DIHYDROFOLATE REDUCTASE GENE MUTATIONS IN *Plasmodium falciparum* AMONG SYMPTOMATIC PATIENTS IN OTA, OGUN STATE, NIGERIA

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A DISSERTATION SUBMITTED TO THE SCHOOL OF POSTGRADUATE STUDIES IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF THE MASTER OF SCIENCE (M.Sc) DEGREE IN MICROBIOLOGY IN THE DEPARTMENT OF BIOLOGICAL SCIENCES, COLLEGE OF SCIENCE AND TECHNOLOGY, COVENANT UNIVERSITY, OTA, OGUN STATE, NIGERIA

AUGUST, 2024

ACCEPTANCE

This is to attest that this dissertation is accepted in partial fulfillment of the requirements for the award of Master of Science (M.Sc) degree in Microbiology in the Department of Biological Sciences, College of Science and Technology, Covenant University, Ota, Ogun State, Nigeria.

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DECLARATION

I **MAMEH, EMMANUEL OJOCHEGBE (22PCQ02459)** declare that this research was carried out by me under the supervision of Prof. Solomon U. Oranusi of the Department of Biological Sciences, College of Science and Technology, Covenant University, Ota, Ogun State, Nigeria. I attest that the dissertations have not been presented wholly or partially for the award of any degree elsewhere. All sources of data and scholarly information used in this dissertation are duly acknowledged.

MAMEH, EMMANUEL OJOCHEGBE

Signature and Date

CERTIFICATION

We certify that this dissertation titled "**DIHYDROFOLATE REDUCTASE GENE MUTATIONS IN *Plasmodium falciparum* AMONG SYMPTOMATIC PATIENTS IN OTA, OGUN STATE, NIGERIA"** is an original research work carried out by **MAMEH, EMMANUEL OJOCHEGBE** (22PCQ02459) in the Department of Biological Sciences, College of Science and Technology, covenant University, Ota, Ogun State Nigeria under the supervision of Prof. Solomon U. Oranusi. We have examined this work and found this work acceptable as part of the requirements for the award of Master of Science in Microbiology.

Prof. Solomon U. Oranusi
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Prof. Anthonia O. Oluduro
(External Examiner) **Signature and Date**

Prof. Akan B. Williams
(Dean, School of Postgraduate Studies) **Signature and Date**

DEDICATION

This research work is dedicated to the Lord Almighty, the source of wisdom, knowledge and understanding. Then to my family for their moral and financial supports.

ACKNOWLEDGEMENTS

My sincere appreciation goes to Almighty God for preserving my life and for His eternal love without which this piece would not have been successful.

I'm quite grateful to Professor Solomon U. Oranusi who is my supervisor, and I appreciate him a great deal. He also happens to be the former HOD of Biological Sciences Department, for all his intellectual support and tremendous contributions especially for all the times he sacrificed right from the moment of choosing a topic, proofreading and correcting my research work, thereby making it a colossal success.

My appreciation also goes to Dr. Akinduti Paul Akinniyi, his direction and expertise in medical microbiology saw me through, and to all the staff members in the Department of Biological Sciences, Covenant University, Ota, Ogun State, Nigeria, for their invaluable contributions. May the lord bless you all.

Appreciation also goes to my sponsor "Covenant Applied Informatics, Communication Africa Centre of Excellence" (CApIC-ACE) team, and the World Bank for this rare privilege, providing me with financial stability and academic resources to advance my studies and contribute meaningfully to malaria research.

My hearty gratitude goes to my loving and ever-caring parents: Elder Sunday, Mameh (Ogah), and Late Mrs. Rhoda, Mameh who sacrificed their earnings to build a future for me. Their foresight, dedication, and immense contribution to my spiritual, moral, and academic training and development set me on the pace of self-discovery and self-development, your labour is not in vain. God will reward you abundantly.

I want to express heartfelt appreciation to my colleagues: Oyegbade Samuel Adeniyi, Balogun Daniel Oluwatobiloba, Aririguzoh Victoria-grace Onyekachi, Atokolo Austine, and Sule Queen Elizabeth, among others, whose contributions were indispensable throughout my postgraduate studies. Also, the entire Living Faith Commission for all immeasurable investment in my life, May God bless and reward each of you abundantly.

Thank you all.

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ABBREVIATIONS

WHO	World Health Organization
COVID-19	Coronavirus disease 2019
ACT	Artemisinin-based combination therapies
AQ-AS	Amodiaquine- artesunate
AL	Arthemether-lumefantrine
SP	Sulfadoxine-pyrimethamine
PF	*Plasmodium falciparum*
PFDHFR	*Plasmodium falciparum dihydrofolate reductase gene*
PFDHPS	*Plasmodium falciparum dihydropteroate synthase gene*
PFCRT	*Plasmodium falciparum chloroquine resistance transporter gene*
PFMDR-1	*Plasmodium falciparum multidrug resistance-1 gene*
ITBNs	Insecticide-treated bed nets
IRS	Indoor residual spray
NMEP	National malaria elimination programme
HIV	Human Immuno-deficiency Virus
AIDS	Acquired Immuno-deficiency Syndrome
HRP	Histidine-rich protein
PQ	Primaquine
PYR	Pyrimethamine
CQ	Chloroquine
QN	Quinine
MQ	Mefloquine
ATQ	Atovaquone
DHA	Dihydroartemisin
TCT	Triple combination therapies
NGS	Next generation sequencing
SNP	Single nucleotide polymorphism

RDT	Rapid diagnostic test
PCR	Polymerase chain reaction
SS	Sanger sequencing
WGS	Whole genome sequencing

ABSTRACT

Malaria significantly impacts global populations, especially in Sub-Saharan Africa, where *Plasmodium falciparum* predominates. The spread of drug-resistant parasites encoded with dihydrofolate reductase (*Pfdhfr*) gene complicates treatment, necessitating strategies for effective disease management and control. The study aimed to evaluate the prevalence of *Plasmodium falciparum* infection and patterns of dihydrofolate reductase (*Pfdhfr*) gene mutations among symptomatic patients in healthcare centers in Ota, Nigeria. Employing a cross-sectional study design with random sampling, blood samples were collected from a total of 300 symptomatic subjects diagnosed of malaria. Thin and thick blood films were prepared and examined microscopically for malaria parasite detection and speciation. Parasitemia was determined and *Plasmodium falciparum* was genotyped for *Pfdhfr* genes using standard methods. Genotyped *Pfdhfr* blood samples were sequenced and analysed for clonal diversity with global genotyped strains. Of the samples collected, 132 (44%) were positive for *P. falciparum* with a higher rate observed in males 71(52.2%) than females 61(37.2%). The age group 11-20 years had the significant high infection rates 58(68.2%) while population >60years had the least 5(20.8%) ($p<0.05$). Genotyped *Pfdhfr* among *Plasmodium falciparum* positive cases were 34.8% (46/132), with higher rates observed in 21-30 years (7.6%) and lowest in population >60 years (2.2%). *Pfdhfr* encoded strains from this study clustered with strains identified in Sudan, Nepal, China, and Nigeria. This study highlights the prevalence of *P. falciparum* with *Pfdhfr* gene mutations among the study population. There is need for continuous surveillance and targeted interventions to manage and control malaria, particularly among younger age groups.

Keywords: Plasmodium falciparum, Pfdhfr gene, Malaria, Prevalence, Parasitemia