DOI Number: 10.37506/v11/i1/2020/ijphrd/193829

Prevalence and Molecular Characterization of Glucose-6-Phosphate Dehydrogenase Deficiency among Brahmins and Muslims of Manipur, India

Nongthombam Achoubi[1], Mohammad Asghar[2], Anand Kumar Gyanendra Singh Wahengbam[3], Soibam Jibonkumar Singh[4], Kallur Nava Saraswathy[5], Benrithung Murry[5]

[1]Research Associate, Department of Anthropology, Manipur University, Manipur, [2]Assistant Professor, Department of Anthropology, Rajiv Gandhi University, Arunachal Pradesh, [3]Research Scholar, Department of Anthropology, University of Delhi, Delhi, [4]Professor, Department of Anthropology, Manipur University, Manipur, [5]Assistant Professor, Department of Anthropology, University of Delhi, Delhi, India

Abstract

G6PD deficiency offers protection against malaria infection and is strongly associated with the distribution of malaria endemicity. Genetic studies, including those of inherited blood disorders at the molecular levels, are very limited in northeastern India including Manipur. The present study aims to determine the prevalence and its molecular characterization of G6PD deficiency among the Brahmins and the Muslims of Manipur. A total of 263 unrelated blood samples (127 Brahmins and 136 Muslims) was screened for G6PD deficiency using Fluorescent Spot Test. DNA was extracted using salting out method and for molecular analysis, PCR was done using 3 most common Indian mutations (G6PD Mediterranean, G6PD Odisha, and G6PD Kerala & Kalyan) and 4 most common Southeast Asian mutations (G6PD Canton, G6PD Kaiping, G6PD Mahidol and G6PD Union). A higher prevalence frequency of 21.32% of G6PD deficient individuals was found among the Muslims as against the Brahmins with 9.45%. Out of the 7 mutations screened for G6PD that are common to Indian populations, only 4 Brahmins are found to have one each of these mutations. The 4 mutations found among the Brahmins were one G6PD Mediterranean, one G6PD Kerala & Kalyan, and two G6PD Mahidol. The Muslim population with a relatively higher frequency of G6PD deficiency as compared to Brahmin population needs special attention by health planners specifically while administering anti-malarial drugs.

Keywords: *G6PD, Meitei Brahmin, Meitei Muslim, Manipur.*

Introduction

Glucose-6-phosphate dehydrogenase (G6PD) deficiency is a common hereditary enzymatic defect worldwide. It is an X-linked inherited disorder most commonly affecting people of Africa, Asia, Mediterranean or Middle-Eastern descent[1]. It is mainly found in Africa, Asia, and Mediterranean Europe, areas where malaria is endemic, or has been endemic[2]. Other evidence supports the idea of a selective advantage of enzyme-deficient cells with regard to *Plasmodium falciparum*. Almost all cases of G6PD deficiency are caused by one amino-acid change due to a point mutation of the genomic DNA, and about 140 molecular abnormalities of the G6PD genotype have been identified[3]. The worldwide prevalence of G6PD deficiency has been estimated to be around 5%[4] but frequency as high as 70% has been reported among the Kurdish Jews[5]. In India, the

Corresponding Author:
Benrithung Murry
Assistant Professor, Department of Anthropology, University of Delhi, Delhi, India
e-mail: benrithungmurry@yahoo.co.in
Mobile: +91 9868616678

prevalence varies from 0 to 27.9% across ethnic groups[6]. The average prevalence of Malaria in Manipur is 1,995 per 1,00,000 populations. Rural inhabitants are almost twice as likely to suffer from malaria and the state has seen a substantial increase in malarial cases from 708 to 1,069 (51%) in the year 2009 and an increase in *Plasmodium falciparum* cases has been documented from 356 to 620 during the same period[7]. Only few studies report the prevalence of specific variants and fewer still have reported the prevalence of different G6PD variants at the DNA level in the north-eastern India. Genetic studies, including those of inherited blood disorders at the molecular levels are very limited in this region, including Manipur and such studies may help explain the origin and spread of these disorders and may also shed some light on the history of these ethnic groups. The present study aims to determine the prevalence and its molecular characterization of G6PD deficiency among the Brahmins and the Muslims of Manipur.

Material and Method

Brahmins belong to the larger Manipuri Meitei community of Manipur. Ethno-historically, the Brahmin settlers represent the eastern-most part of the so called Caucasoid types but consequent upon their inter-marriages with Meitei women over a long period of time they show both Caucasoid and Mongoloid features[8]. They inhabit the four valley districts of Manipur namely, Imphal-East, Imphal-West, Thoubal and Bishnupur. But the bulk of the Brahmins is scattered in BamonLeikai of Imphal-East. Muslims are a Caucasoid group, in contrast to the neighboring Mongoloid groups[9]. They spread throughout Manipur and practice consanguineous marriages. However, most of them inhabit the Imphal-East and Thoubal districts of Manipur. The Brahmin samples were collected from Imphal-East district and for Muslims, sampling was done from both Imphal-East and Thoubal districts of Manipur. For screening of G6PD deficiency, a total of 263 (only males) unrelated blood samples (127 Brahmins and 136 Muslims) were collected by finger prick after obtaining prior informed written consent. All the blood samples were tested G6PD deficiency using Fluorescent Spot Test[10]. Intravenous blood (5ml each) was collected from G6PD deficient individuals for further molecular analysis after obtaining prior informed written consent and DNA isolation was done following the standard Salting out method[11]. Molecular analysis was done by using the protocols given in table-1 below.

Results

Out of 127 Brahmin males screened for G6PD deficiency only 12 were found to be deficient with the frequency of 9.45% and of the 136 Muslim males, a higher frequency of 21.32% of G6PD deficient individuals (29) were found among them as shown in Table 2 below. The allele frequencies of G6PD deficiency of Meitei Brahmins (0.0945) and Meitei Muslims (0.2132) are the same as their percentile phenotype frequencies. A Chi square comparison of both the populations with respect to the allele frequencies showed that there is a significant difference between the studied populations ($\chi 2 = 7.0373$; $p= 0.007983$).Of the many mutations reported for G6PD deficiency only 3 most common Indian mutations and 4 most common Southeast Asian variants are selected for the present study. This is because both Brahmins and Muslims are expected to be admixture populations of India and South-East Asian ethnic elements.All the 41 G6PD deficient individuals were screened for the selected 7 mutations. Only 4 individuals out of the 12 Brahmin G6PD deficient could be characterized at the molecular level i.e. G6PD Mediterranean (8.33%) in one individual, G6PD Odisha (8.33%) in one individual and G6PD Mahidol (16.67%) in two individuals, whereas none of the 29 G6PD deficient Muslims could be characterized.

Table 1: Protocols used for molecular characterization of G6PD deficient individuals.

G6PD deficiency variant	Markers	References
Common Indian variants	G6PD Mediterranean (563 C→T)	Kaeda *et al*., 1995
	G6PD Odisha (131 C→G)	
	G6PD Kerala and Kalyan (131 C→G)	Ahluwalia *et al*., 1992
Common South-East Asian variants	G6PD Canton (1376 G→T)	Nuchprayoon*et al*., 2007
	G6PD Kaiping (1388 →A)	
	G6PD Mahidol (487 G→A)	
	G6PD Union (1360 C→T)	

Table 2: Distribution of G6PD deficiency among Brahmins and Muslims of Manipur.

Population	Total No. Tested	G6PD				P-value
		Normal		Deficient		
		No.	Percentile	No.	Percentile	
Meitei Brahmin	127	115	0.9055	12	0.0945	0.007
Meitei Muslim	136	107	0.7868	29	0.2132	

*significant at $p < 0.05$

Discussion

The G6PD deficiency prevalence frequency in India varies from 1% to 27% in different communities and regions of India[12]. In eastern India, the frequency is the highest in Angami Nagas (27.1%), followed by Adi (19.4%), Apatani (16.7%), Nishi (16%), Rabha (15.8%), Mikir (15.6%), Santhal (14.1%), etc.[13]. The distribution of G6PD deficiency among the Brahmins (9.45%) and Muslims (21.32%) of the present study is within the reported ranges of India. The reported frequencies of G6PD deficiency in India vary from complete absence among the Ganchha of Rajasthan[14], Lepchas of Assam[15], Dharwa, Halba and Maria of Madhya Pradesh, and Marathi of Maharashtra[16], Brahmin of Manipur and Jamatia of Tripura[17] to 0.279 among Vataliya Prajapati of Surat, Gujarat[18]. Very limited studies have been carried out in Manipur, the prevalence of G6PD deficiency among the Brahmins was reported to be 2.8%[19] and Kabui (7.8%)[20]. In the north-eastern region of India, the highest frequency (27.1%) has been reported among the Angami Nagas of Nagaland[21]. However, zero frequency has been reported among the Lepchas[15]. Among the populations of the eastern region of India, highest frequency (17%) of G6PD deficiency was reported among the Warli of Orissa[22] followed by Munda and Paraja having the same frequency of 15.9%[23]. The lowest frequency (3.6%) was reported among the Hindus of West Bengal[24]. The frequency of G6PD deficiency in Northern India ranges from 1.5% among the Rajputs of Himachal Pradesh[25] to 12% among the Tharu of North India[17]. The frequencies of G6PD deficiency were also reported among the Dhurwa, Halba and Maria of Madhya Pradesh[16]. The frequency of G6PD deficiency range of western India is 2.1% among the Rajputs of Dadra Nagar Haveli[26] to 27.9% among the Vataliya Prajapati of Surat[18].

Conclusion

The spectrum of mutations causing G6PD deficiency in India has not been well elucidated and several studies have revealed that the G6PD Mediterranean mutation is the most common variant followed by G6PD Kerala-Kalyan and G6PD Odisha[27].The prevalence of G6PD deficiency individuals were significantly high among Muslims as compared to that of Brahmins. However, though 4 common mutations were detected in the Brahmin population, all the 7 common mutations screened for in the populations were found to be absent among Muslim G6PD deficient individuals, suggesting a different origin and migrational history in these two groups which is in conformity to their historical records. Further, the Muslim population with a relatively higher frequency of G6PD deficiency are expected to have mutations that are not common to Indian populations. Hence, the Muslim population needs special attention and further in depth molecular research in respect to G6PD deficiency.

Conflict of Interest: No

Ethical Clearance and Consent: The present study is approved by the Ethical Committee of the Department of Anthropology, University of Delhi. Blood samples were collected after obtaining duly signed prior informed written consent from the participants.

Acknowledgements: The authors are thankful to the University Grants Commission, Delhi for funding this study.

References

1. WHO. Glucose-6-phosphate dehydrogenase deficiency. Bull. World Health Orga. 1989; 67, 601-611.
2. Tishkoff S.A., Varkonyi R., Cahinhinan N., Abbes S., Argyropoulos G., Destro-Bisol G., et al. Haplotype diversity and linkage disequilibrium at human G6PD: Recent origin of alleles that confer malarial resistance. Science. 2001; 293: 455-462.
3. Cappellini M, Fiorelli G. Glucose-6-phosphate

dehydrogenase deficiency. The Lancet. 2008; 371(9606):64-74.

4. Nkhoma E.T., Poole, C., Vannappagari, V., et al. The global prevalence of glucose-6-phosphate dehydrogenase deficiency: a systematic review and meta-analysis. – Blood. Cells. Mol. Dis. 2009; 42: 267-278.
5. Beutler, E. G6PD deficiency. – Blood. 1994; 84(11), 3613-3636.
6. Tripathy, V. & Reddy, B.M. Present status of understanding on the G6PD deficiency and natural selection. – J. Postgrad. Med. 2007; 53(3), 193-202.
7. Office of the Registrar General & Census Commissioner, India [Internet]. Censusindia.gov. in. 2019 [cited 10 April 2019]. Available from: http://www.censusindia.gov.in/
8. Chakraborty R, Walter H, Sauber P, Mukherjee B, Malhotra K, Banerjee S et al. Immunoglobulin (Gm and Km) allotypes in nine endogamous groups of West Bengal, India. Annals of Human Biology. 1987;14(2):155-167.
9. Shah, L. Bio-anthropology of the Muslim of Manipur. - Unpublished thesis, Manipur University, Imphal, Manipur.1990.
10. Beutler, E. & Mitchell, M. Special modification of the fluorescent screening method for glucose-6-phosphate dehydrogenase deficiency. – Blood. 1968; 32, 816–818.
11. Miller, S.A., Dykes, D.D. & Polysky, H.F. A simple salting out procedure for extracting DNA from human nucleated cells. – Nucl. Acids. Res. 1988; 16, 12-15.
12. Bhasin M. Genetics of Castes and Tribes of India: Glucose-6-Phosphate Dehydrogenase Deficiency and Abnormal Haemoglobins (HbS and HbE). International Journal of Human Genetics. 2006; 6(1):49-72.
13. Balgir R. Do tribal communities show an inverse relationship between sickle cell disorders and glucose-6-phosphate dehydrogenase deficiency in malaria endemic areas of Central-Eastern India?. HOMO. 2006; 57(2):163-176.
14. Choubisa, S.L. Erythrocyte glucose-6-phosphate dehydrogenase deficiency and thalassaemic genes in the scheduled castes of Rajasthan. - Indian Journal Medical Research. 1985; 82, 554–558.
15. Saha, N., Bhattacharyya, S.P., Mukhopadhyay, B., Bhattacharyya, S.K., Gupta, R. & Basu, A.A. Genetic study among the Lepchas of the Darjeeling area of eastern India. – Hum. Hered. 1987; 37, 113-121.
16. Das K, Roy M, Das M, Sahu P, Bhattacharya S, Malhotra K et al. Study of enzyme polymorphism and haemoglobin patterns amongst sixteen tribal populations of central India (Orissa, Madhya Pradesh, and Maharashtra). The Japanese Journal of Human Genetics. 1993;38(3):297-313.
17. Sarkar, S., Biswas, N.K., Dey, B., Mukhopadhyay, D. & Majumder, P.P. A large, systematic molecular-genetic study of G6PD in Indian populations identifies a new non-synonymous variant and supports recent infection. - Genetics and Evolution. 2010; 10.1228-1238.
18. Joshi, Sukumar, Patel, Colah, Patel. High prevalence of G6PD deficiency in Vataliya Prajapati community in western India. Haematologia. 2001;31(1):57-60.
19. Singh, K.S., Mukherjee, B.M., Walter, H., Lindenberg, P., Gilbert, K., Dannewitz, A., Malhotra, K.C., Banerjee, S., Roy, M. & Dey, B. Genetic markers among Meiteis and Brahmins of Manipur India. – Hum. Hered. 1986; 36, 177-187.
20. Achoubi N, Asghar M, Meitei S, Sachdeva M, Saraswathy K, Murry B. Haemoglobinopathies and glucose-6-phosphate dehydrogenase deficiency in a malaria endemic region of Manipur, northeast India. Anthropological Science. 2010;118(3):201-204.
21. Seth, P.K. & Seth, S. Biogenetical studies of Nagas: Glucose-6-phosphate dehydrogenase deficiency in Angami Nagas. – Hum. Biol. 1971; 3, 557-561.
22. Balgir R. The spectrum of haemoglobin variants in two scheduled tribes of Sundargarh district in north-western Orissa, India. Annals of Human Biology. 2005; 32(5):560-573.
23. Balgir R, Dash B, Murmu B. Blood Groups, Hemoglobinopathy and G-6-PD Deficiency Investigations Among Fifteen Major Scheduled Tribes of Orissa, India. The Anthropologist. 2004; 6(1):69-75.
24. Kotea R, Kaeda J, Yan S, SemFa N, Beesoon S, Jankee S et al. Three major G6PD-deficient polymorphic variants identified among the

Mauritian population. British Journal of Haematology. 1999;104(4):849-854.

25. Kabita S, Khurana P, Saraswathy K, Sachdeva M. Glucose-6-Phosphate Dehydrogenase Deficiency among the Rajputs and Brahmins of Solan District, Himachal Pradesh. The Anthropologist. 2011;13(1):39-41.
26. Devi N, Sachdeva M. Sickle Cell Haemoglobin and Glucose-6- Phosphate Dehydrogenase Deficiency among the Rajputs of Dadra and Nagar Haveli. The Anthropologist. 2009;11(1):45-47.
27. Mukherjee, M. B., Colah, R. B., Martin, S., & Ghosh, K. Glucose-6-phosphate dehydrogenase (G6PD) deficiency among tribal populations of India - Country scenario. The Indian journal of medical research. 2015; 141(5), 516-20.