator	Full name	Function
PfEMP1 Table (var genes)	Plasmodium falciparum erythrocyte membrane protein 1	Immune evasion and survival strategies gene
PfSIR2A	Plasmodium falciparum Silent Information Regulator 2A;	Var genes repressor gene
HP1	Heterochromatin Protein 1	Genome stability maintenance gene
H3K9me3	Histone H3 lysine 9 trimethylation	Epigenetic memory (pattern of gene activity) gene

Core Genes/Regulators

Table 3.4: (Core stress/drug pressures)

Stress signal	symbol	Selectors
Stress (general host/drug pressure)	ω	Immune system, drug pressure, etc. response to attack

Biological Interaction/Update Rules Functions

PfEMP1: The alteration in the genetic make of this gene encodes a major virulence factor responsible for cytoadherence and antigenic variation. It is silenced by HP1 and PfSIR2A through heterochromatin formation and histone deacetylation, and its activation is influenced by stress.

Thus, PfEMP1 (t +1) = NOT HP1 AND NOT PfSIR2A AND STRESS.

$$= \neg \text{HP1} \wedge \neg \text{PfSIR2A} \wedge \text{STRESS} \quad 3.1$$

PfEMP1 is active only if epigenetic silencing is OFF. Only one PfEMP1 variant is expressed at a time to avoid immune detection, which is regulated epigenetically by heterochromatin (HP1 and PfSIR2A).

PfSIR2A: This is a dependent histone deacetylase regulating gene silencing, and PfSIR2A's expression or activity changes in response to host immune-related stress signals, which supports the idea that immune stress can modulate PfSIR2A repression. It indirectly influences the repression of PfEMP1 via chromatin modification.

Thus, PfSIR2A (t + 1) = NOT Stress = ¬ Stress Signal 3.2

It serves as an environmental sensor, helping the parasite repress virulence genes when stress is low.

HP1: A chromatin-associated protein is crucial for gene silencing via heterochromatin formation modulated by H3K9me3 and supported by PfSIR2A.

Thus, HP1 (t + 1) = H3K9me3 AND PfSIR2A

$$= \text{H3K9me3} \wedge \text{PfSIR2A} \quad 3.3$$

It represses var gene expression (PfEMP1) and gametocyte switch genes to avoid parasite proliferation.

H3K9me3: This is an Epigenetic mark for heterochromatin promoted by PfSIR2A**.**

Thus, H3K9me3 (t + 1) = PfSIR2A 3.4

It helps recruit HP1 to a specific luminous protein for transcriptional repression.

The Logic Rules and State Transitions

The rules below were used to convert the logic rules and state transitions update

A OR B = A ∨ B = A + B + A · B

A OR B OR C = A ∨ B ∨ C = A + B + C +AB + AC + BC + ABC

A AND B = A ∧ B = A · B

NOT A = ¬ A = 1 + A

We now have the logic rules and state transitions as follows:

PfEMP1 (t +1) = ¬ HP1 ∧ ¬ PfSIR2A ∧ Stress

$$PfEMP1\ (t+1) = (1 + HP1(t)) \cdot (1 + PfSIR2A\ (t)) \cdot \omega \quad 3.5$$

PfSIR2A (t +1) = ¬ Stress Signal.

$$PfSIR2A\ (t+1) = 1 + Stress = (1 + \omega) \quad 3.6$$

HP1 (t + 1) = H3K9me3 ∧ PfSIR2A

$$HP1\ (t+1) = H3K9me3(t) \cdot PfSIR2A(t) \quad 3.7$$

H3K9me3 (t + 1) = PfSIR2A(t)

$$H3K9me3\ (t+1) = PfSIR2A(t) \quad 3.8$$

Square Free Polynomials

we convert the update functions from logic expressions into square-free polynomials. To analyze the model for convenience, we rename the variables as follows:

PfEMP1 → X_1

PfSIR2A → X_2

HP1 → X_3

H3K9me3 → X_4

The polynomial form of the Equation (3.5 – 3.8) is as follows:

$$X_1(t+1) = (1 + X_3(t)) \cdot (1 + X_2(t)) \cdot \omega \quad 3.9$$

$$X_2(t+1) = (1 + \omega) \quad 3.10$$

$$X_3(t+1) = X_4(t) \cdot X_2(t) \quad 3.11$$

$$X_4(t+1) = X_2(t) \quad 3.12$$

Solving for Fixed Point Equations In Gf_2

Recall: At a fixed point, $X_i(t+1) = X_i(t)$ in F_2

From Equation (3.10):

$(X_2\ (t+1)) = (1+\omega)$

But for a fixed point, $X_2(t+1) = X_2(t)$

Hence, $X_2(t) = (1+\omega)$ 3.13

From Equation (3.12):

$(X_4\ (t+1)) = X_2(t)$

But for a fixed point, $X_4(t+1) = X_4(t)$

Which implies that: $X_4(t+1) = X_4(t) = X_2(t)$

Combining with equation (3.13), we have

$(X_4\ (t+1)) = X_4(t) = X_2(t) = (1+\omega)$

Hence, $X_4\ (t) = (1+\omega)$ 3.14

From Equation (3.11):

$(X_3\ (t+1)) = X_4(t)\cdot X_2(t)$

But for a fixed point, $X_3(t+1) = X_3(t)$

Which implies that: $X_3(t+1) = X_3(t) = X_4(t)\cdot X_2(t)$

Combining with equation (3.13 and 3.14), we have

$(X_3\ (t+1)) = X_3(t) = X_4(t)\cdot X_2(t) = (1+\omega)\ (1+\omega) = 1+\omega+\omega+\omega\cdot\omega$

But $\omega\cdot\omega = \omega$, and $\omega+\omega = 0$.

Hence, $X_3\ (t) = 1+\omega$ 3.15

From equation (3.9)

$X_1(t+1) = (1+X_3(t))\cdot(1+X_2(t))\cdot\omega = \omega+\omega\ (X_3+X_2)+\omega\ (X_3\cdot X_2)$

But for a fixed point, $(X_1\ (t+1)) = X_1\ (t)$

$X_1(t+1) = X_1(t) = \omega+\omega\ (X_3+X_2)+\omega\ (X_3\cdot X_2)$

Combining with equation (3.13) and equation (3.15), we have

$X_1(t) = \omega+\omega\ (1+\omega+1+\omega)+\omega\ (1+\omega)(1+\omega)$

$X_1(t) = \omega+\omega(1+\omega)(1+\omega) = \omega+\omega(1+\omega) = \omega$

Hence, $X_1(t) = \omega$. 3.16

Putting all the fixed-point equations together, we have

$X_1(t+1) = \omega$

$X_2(t) = (1 + \omega)$

$X_3(t) = (1 + \omega)$

$X_4(t) = (1 + \omega)$

RESULT/FINDINGS

Finite field (GF2) fixed-point result

Now, we find the fixed point(s) for **stress (ω) = 0** and **stress (ω) = 1**, using the equations (3.13) – (3.16).

For stress (ω) = 0

$X_1(t+1) = \omega = \mathbf{0}$

$X_2(t+1) = (1 + \omega) = 1 + 0 = 1$

$X_3(t+1) = (1 + \omega) = 1 + 0 = 1$

$X_4(t+1) = (1 + \omega) = 1 + 0 = 1$

The Fixed points for all stress = 0 are:

Fixed Point = (0, 1, 1, 1),

$(X_1(t), \ldots, X_4(t))$ = (PfEMP1, PfSIR2A, HP1, H3K9me3).

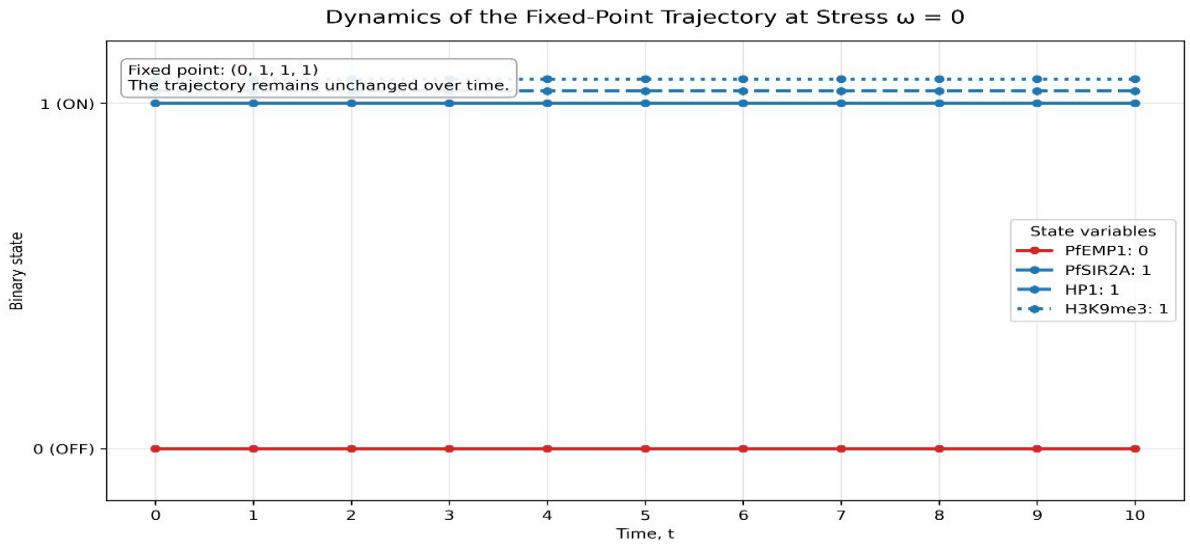

Figure 3.1: Dynamic fixed-point trajectory (stress (ω) = 0) fixed point = (0,1,1,1)

Remark: PfEMP1 remains at 0; PfSIR2A, HP1, and H3K9me3 remain at 1 for all times (stable fixed point).

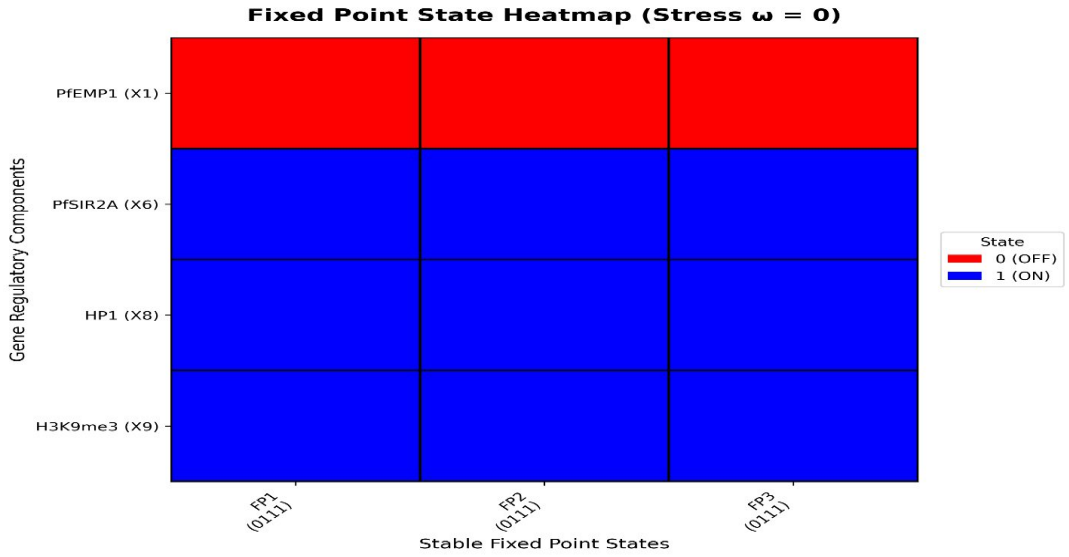

Figure 3.2: Fixed point state heatmap (stress (ω) = 0) fixed point = (0,1,1,1)

Remark: : PfEMP1 remains at 0; PfSIR2A, HP1, and H3K9me3 remain at 1 for all times confirming that the system reached a stable phenotype immediately, which indicates that it is a fixed point.

For stress (ω) = 1

$X_1(t+1) = \omega = \mathbf{1}$

$X_2(t+1) = (1+\omega) = 1+1 = 0$

$X_3(t+1) = (1+\omega) \quad = 1+1 = 0$

$X_4(t+1) = (1+\omega) \quad = 1+1 = 0$

The Fixed points for stress = 1 is:

Fixed point: (1, 0, 0, 0).

$(X_1(t), \ldots, X_4(t))$ = (PfEMP1, PfSIR2A, HP1, H3K9me3).

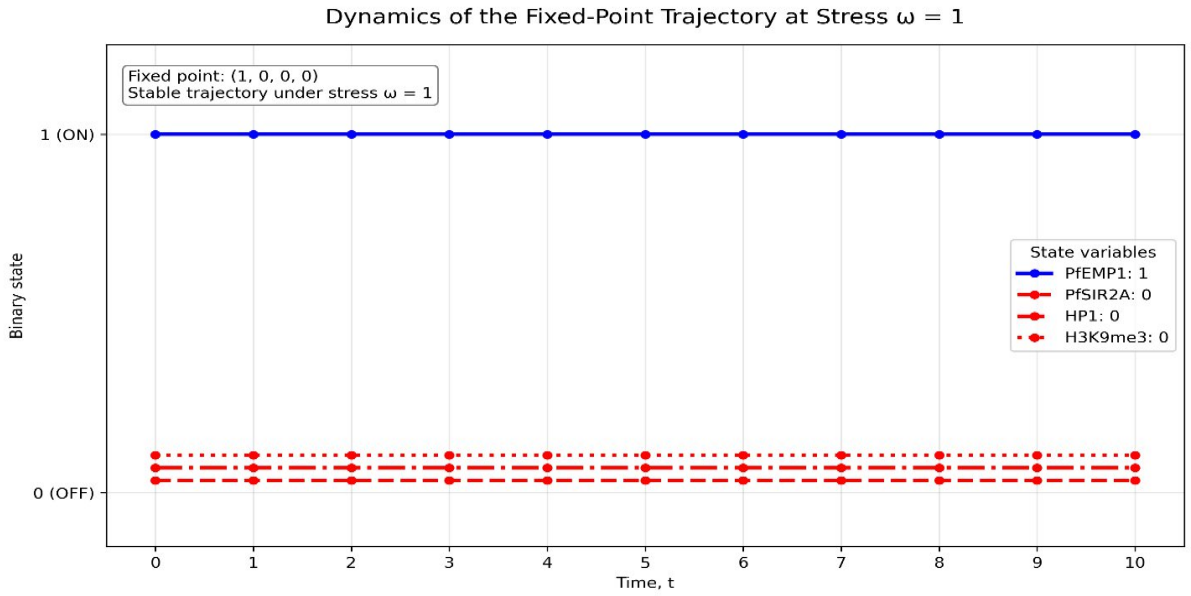

Figure 3.3: Dynamic fixed-point trajectory (stress (ω) = 1) fixed point = (1,0,0,0)

Remark: PfEMP1 remains at 1; PfSIR2A, HP1, and H3K9me3 remain at 0 for all times (stable fixed point).

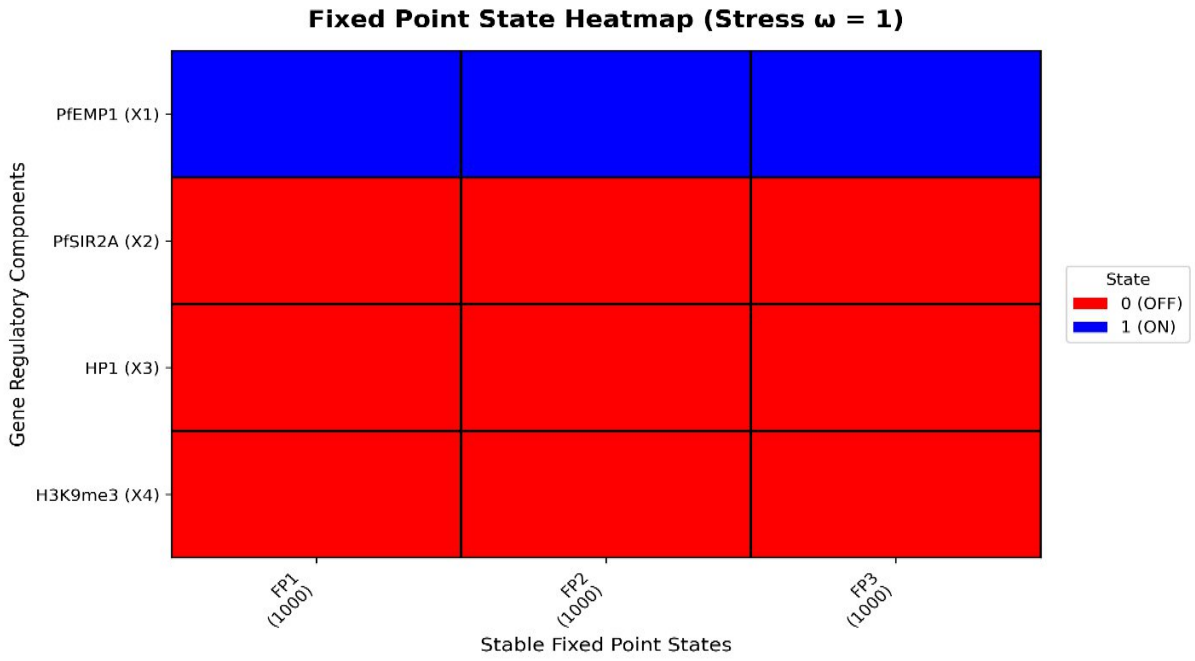

Figure 3.4: Fixed-point state heatmap (stress (ω) = 1) fixed point = (1,0,0,0)

Remark: PfEMP1 remains at 1; PfSIR2A, HP1, and H3K9me3 remain at 0 for all times, confirming that the system reached a stable phenotype immediately, which indicates that it is a fixed point.

Theorem

Let $GF(2) = \{0, 1\}$ represent a binary gene expression system Consider the regulatory system F: $K^4 \rightarrow K^4$ be the regulatory map over $K = GF(2)$ with state vectors $X_i(t) = (X_1(t),\ldots, X_4(t))$

where $(X_1(t),\ldots, X_4(t))$ = (PfEMP1, PfSIR2A, HP1, $H_3K_9me_3$). Let $Fix(F) = \{Xi: F(Xi) = Xi\}$ be the fixed-point set. The fixed-point solutions obtained from the parasite regulatory model show that in $GF(2)$, when PfEMP1 = 0, (PfSIR2A, HP1, H3K9me3) = (1, 1, 1) and when PfEMP1= 1, (PfSIR2A, HP1, H3K9me3) = (0, 0, 0). Therefore, PfEMP1 expression is inversely associated with the epigenetic repression state (PfSIR2A, HP1, H3K9me3).

Proof

A fixed point satisfies $F(X_i(t+1)) = X_i(t)$, meaning that every gene expression state remains unchanged after the regulatory update.
Define the epigenetic projection $E(X_i(t)) = (X_2(t), X_3(t), X_4(t))$, where $X_2(t)$ = PfSIR2A, $X_3(t)$= HP1, and $X_4(t)$ = H3K9me3, which represents the chromatin-associated regulatory state.

Under a stress-free environment $(\omega) = 0$, the fixed points show two possible complementary states:
$X_1(t) = \omega = 0$
$X_2(t) = (1 + \omega) = 1 + 0 = 1$
$X_3(t) = (1 + \omega) = 1 + 0 = 1$

$X_4(t) = (1 + \omega) = 1 + 0 = 1$
Hence, when $X_1(t) = 0$, the epigenetic projection gives $E(X_i(t)) = (1, 1, 1)$.

Under a stress environment $(\omega) = 1$, the fixed points show two possible complementary states:
$X_1(t) = \omega = 1$
$X_2(t) = (1 + \omega) = 1 + 1 = 0$
$X_3(t) = (1 + \omega) = 1 + 1 = 0$

$X_4(t) = (1 + \omega) = 1 + 1 = 0$
Hence, when $X_1(t) = 1$, the epigenetic projection gives $E(X_i(t)) = (0, 0, 0)$.
Hence, the transition satisfies:
PfEMP1 OFF ↔ epigenetic repression ON and PfEMP1 ON ↔ epigenetic repression OFF.
Therefore, PfEMP1 and the repression subsystem exhibit complementary behaviour.

Corollary:

If a regulatory network contains fixed points satisfying PfEMP1= φ and (PfSIR2A, HP1, H3K9me3) = (μ,μ,μ), where increasing φ corresponds to decreasing μ, then PfEMP1 expression is controlled by an epigenetic switching mechanism.

Proof

From the model in $GF(2)$,
PfEMP1 activations is associated with loss of repression markers.

The PfEMP1 component fixed-point condition is $X_1(t) = \omega$
The PfSIR2A component fixed-point condition is $X_2(t) = (1 + \omega)$

The HP1 component fixed-point condition is $X_3(t) = (1 + \omega)$

The H3K9me3 component fixed point condition is $X_4(t) = (1 + \omega)$ For PfEMP1= $\varphi = \{0, 1\}$, (PfSIR2A, HP1, H3K9me3) $= (\mu,\mu,\mu) = (1, 1, 1,)$, (0, 0, 0).

Therefore, the parasite regulatory system possesses an epigenetic switch controlling PfEMP1 expression.

Remark: The mathematical result does not imply that PfSIR2A, HP1 and H3K9me3 are the only regulators of PfEMP1 expression. Rather, the two attractor states indicates that these variables are strongly associated with PfEMP1 regulation.

The model supports the following interpretations;

1. High PfSIR2A/HP1/H3K9me3 maintains chromatin repression, which keeps PfEMP1 inactive.
2. Reduced PfSIR2A/HP1/H3K9me3 allows chromatin expression, which permits PfEMP1 activation

DISCUSSIONS

The fixed-point analysis provides a mathematical basis for identifying critical regulatory components within the parasite gene-expression network. The existence of two distinct fixed points, corresponding to the PfEMP1 OFF (0, 1, 1, 1) and PfEMP1 ON (1, 0, 0, 0) configurations, indicates a bistable regulatory structure in which the system can occupy alternative stable states. This property suggests that PfEMP1 expression is closely coupled to the epigenetic state of the parasite, and therefore, this relationship provides an opportunity to investigate therapeutic strategies that target not only PfEMP1 itself but also the regulatory mechanisms that determine whether PfEMP1 is expressed.

Framework for new drug/ Vaccine development

The fixed-point analysis from a drug-development perspective, PfSIR2A, HP1, and the molecular machinery responsible for establishing or maintaining H3K9me3 represent potential regulatory targets. Pharmacological modulation of these components could, in principle, alter the stability of the PfEMP1-expressing state and promote a less virulent or more immunologically vulnerable parasite phenotype. The fixed-point framework therefore provides a means of prioritizing candidate regulatory molecules for experimental validation and identifying points within the epigenetic network where therapeutic intervention may disrupt PfEMP1 regulation.

For vaccine development, the fixed-point analysis highlights an important challenge associated with PfEMP1 antigenic variation. Since the parasite can switch between different expression states and PfEMP1 variants, vaccines directed against individual highly variable PfEMP1 forms may have limited breadth. The regulatory framework therefore encourages investigation of conserved molecular features associated with PfEMP1 regulation, as well as conserved regions of PfEMP1 that remain functionally important across variants. Targeting conserved determinants could potentially provide broader protection than targeting a single variable PfEMP1 antigen.

SUMMARY

This study used a finite-field framework to represent the parasite regulatory network as a nine-variable discrete dynamical system over GF(2). The analysis focused particularly on the relationship between PfEMP1 expression and three epigenetic factors associated with var gene silencing: PfSIR2A, HP1, and H3K9me3. The fixed-point solutions revealed a clear inverse association between PfEMP1 expression and the coordinated activity of the three repression-associated regulators. PfEMP1 was silenced when PfSIR2A, HP1, and H3K9me3 occupied the active state (1,1,1), whereas PfEMP1 expression was permitted when these factors occupied the inactive state (0,0,0). This pattern supports a switch-like regulatory system in which coordinated epigenetic repression stabilizes the silent var gene state, while release from repression favors PfEMP1 activation. The finite-field model therefore provides a concise mathematical representation of stable regulatory states that may underlie antigenic switching in the parasite.

CONCLUSION

The finite-field (GF2) analysis indicates that PfEMP1 expression is governed by a coordinated epigenetic switching mechanism involving PfSIR2A, HP1, and H3K9me3. The stable-state structure of the model places active epigenetic repression in opposition to PfEMP1 expression. The combined active state of the three regulators corresponds to PfEMP1 silencing, while their combined inactive state corresponds to PfEMP1 activation. These findings are consistent with the central role of heterochromatin-based regulation in maintaining mutually exclusive var gene expression and enabling antigenic variation. This model also shows that finite-field methods can be applied to parasite gene-regulatory systems to identify attractor states and clarify relationships among regulatory components.

Future Research

Future work should extend the network by incorporating additional transcriptional and chromatin regulators, multilevel or probabilistic expression states, and experimentally measured transition dynamics. Validation against transcriptomic, epigenomic, and chromatin-occupancy data would further determine how accurately the predicted fixed points reflect biological states. Such extensions could deepen understanding of PfEMP1 switching and support the identification of regulatory vulnerabilities relevant to malaria pathogenesis and therapeutic research.

REFERENCES

Adegbola, A. J., Ijarotimi, O. A., Ubom, A. E., Adesoji, B. A., Babalola, O. E., Hocke, E. F., Hansson, H., Mousa, A., Bolaji, O. O., Alifrangis, M., & Roper, C. (2023). A snapshot of the prevalence of dihydropteroate synthase-431V mutation and other sulfadoxine-pyrimethamine resistance markers in Plasmodium falciparum isolates in Nigeria. *Malaria journal, 22*(1*),* 71. https://doi.org/10.1186/s12936-023-04487-5

Akerejola R. F., Elakhe O. A., and Isere A. O. (2024). A non-standard finite difference discretization scheme applied to a malaria model, *African Journal of Mathematics and Statistics Studies, Vol. 7*(4), 226 - 247.

Aleem H. (2010). An Algebraic Approach to Modelling the Regulation of Gene Expression. *Ph.D. Thesis*, University of Manchester.

Balau, A., Sobral, D., Abrantes, P., Santos, I., Mixão, V., Gomes, J. P., Antunes, S., & Arez, A. P. (2023). Differential Gene Expression of Malaria Parasite in Response to Red Blood Cell-Specific Glycolytic Intermediate 2,3-Diphosphoglycerate (2,3-DPG). *International journal of molecular sciences, 24*(23), 16869. https://doi.org/10.3390/ijms242316869

Bakary, T., Boureima, S., and Sado, T. (2018). A mathematical model of malaria transmission in a periodic environment. *Journal of Biological Dynamics,12*(1), 400–432, https://doi.org/10.1080/17513758.2018.1468935

Barbuti, R., Gori, R., Milazzo, P. *et al.* (2020). A survey of gene regulatory networks modelling methods: from differential equations, to Boolean and qualitative bioinspired models. *J Membr Comput 2*, 207–226. https://doi.org/10.1007/s41965-020-00046-y

Blake TCA, Haase S, Baum J (2020) Actomyosin forces and the energetics of red blood cell invasion by the malaria parasite Plasmodium falciparum. *PLoS Pathog 16*(10): e1009007. https://doi.org/10.1371/journal.ppat.1009007

Bui, H. T. N., Passecker, A., Brancucci, N. M. B., & Voss, T. S. (2021). Investigation of Heterochromatin Protein 1 Function in the Malaria Parasite Plasmodium falciparum Using a Conditional Domain Deletion and Swapping Approach. *mSphere*, *6*(1), e01220-20. https://doi.org/10.1128/mSphere.01220-20

Chen, J., Wang, Q., He, X., & Yang, B. (2025). Malaria Vaccines: Current Achievements and Path Forward. *Vaccines*, *13*(5), 542. https://doi.org/10.3390/vaccines13050542

Diffendall, G., & Scherf, A. (2024). Deciphering the Plasmodium falciparum perinuclear var gene expression site. *Trends in Parasitology. 40*. 10.1016/j.pt.2024.06.002.

Harvey F. L., Arnold B., Chris K., K., Anthony B., Hidde L. P., Kelsey C. M., Michael B. Y., Angelika A. (2021). Molecular Cell Biology, 9th Edition, by Lodish (January 15, 2021), ISBN 10: 1319208525 / ISBN 13: 9781319208523, published by W. H. Freeman, 2021.

Hviid, L., Jensen, A. R., & Deitsch, K. W. (2024). PfEMP1 and var genes - Still of key importance in Plasmodium falciparum malaria pathogenesis and immunity. *Advances in parasitology*, *125*, 53–103. https://doi.org/10.1016/bs.apar.2024.02.001

Isere A. O., Osemwenkhae J. E. and Okuonghae D. (2014). Optimal control model for the outbreak of cholera in Nigeria. *African Journal of Mathematics and Computer Science research, 7*(2), 24-30.

Isere A. O. (2024), A mathematical model of the effects of strikes on Nigerian universities. *arXiv:2409.02945v1* [math. HO], 28 Aug 2024

Jensen, A. R., Adams, Y., & Hviid, L. (2020). Cerebral Plasmodium falciparum malaria: The role of PfEMP1 in its pathogenesis and immunity, and PfEMP1-based vaccines to prevent it. *Immunological reviews*, *293*(1), 230–252. https://doi.org/10.1111/imr.12807

Kayode, A. T., Akano, K., Ajogbasile, F. V., Uwanibe, J. N., Oluniyi, P. E., Bankole, B. E., Eromon, P. J., Sowunmi, A., Folarin, O. A., Volkman, S. K., McInnis, B., Sabeti, P., Wirth, D. F., & Happi, C. T. (2021). Polymorphisms in Plasmodium falciparum chloroquine resistance transporter (Pfcrt) and multidrug-resistant gene 1 (Pfmdr-1) in Nigerian children 10 years post-adoption of artemisinin-based combination treatments. *International journal for parasitology*, *51*(4), 301–310. https://doi.org/10.1016/j.ijpara.2020.10.001

Ma, R., Salinas, N. D., Orr-Gonzalez, S., Richardson, B., Ouahes, T., Torano, H., Jenkins, B. J., Dickey, T. H., Neal, J., Duan, J., Morrison, R. D., Gittis, A. G., Doritchamou, J. Y. A., Zaidi, I., Lambert, L. E., Duffy, P. E., & Tolia, N. H. (2024). Structure-guided design of VAR2CSA-based immunogens and a cocktail strategy for a placental malaria

vaccine. *PLoS pathogens*, *20*(3), e1011879. https://doi.org/10.1371/journal.ppat.1011879

Maiga, H., Grivoyannis, A., Sagara, I., Traore, K., Traore, O. B., Tolo, Y., Traore, A., Bamadio, A., Traore, Z. I., Sanogo, K., Doumbo, O. K., Plowe, C. V., & Djimde, A. A. (2021). Selection of *pfcrt* K76 and *pfmdr1* N86 Coding Alleles after Uncomplicated Malaria Treatment by Artemether-Lumefantrine in Mali. *International journal of molecular sciences*, *22*(11), 6057. https://doi.org/10.3390/ijms22116057

Melong, C. S. M., Peloewetse, E., Russo, G., Tamgue, O., Tchoumbougnang, F., & Paganotti, G. M. (2024). Correction to: An overview of artemisinin-resistant malaria and associated Pfk13 gene mutations in Central Africa. *Parasitology research*, *123*(8), 309. https://doi.org/10.1007/s00436-024-08315-w

Ndung'u, L., Thiong'o, K., Karani, L., Gitahi, S., Kimani, F., Ngugi, M. P., & Kiboi, D. (2026). Persistent and Circulating *Plasmodium falciparum dhfr* and *dhps* Mutations in Busia County, Western Kenya. *Pathogens (Basel, Switzerland)*, *15*(2), 233. https://doi.org/10.3390/pathogens15020233

Nezamuldeen, Leena, & Jafri, M. Seleet. (2024). Boolean Modeling of Biological Network Applied to Protein-Protein Interaction Network of Autism Patients. *Biology*, *13*(8), 606. https://doi.org/10.3390/biology13080606

Olivier S., John B., Sulymon A. S., Marie G. U., Alex M. K., Abraham O., Samuel O. D., Lewis T. B., Beloved of God Agbelemoge, and Richard O. O. (2025); Routine malaria vaccination in Africa: a step toward malaria eradication? *Malaria Journal (2025) 24:1* https://doi.org/10.1186/s12936-024-05235-z

Raina R. and Macauley M. (2019). *Algebraic and Computational Biology*. Mathematics in Science and Engineering. Series Editor Goong Chen

Rajneesh, Tiwari R., Singh V.K., Kumar A., Gupta R.P., Singh A.K., Gautam V., Kumar R. (2023). Advancements and Challenges in Developing Malaria Vaccines: Targeting Multiple Stages of the Parasite Life Cycle. *ACS Infect. Dis. 9*:1795–1814. doi: 10.1021/acsinfecdis.3c00332

Real, E., Howick, V. M., Dahalan, F. A., Witmer, K., Cudini, J., Andradi-Brown, C., Blight, J., Davidson, M. S., Dogga, S. K., Reid, A. J., Baum, J., & Lawniczak, M. K. N. (2021). A single-cell atlas of Plasmodium falciparum transmission through the mosquito. *Nature communications*, *12*(1), 3196. https://doi.org/10.1038/s41467-021-23434-z

Rocamora, F., & Winzeler, E. A. (2020). Genomic Approaches to Drug Resistance in Malaria. *Annual review of microbiology*, *74*, 761–786. https://doi.org/10.1146/annurev-micro-012220-064343

Schmedes, S. E., Patel, D., Dhal, S., Kelley, J., Svigel, S. S., Dimbu, P. R., Adeothy, A. L., Kahunu, G. M., Nkoli, P. M., Beavogui, A. H., Kariuki, S., Mathanga, D. P., Koita, O., Ishengoma, D., Mohamad, A., Hawela, M., Moriarty, L. F., Samuels, A. M., Gutman, J., Plucinski, M. M., … Talundzic, E. (2021). Plasmodium falciparum kelch 13 Mutations, 9 Countries in Africa, 2014-2018. *Emerging infectious diseases*, *27*(7), 1902–1908. https://doi.org/10.3201/eid2707.203230

Serrano-Durán, R., López-Farfán, D., & Gómez-Díaz, E. (2022). Epigenetic and Epitranscriptomic Gene Regulation in *Plasmodium falciparum* and How We Can Use It against Malaria. *Genes*, *13*(10), 1734. https://doi.org/10.3390/genes13101734

Stewart, L. B., Freville, A., Voss, T. S., Baker, D. A., Awandare, G. A., & Conway, D. J. (2022). Plasmodium falciparum Sexual Commitment Rate Variation among Clinical Isolates and Diverse Laboratory-Adapted Lines. *Microbiology spectrum*, *10*(6), e0223422. https://doi.org/10.1128/spectrum.02234-22

Thakar J. (2024). Pillars of biology: Boolean modeling of gene-regulatory networks. *Journal of theoretical biology*, *578*, 111682. https://doi.org/10.1016/j.jtbi.2023.111682

Tripathi, J., Zhu, L., Nayak, S., Stoklasa, M., & Bozdech, Z. (2022). Stochastic expression of invasion genes in Plasmodium falciparum schizonts. *Nature communications*, *13*(1), 3004. https://doi.org/10.1038/s41467-022-30605-z

Walker, A., Ross, A., & Nsanzabana, C. (2025). Changes in the Prevalence of Antimalarial Partner Drug Resistance Markers and Policy in 6 Sub-Saharan African Countries From 2000 to 2021: A Systematic Review. *Open forum infectious diseases*, *12*(9), ofaf508. https://doi.org/10.1093/ofid/ofaf508

World Health Organization (2022). *World Malaria Report* 2022. Geneva: WHO.

World Health Organization (2024). Strategy to respond to anti-malaria drug resistance in Africa. *Malaria policy advisory group* meeting in Yaoundé, Cameroon.

World Health Organization (2025*). Question & Answer*: Artemisinin partial resistance.

Wyss, M., Kanyal, A., Niederwieser, I., Bartfai, R., & Voss, T. S. (2024). The *Plasmodium falciparum* histone methyltransferase PfSET10 is dispensable for the regulation of antigenic variation and gene expression in blood-stage parasites. *mSphere*, *9*(11), e0054624. https://doi.org/10.1128/msphere.00546-24

Zhou, R., Li, S., Ji, P., Ruan, S., Liu, Y., Yang, C., Qian, D., He, Z., Wang, D., Lu, D., Zhang, H., & Deng, Y. (2024). Prevalence of molecular markers of sulfadoxine-pyrimethamine resistance in Plasmodium falciparum isolates from West Africa during 2012-2022. *Scientific reports*, *14*(1), 26567. https://doi.org/10.1038/s41598-024-75828-w