

EISSN: 3043-6052

Vol 1, No 2: September, 2024

An open Access Peer-Reviewed Journal

Original Article

ALLELIC VARIANT-R OF THE PLASMODIUM FALCIPARUM MEROZOITE SURFACE PROTEIN 1 GENE AMONG CHILDREN POPULATION IN YENAGOA

Pughikumo DT.,¹ Pughikumo OC.,² & Donald AA¹

Affiliation: ¹Department of Human Physiology, Faculty of Basic Medical Sciences, Niger Delta University

²Department of Hematology, Faculty of Basic Clinical Sciences, Niger Delta University

Corresponding Author: Pughikumo DT, Department of Human Physiology, Faculty of Basic Medical Sciences, Niger Delta University

ABSTRACT

Merozoite surface proteins (MSPs) and their epidemiological relevance in research and healthcare for effective malaria diagnosis and control measures are garnering scientific endorsement with advancement in molecular biology. In this study, researchers investigated for an allele of MSPs designated as MSP1 among a cross-section of the population, with twenty-five patients aged between one and five years randomly selected.; among whom males accounted for 52% and females 48%. Blood samples were collected and standard protocols for determination of malaria parasites as well as genetic procedure for MSPs confirmation were performed in a reputable laboratory. 12% of patient had MSP1 –R, among whom 33% was female and 67% was male. It is informative that allelic variant MSP1 –R is present in malaria-infected blood sample of children population investigated.

How to cite: Pughikumo DT., Pughikumo OC., & Donald AA. Allelic Variant-R of the Plasmodium Falciparum Merozoite Surface Protein 1 Gene among Children Population in Yenagoa. *Global Professionals Multidisciplinary Practices Journal*. 2024, 1(2):102-109

Key words: Allelic variant – R, merozoite surface protein, age, sex, plasmodium falciparium

INTRODUCTION

The Merozoite surface protein 1 (MSP1) variant R is a specific allele of the MSP1 gene carried by a Plasmodium parasite, which causes an acute febrile illness (malaria) that are spread to people through the bites of infected female Anopheles mosquitoes. Malaria is a life-threatening disease primarily found in tropical countries. It is preventable and curable, (Bennink et al, 2016). However, without prompt diagnosis and effective treatment, a case of uncomplicated malaria can progress to a severe form of the disease, which is often fatal without treatment. Malaria is not contagious and cannot spread from one person to another. Five species of parasites can cause malaria in humans and 2 of species. Plasmodium these falciparum and Plasmodium vivax – pose the greatest threat. There are over 400 different species of Anopheles mosquitoes and around 40, known as vector species, can transmit the disease, (Bennink et al, 2016; Picot et al, 2020). The natural history of malaria involves cyclical infection of humans and female Anopheles mosquitoes. In humans, the parasites grow and multiply first in the liver cells and then in the red cells of the blood.

In the blood, successive broods of parasites grow inside the red cells and destroy them; releasing daughter parasites ("merozoites") that continue the cycle by invading other red cells. The blood stage parasites are those that cause the symptoms of malaria. The MSP1 is significant in understanding its evolution and potential impact on public health. MSP1 variants can affect various aspects of disease transmission. The objective of the study is to analyze the prevalence and distribution of MSP-1 variant-R among children aged 1-5yrs to enhance our understanding of malaria transmission dynamics and improve control measures, strategies for malaria control and understanding its association with drug resistance can guide the selection and deployment of antimalarial drugs.

METHODOLOGY

The research took place at Nucleometrix Laboratory, along Imiringi road, Yenagoa, Bayelsa, in which 25 samples were selected using random sampling technique; to collect blood samples from the patients at Tobi's clinic as cross section study.

Method used to carry out Field Staining

Preparation of Thin Blood Smear: A thin blood smear was prepared on a clean glass slide, by placing a drop of blood on one end of the slide, using another slide to spread the blood into a thin film, and allowed to air dry.

Fixation: Once the blood smear was dried, fixation is through one minute methanol emersion; helping to preserve cellular morphology, preventing cells from washing away during staining too.

Staining Solution Preparation: Field staining solution was prepared by diluting Field stain in buffered water or methanol.

Staining: The fixed blood smear slide was placed in a staining dish and covered with the prepared Field staining solution. Incubation of slide in the staining solution followed thereafter for 15 minutes.

Rinsing: After the staining period, the slide was carefully rinsed with tap water to remove excess stain.

Drying: Slide is allowed to air dry completely before examining it under a microscope.

Examination: Once the slide is dry, examination followed under a microscope using oil immersion at high magnification (100 x).

Malaria parasites appearing as purple-stained structures within the red blood cells, allowed identification and enumeration.

Interpretation: Staining pattern and morphology of the malaria parasites were interpreted according to established criteria for each species. Different species of Plasmodium may exhibit distinct morphological features under Field staining

Rapid Diagnostic Test: The standard procedure in malaria Rapid Diagnostic Test (RDT) was done using the test kit. Preparation, patient preparation, blood collection, and application of collected blood onto the designated area of the test cassette /strip were carried out in line with specific instructions provided in the RDT kit for the correct application technique. The buffer solution was added to the designated area of the test strip, facilitating the movement of the blood sample and reaction with test components. The test was allowed to develop according to the recommended incubation time, Interpretation of Results: After the incubation period, visual inspection of the test strip for the presence of colour lines or other indicators that signal a positive or negative result for malaria infection were done. Results interpreted according to the manufacturer's instructions.

Safe disposal of used lancets, bloodcontaminated materials, and any other waste generated during the testing process was done according to medical waste disposal guidelines **Documentation:** All test results were recorded in the patient's medical records, including the date, time, and interpretation of the test. Polymerase chain reaction was performed following standard protocol.

RESULTS

The general spread of RDT, MSP1-M, and MSP1-R

S/N Gender		Age in years	RDT	MSP1	MSP1
		•		M	R
1	F	4	+		
2	M	5	+	+	
3	M	5			
4	M	2	+		
5	F	4			
6	M	1	+	+	
7	F	2			
8	M	3	+		
9	M	4	+		
10	F	2		+	
11	F	5			+
12	F	1	+		
13	M	4			
14	M	4			+
15	M	2	+		
16	F	5			
17	M	2			
18	F	1			
19	F	2	+	+	
20	M	2		+	
21	F	1	+		
22	M	1			+
23	M	5			
24	F	3	+		
25	F	3	+		

RDT by age and sex

AGE	TOTAL NO. OF MALE INFECTED	TOTAL NO. OF MALE EXAMINED	OF	TOTAL NO. OF FEMALE EXAMINED	TOTAL NO. INFECTED	TOTAL NO. EXAMINED
1	1	2	2	3	3	5
2	2	4	1	3	3	7
3	1	1	2	2	3	3
4	1	3	1	2	2	5
5	1	3	0	2	1	5
TOTAL	6	13	6	12	12	25

MSP1-R by age and sex

AGE	TOTAL NO. OF MALE	TOTAL NO. OF MALE		TOTAL NO. OF	TOTAL NO. INFECTED	TOTAL NO. EXAMINED
	INFECTED	EXAMINED	FEMALE INFECTED	FEMALE EXAMINED		
1	1	2	0	3	1	5
2	0	4	0	3	0	7
3	0	1	0	2	0	3
4	1	3	0	2	1	5
5	0	3	1	2	1	5
TOTAL	2	13	1	12	3	25

MSP1-M by age and sex

AGE	TOTAL	TOTAL NO.		TOTAL NO.		TOTAL NO.
	NO. OF	OF MALE		OF	NO.	EXAMINED
	MALE	EXAMINED		FEMALE	INFECTED	
	INFECTED		INFECTED	EXAMINED		
1	1	2	0	3	1	5
2	1	4	2	3	3	7
3	0	1	0	2	0	3
4	0	3	0	2	0	5
5	1	3	0	2	1	5
TOTAL	3	13	2	12	5	25

Discussion

In the current study, twenty-five patients aged between one and five years were randomly selected. Among them, 52% were male female accounted for whereas 48%. Following blood samples collection and standard procedure for evaluating the presence of malaria parasites as well as genetic protocol for MSPs confirmation in a reputable laboratory; 12% of patient were diagnosed with MSP1 –R, among whom 33% was female and 67% was male. Also MSP1 -M was found in 25% of the population, wherein male was 60% and female 40%. It is informative that allelic variant MSP1 -R and MSP-M are present in malaria-infected blood sample of children population investigated, in a ratio appearing more in male; and as continuous progress is being achieved to understand malaria parasite, merozoites surface proteins and allelic variants, this may contribute to overall quest for vaccines and control of malaria

protein 1 (MSP1) variant R is a specific allele of the MSP1 gene carried by a Plasmodium parasite, which causes an acute febrile illness (malaria) that are spread to people through the bites of infected female Anopheles mosquitoes. Malaria is a life-threatening disease primarily found in tropical countries. It is preventable and curable.

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