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Glucose-6-Phosphate Dehydrogenase (G6PD) Deficiency among Children Diagnosed with Plasmodium falciparum Malaria Residing in the Rural and Urban Communities of Katsina State, Northern Nigeria

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Abstract:

About 96 million people out of 400 million having Glucose-6-phosphate dehydrogenase (G6PD) deficiency worldwide are known to reside in malaria endemic countries and this G6PD deficiency has been shown to protect against malaria infection, a disease which affects mostly children less than 5 years of age. G6PD deficiency is an X-linked genetic defect and is characterized by considerable biochemical and molecular heterogeneity. This study was prompted by the scarcity of scientific information on G6PD deficiency for malaria infected children in the rural areas of the state. G6PD deficiency was reported in Nigeria more than 48 years ago. Blood samples were collected from children with *Plasmodium falciparum* malaria attending the six selected hospitals located across the three senatorial zones of the state from June 2020 to December 2020. Children's inform consent was obtained, their socio-demographic information and clinical presentations were also taken with the aid of structured questionnaire. G6PD deficiency was detected qualitatively using G6PD screening test. The prevalence varies from 4.3 to 26.0 percent with an overall prevalence of 16.0 percent in different rural areas were significantly associated ($p<0.05$) with malaria. Since the children live in remote areas where malaria is/has been endemic, irrational use of antimalarial drugs could result in an increased number of cases with drug induced haemolysis. Therefore, before giving antimalarial therapy, routine screening for G6PD deficiency should be undertaken in those children from the rural communities where its prevalence is high.

Keywords: G6PD, *Plasmodium falciparum*, Malaria, Rural communities, Children and Katsina

1. INTRODUCTION

Glucose-6-phosphate dehydrogenase (G6PD) is an enzyme found in the cytoplasm of all cells catalyzing the first reaction in the pentose phosphate pathway, providing reducing power to all cells in the form of NADPH. NADPH enables cells to neutralize oxidative stress that can be activated by several oxidant agents, and to preserve the reduced form of glutathione. Since red blood cells do not contain mitochondria, the pentose phosphate pathway is their only source of NADPH; therefore, defence against oxidative damage is dependent on G6PD [1].

Glucose-6-Phosphate Dehydrogenase (G6PD) deficiency has been shown to protect against malaria infection which affect 241 million people worldwide with an estimate of 627 000 malaria deaths in 2020 of which 95% in

the African region and 80% were children under 5 years of age [2].

G6PD deficient individual has been linked with immunity against *Plasmodium falciparum* malaria [3, 4]. G6PD deficiency is one of the most frequent human enzyme deficiencies in the world. It is particularly frequent in populations living in malaria-endemic areas. The highest frequencies are examined in Africa, Asia, the Mediterranean region, and in the middle-east; owing to recent migrations, however, the abnormality is also found in North and South America and in northern European countries [5].

G6PD deficiency is one of the most prequent X-linked recessive hereditary genetic disorder caused by mutations in the G6PD gene, resulting in protein variants with many levels of enzyme activity, that are linked with a wide range of biochemical and clinical phenotypes. It acts on the erythrocyte metabolism, featuring non-immune haemolytic anaemia in response to a number of causes [6]. Patients with G6PD deficiency develops haemolytic anaemia during acute malaria infection and when treated with certain therapeutic agents such as anti-malarials, antipyretics and antibiotics which have oxidant properties. Increased oxidative stress in G6PD deficient cells is well recorded [7]. However, erythrocyte exposure to oxidative stress causes haemoglobin denaturation, ultimately resulting in haemolysis. Other clinical conditions include neonatal jaundice, which may result in neurological difficulties and death [8].

More than 176 mutations and 500 non-identical variants have been described to date for the G6PD gene; moreover, most are single base changes, leading to amino acid substitutions [9, 10]. The world health organization grouped the G6PD variants into five classes based on their enzyme activity and clinical embodiments, with class I demonstrating the severely deficient cases that are linked with chronic non-spherocytic haemolytic anaemia [11].

Malaria is a febrile sickness rooted by sporozoa of the genus *Plasmodium,* four species of which infect humans: *P. falciparum, P. vivax, P. ovale,* and *P. malariae.* The malaria parasites undergo a transformational cycle in the female anopheles mosquito, which is the vector. They are transfer to the human host following a bite by the mosquito, quickly enter the liver where they undergo a transformational phase of different duration among the four species (pre-erythrocytic phase), and then enter the red blood cell (intra-erythrocytic phase) where they continue their replication cycle. The asexual erythrocytic parasite is the stage in the life cycle that causes disease. This is characterized by fever, chills and sweats (which vary in regularly among the various species), anaemia, expansion of the liver and spleen [12].

The mechanism of *Plasmodium falciparum* malaria resistance in deficient individuals is likely linked to an impaired antioxidant defence in deficient ring-stage parasitized red cell evolving membrane damage, which activates a profuse removal of parasite by phagocytosis before it developed to trophozoite-stage parasitized red cell and to schizonts [12].

Regardless of the clinical and epidemiological importance of the interplay between G6PD deficiency and malaria, the magnitude of its incidence and significance has not been accurately measured. Examining and identification of G6PD deficient individuals will helps in curtailing such incidence, through proper selection of treatment, patient counseling, and avoidance from disease-precipitating drugs such as anti-malarial and other agents.

2. MATERIAL AND METHODS

2.1 Study Population

The study population was consisted of children aged ≤5 years admitted or presented with *Plasmodium falciparum* malaria cases to the selected hospitals. Two key hospitals were selected from each of the three senatorial districts of Katsina state. The hospitals selected for the study are; General Hospital Funtua and General Hospital Malumfashi (South), General Hospital Dutsin-ma and General Hospital Katsina (Central), General Hospital Baure and General Hospital Daura (North).

2.2 Ethical Approval

The ethical approval for this study was obtained from Katsina State Ministry of Health Ethical Research Committee that grants ethical clearance for research that involves human subjects (MOH/ADM/SUB/1152/1/276).

2.3 Sample Size

The sample size for this study was determined using the formula of [13] at 95% confidence level and a reported 15% prevalence of Glucose-6-phosphate dehydrogenase deficiency in Nigeria [13]. The calculated sample size is 195.9216; hence a total of 200 samples were used for the study.

2.4 Collection and Transport of Samples

Venous blood samples (2.0mls) were withdrawn from each child of the study population at the selected hospital by the laboratory technician and ethical guidelines was followed. All of the variables including age, sex, presence of anaemia, cause of anaemia, presence of jaundice, duration of jaundice, family history of G6PD deficiency and parent knowledge about G6PD deficiency were recorded based on a scheduled questionnaire. The samples were collected in EDTA (ethylene diamine tetra-acetic acid) tubes and transported immediately in ice-cooler box to the Laboratory Department of General Hospital Dutsin-ma for G6PD screening.

2.5 Biochemical Analysis of Samples

2.5.1 Detection of Plasmodium falciparum parasite using Rapid Test Device

The children were diagnosed using Biopanda Malaria Rapid Test device (Biopanda Reagents, United Kingdom) which is specifically and qualitatively detects *Plasmodium falciparum* (P.f.) antigen in human whole blood samples. This test applies lateral flow immuno-chromatography and is a tool to assist in the diagnosis of malaria.

3. RESULTS

3.1 General characteristics of the study subjects/population

During the study period, a total of 200 study population were recruited, of these, 119 (59.50%) were males and 81 (40.50%) were females. Moreover, 120 (60.00%) subjects come from the rural areas of the study population while 80 (40.00%) were from the urban areas. Majority of the children in study population were within the range of 0-12months 62 (31.00%) and decreases as the months increases i.e. 13-24, 25-36, 37-48 and 49-60months with the following numbers and percentages 49 (24.50%), 34 (17.00%), 30 (15.00%), and 25 (12.50%) respectively. However, the cause of anaemia in the study children were mainly as a result of taking certain food 179 (89.50%) while very few shows anaemia as a result of taking drug 21 (10.50%) and none of the children were shown to have crisis due to infection 0 (0.00%). Greater number 160 (80.00%) develops jaundice during anaemia and mostly last within 1-2weeks while the remaining 40 (20.00%) do not.

3.2 Prevalence of G6PD deficiency based on sampling area among children diagnosed with Plasmodium falciparum malaria in Katsina State, Nigeria

Figure 3.1 shows the distribution G6PD deficiency based on sampling area among children diagnosed with *P. falciparum* malaria in Katsina State, Nigeria. In this study, the prevalence of G6PD deficiency did not differ between the sampling area, (χ^2= 0.0127, d.f = 2, P=0.9937). The prevalence of G6PD deficiency was 34.30%, 34.30% and 31.40% for Katsina Central, Katsina North and Katsina South respectively.

2.5.2 G6PD Enzyme Detection using Test kits

In this work, the activity of G6PD enzyme was measured qualitatively using commercially available G6PD screening test (Biorapid Diagnostics Nig. Ltd.) according to manufacturer's instructions using fresh blood samples as enzyme activity reduces on refrigeration.

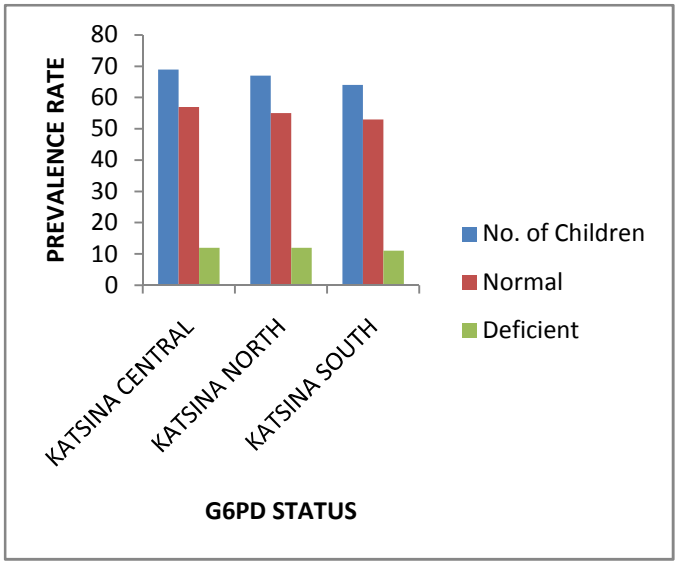

Figure 3.1 Distribution G6PD deficiency based on sampling area among children diagnosed with *Plasmodium falciparum* malaria in Katsina State, Nigeria

4. DISCUSSION

4.1 Distribution G6PD deficiency based on sampling area among children diagnosed with Plasmodium falciparum malaria in Katsina State, Nigeria

This research work shows that there was no interdependence statistically between G6PD deficiencies with either of the Senatorial zones i.e the sampling areas (Central, North and South senatorial zones). Therefore this shows that G6PD deficiency does not hang on the locality of the subjects within the state. Despite of the senatorial zone of origin a child may have the G6PD deficiency or not. The geographical distribution of G6PD deficiency proposes that some polymorphisms give out immunity to *Plasmodium falciparum* malaria [14]. This phenomenon has been studied mainly for the African variant (G6PD A-), showing that it also confers immunity against lethal falciparum malaria [15]. The higher prevalence of G6PD deficiency in malaria endemic countries is a sign that malaria infection has applied a giant selective pressure in many human populations [12,

16]. In *Plasmodium falciparum* infection it has been illustrated that shorter half-life and rapid clearance of red blood cells of G6PD deficient subjects make them less susceptible to malaria ambush from these parasites [17].

In western India, the largest frequency of G6PD deficiency was shown by the Parsis (17-19%). This large value may be due to the fact that Parsi is an endogamous group with a very laege degree of brotherhood. There was no deficiency recorded among Hindus and Christians of Bombay. Gujrati show scanty frequency of G6PD gene except for the Cutchi-Bhamushali group in which the rate is as large as 13.8%. North India comes up with a heterogeneous structure of enzyme deficiency (2% to 11.1%). G6PD deficiency of 6% from Agra and 11.1% from Allahabad was outlined [18]. In central Himalayas, Rajput of Baskat, U.P. and western Himalayas the frequency is not very high [19].

High Frequency of G6PD deficiency has been recorded among Angami Nagas of Nagaland (27.06%); 15.66 % [20]; santhal tribe 14.3% [21] and Rajhanski 11.65% [20] in the population group of east India. These reports show that the tribal populations present a higher prevalence of G6PD deficiency than the non-tribal population. Available reports show that the occurrence among Muslims of Madhya Pradesh is 3.9% while in Hindus is 7.5% [22]. Highest frequency (12.5%) is documented among Kurumbas of Nilgiri hills, Madras while Hindus of Hyderabad the rate lies between 9-12.5% in south India.

In UAE the frequency of G6PD deficiency in national populations is higher significantly (7.4%) unlike the non-nationals (3.8%) ($p < 0.001$) [23]. This reflects high occurrence of the deficiency in the region. Past research on G6PD deficiency between UAE national males living in Al-Ain have documented rates of 9.1 to 11% [24, 25] which is a little bit larger than the rate of 7.4% observed in the present research. The occurrence of G6PD deficiency in the UAE is lower than that in Kuwait (19%), Bahrain (21%), and Oman (27%) [26, 27].

However, the variation of G6PD deficiency within various geographical locations in Oman lies between 8.7 and 29% [28] while the frequency of G6PD in the UAE population is much larger than that in the populations living in some other Mediterranean countries like Italy (1 to 2%) [29], Spain (1%) [30], Turkey (1.2%) [31]. The occurrences of G6PD deficiency which have been documented all over the Eastern Mediterranean region lies between 3.6% in Jordan to 39.8% in Eastern Saudi Arabia [32].

One of the broadest review in India, shows that the large differences of G6PD deficiency has been observed ranging from 0% - 30.7% between the various caste, ethnic, and linguistic groups of India. The distribution in different areas reported rate of G6PD deficiency extending between 0% - 30.70% in Eastern India to 0% - 27.9% in Western India. The frequency of G6PD deficiency was reported extending from 0% - 23.21% in Northern India to 0% - 18% in Southern India. However, it was reported from 1.86% - 15.71% in North-eastern India and 0% - 19.23% and in Central India, the rate in the island regions of India was observed to be less [33].

4. CONCLUSION

The study shows that the prevalence of G6PD deficiency did not differ among the three senatorial zones (Katsina Central, Katsina North and Katsina South) and therefore, high percentages of G6PD deficiency prevalence in many malaria endemic areas account for considerable difficulties in effort to eradicate malaria. This highlights the need for comprehensive estimates of G6PD deficiency in malaria endemic regions and its clinical consequences. The most important way for prevention and reduction the incidence rate of clinical symptoms of G6PD deficiency is to avoid triggering agents like infection, Fava beans and oxidative drugs that induce haemolysis and also screening of newborns for early diagnosis of the deficiency.

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Competing Interest

The authors declare that there is no competing interest

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