Haemoglobin Variants among Voluntary Blood Donors in Jos, Nigeria: The Implications on Blood Transfusion

DAMULAK OD, BOLORUNDURO SA, EGESIE JO, YAKUBU K, GODIT P, SMITH OA

INTRODUCTION
The normal haemoglobin is an efficient transporter of oxygen to the tissues and carbondioxide from tissues to the lungs for elimination. Various abnormal haemoglobin variants including the sickle cell diseases, have been described with varying sickling tendencies.

AIMS. This study aimed to determine the haemoglobin variants among voluntary blood donors in Jos.

METHOD: Records of the age, sex, Haemoglobin level, and the haemoglobin genotype of all voluntary blood donors who donated blood at the National Blood Transfusion Service Centre, Jos, Nigeria between January 2011 and April 2012; and their haemoglobin levels and protein electrophoresis determined, were reviewed.

RESULTS: A total of 937 blood donors, 658 (70.23%) males and 279 (29.79%) females, mean age 32.4 years, donated blood voluntarily, their haemoglobin electrophoretic patterns determined by alkaline cellulose acetate electrophoresis. Donor blood haemoglobin levels were determined by automation. Haemoglobin protein electrophoretic patterns identified among our donors were 77.70% AA, 21.88% AS, 0.22% SC, 0.11% AC and 0.11% SS. Mean haemoglobin levels of the donors according to their haemoglobin proteins electrophoretic patterns were, 150.4 ± 12.5gms/l for AA, 151.9 ± 13.8gms/l for AS and 131.1 ± 5.0gms/l for haemoglobin SC.

CONCLUSION: Determination of haemoglobin protein electrophoretic patterns of blood unit for transfusion could enhance selective blood issuing based on recipient's haemoglobin type.

KEY WORDS: Haemoglobin Variants, Effective Transfusion.

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INTRODUCTION
The normal haemoglobin (HbAA) is an efficient transporter of oxygen to the tissues and carbondioxide from tissues to the lungs for elimination. Various abnormal haemoglobin variants have been described with varying sickling tendencies; the sickle cell diseases such as Hb SS, SC, SD and SB-Thal. Sickling in haemoglobin SC and SB-Thal could be more severe than can be in homozygous haemoglobin S (HbSS) though less frequent. The compound heterozygous sickle cell disease; Hb SD has the mildest disease and the red blood cells rarely sickle. Individuals with the sickle cell trait (Hb AS) do not sickle except under severe hypoxia as can occur when under general anaesthesia, flying in unpressurized planes, or post surgery.

The sickle haemoglobin in the deoxy form (deoxy HbS) polymerizes; a state cardinal for the formation of sickle erythrocytes and the clinical complications of the disease. The expression of the sickle gene is modulated by a number of pathophysiological mechanisms that generate a vast clinical diversity in the sickle cell patients. These mechanisms include genotypic variations, modification of polymerization of haemoglobin molecules, abnormal red blood cell hydration and membrane defects. The extent of haemoglobin polymerization is determined by the intracellular composition of Hb spaces, HbSS, HbAS, HbASn, HbSF, mean cell haemoglobin concentration (MCHC), oxygen saturation as well as PH and diphosphagycerate (DPG). Blood for transfusion to patients with abnormal haemoglobin might be ineffective if the units are also of abnormal haemoglobin combination such as SC, AS and SD that can sickle under hypoxic conditions. Researchers have described varying Hb phenotypes among various populations.

In Port Harcourt, Southern Nigeria, Zacchueus reported abnormal Hb variants among students of African descent. He found HbAA (80.32%) and HbAS (19.68%). Another study from the same region reported the prevalence of 70.00% HbAA, 29.44% AS, and 0.56% SS among pregnant women at first ante-natal care visit. A research report from the Niger Delta of Nigeria found more abnormal haemoglobin variants with 66% AA, 26% AS, 2% SS, 2% AC and 4% SC. In the Bonny area of Rivers State Nigeria, Hb AA was 73%, AS 22% while SS and AC were 4% and 1% respectively. A similar finding was reported from Uyo where Hb AA (78.7%), AS (19.6%), SS (1.5%), AC (0.2%) and SC (0.04%) were the haemoglobin variants indentified. Studies from South Western and Northern Nigeria show similar diversity of haemoglobin variants in their populations. A report from work done in Ogbomosho shows Hb AA constituting 68.1% while AS, AC, SS, SC and CC accounted for 21%, 5.7%, 3.0%, 2% and 0.3% respectively. Another report from Lagos Nigeria, is not

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different as 73.1% of their subjects were Hb AA, genotype AS 24.5% and SS 2.4%. A report from a study done in Keffi found 78.5% HbAA, 21.5% HbAS among the population. A similar research from Nekede in Northern Nigeria found only Hb AA and AS at the prevalence of 79% and 21% respectively with a significant Hb AS among the female population. A research report from a hospital base blood bank in Maiduguri found 78.94% HbAA, 21% AS and 0.06% SC.

A proper transfusion practice requires that component of blood for transfusion must fit purpose, hence the need to not only characterize the haemoglobin variants and their prevalence among our donors but as well have haemoglobin type of all blood units for transfusion determined.

METHOD
This retrospective study was carried out at the North Central Zonal Centre of the National Blood Transfusion Service, Jos, Nigeria. Records of the age, sex, haemoglobin level, and the haemoglobin genotype of all voluntary blood donors who donated blood to the centre between January 2009 and April 2012 were reviewed. The methods used for haemoglobin level and genotype determination were also studied. Appropriate statistical package was applied for result analysis. Results are presented in tables.

RESULTS
A total of 937 blood donors, 658 (70.22%) males and 279 (29.78%) females, mean age 32.4 years, donated blood voluntarily at the North Central Zonal Centre of the National Blood Transfusion Service, in Jos within the study period and had their Hb electrophoretic pattern determined. Donor blood haemoglobin levels were determined by automation using Haemocue Hb 301, Sweden, and protein electrophoretic pattern by alkaline cellulose acetate electrophoresis using Shandon 600000001 electrophoretic tank, Shandon Southern Products Ltd, England. Five haemoglobin protein electrophoretic patterns were identified among our blood donors (AA, AS, SC, AC and SS); 77.70% were AA, 21.88% AS, 0.22% SC, 0.11% AC and 0.11% SS. 153 (23.25%) of male donors and 55 (19.71%) of the female donors had sickle haemoglobin. Table 1.

<table>
<thead>
<tr>
<th>Hb variants</th>
<th>Male (%)</th>
<th>Female (%)</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>504 (53.79)</td>
<td>224 (23.91)</td>
<td>728 (77.70)</td>
</tr>
<tr>
<td>AS</td>
<td>152 (16.22)</td>
<td>53 (5.66)</td>
<td>205 (21.88)</td>
</tr>
<tr>
<td>SC</td>
<td>1 (0.11)</td>
<td>1 (0.11)</td>
<td>2 (0.22)</td>
</tr>
<tr>
<td>AC</td>
<td>1 (0.11)</td>
<td>0 (0.00)</td>
<td>1 (0.11)</td>
</tr>
<tr>
<td>SS</td>
<td>0 (0.00)</td>
<td>1 (0.11)</td>
<td>1 (0.11)</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>658 (70.22)</td>
<td>279 (29.78)</td>
<td>937 (100.00)</td>
</tr>
</tbody>
</table>

Sex distribution of Hb protein electrophoretic patterns of donors

This difference in the prevalence of sickle haemoglobin between male and female is not significant (P=0.2). The sex distribution of the haemoglobin proteins, male to female were; 53.79%: 23.91%, 16.22%: 5.66%, and 0.11%: 0.11% haemoglobin AA, AS and SC respectively. Abnormal haemoglobin proteins were 98.09% AS, 0.96% SC, 0.48% AC and 0.48% SS.

Distribution of donors with abnormal haemoglobin electrophoretic pattern

Mean haemoglobin levels of the blood donors according to their haemoglobin proteins electrophoretic patterns were, 150.4± 12.5g/l for AA, 151.9± 13.8 for AS and 131.1 ± 5.0g/l for haemoglobin SC donors. The haemoglobin level of the only AC blood donor was 159.0gms/l, while the haemoglobin SS who donated blood had a haemoglobin level of 138.0gms/l. There was no statistical significant difference between the haemoglobin level of AA compared to AS donors (P=0.1), AA compared to SC (P=0.2) and AS compared to SC (P=0.1). Table 3.

Comparative blood haemoglobin levels and electrophoretic patterns

DISCUSSION
Voluntary blood donation provides a platform for community service that allows prospective blood donors to freely donate blood for altruistic reasons. Sensitization on blood donation further enlightens individuals to self evaluate their current health conditions as well as risk exposure to contracting and transmitting transfusion transmissible infections. This evaluation empowers the individual to volunteer blood donation at no risk to self, or the prospective recipients. This sincere evaluation allows prospective donors with various haemoglobin type(s), random voluntary recruitment and donation.

We identified five haemoglobin combinations among our donors (AA, AS, SC, AC, and SS), table 1. The prevalence of 77.70% hemoglobin AA in our study is similar to the 80.32% reported by Zaccheaus and colleagues in Port Harcourt Nigeria. It is also similar to that reported in the Borny area of Rivers state and Uyo Nigeria, where Hb AA accounted for 73% and 78% respectively. The prevalence of Hb AA in our study is still similar to another finding in South Western Nigeria where a rate of 73.1% was documented in a Lagos
study. Our finding of 77.70% Hb AA concurred with the prevalence of 78.5% reported by Pennep in Keffi and 78.94% from a hospital based blood service in Maiduguri both in Northern Nigeria. It is however higher than the 70% reported among ante natal care patients in Port Harcourt and 66% reported by Egesie and others among undergraduate students in the Niger Delta of Nigeria.

Haemoglobin AS, found in 21.88% of voluntary blood donors in our study, is similar to 19.68% earlier reported by Zaccheus et al in Port Harcourt Nigeria and 21% reported in Ogbomosho, South Western Nigeria. It is also similar to the reports from work done in Northern Nigeria where Hb AS accounted for 21.5% in Keffi, 21% in both Nekede and the University of Maiduguri Teaching Hospital. Our finding is however lower than 29.4% Hb AS documented by Erhabor and others, among students in the Niger Delta of Nigeria and 26% reported among students of cell biology and genetics of the University of Lagos. Haemoglobin AS in our study was higher than the 8.08% and 7.75% reported by Fabritius and colleagues who systematically screened blood donors for haemoglobinopathies in Guadeloupe, French West Indies. These findings suggest both local and international variations in the prevalence of the sickle gene, which can be a mirror of the effectiveness of malaria control measures. The prevalence of 21.88% AS, 0.11% AC, 0.22% SC and 0.11% SS haemoglobin combinations in our study suggests a high potential of homozygous inheritance of the sickle haemoglobin gene which could be heightened further if stigmatization with inevitable selective mating is not controlled. There is need therefore to focus on, and commit more resources in Nigeria and Africa to the control of malaria, the stimulus for the evolution and persistence of the sickle haemoglobin. Further similar studies could measure the impact of such interventions. The mean Hb level of 150.4 gms/l and 151.9 gms/l of blood donors with Hb electrophoretic patterns AA and AS respectively show that adequate haemoglobin levels are attained in the presence of normal (Hb A) genotype.

Even though Pennap and his colleagues did not record any sickle cell disease haemoglobin electrophoretic patterns in their study in the same geopolitical region with ours, just as Omenka and others documented in Nekede, we found sickle cell disease patterns in 33%; 0.22% Hb SC and 0.11% SS among our blood donors with mean haemoglobin levels of 131.1 gms/l and 138.0 gms/l respectively. The prevalence of 0.11% Hb SS and 0.22% Hb SC in our study are similar to the 0.54% and 0.80% respectively reported by Akhigbe et al in their study of haemoglobin electrophoretic patterns among students of Ladoke Akintola University of Technology Ogbomosho Nigeria. These similarities may be due to the exclusion of students with known abnormal Hb electrophoretic pattern, which would include candidates with diagnosed sickle cell anaemia and other sickle cell diseases, from their study and blood donation in our blood service. It could also be due to the inability of many sickle cell disease children to successfully pursue their education to tertiary levels as a result of recurrent illness and childhood mortality in the face of poorly developed health sector of developing countries. The identification of these sickle cell disease Hb electrophoretic patterns in both homozygous (SS) and heterozygous (SC) states, with adequate Hb levels to meet haemoglobin requirement for blood donation, suggests that ineffective transfusion could be the end result of the transfusion involving such blood units. The homozygous sickle cell disease documented in our work may be due to the inability of the alkaline cellulose acetate electrophoretic method to separate haemoglobin D and or G from S. Stress occasioned by ill health, set the stage for the sickling of transfused blood from donors with sickle cell gene(s), subsequent destruction of such red blood cells and failure to correct anaemia. The determination of Hb proteins electrophoretic pattern should go beyond alkaline cellulose acetate method currently in use to more advanced methods like agarose gel electrophoresis, polyacrylamide gel electrophoresis and iso electric focusing that can separate the mobility of haemoglobin D and G from S. The further development of the blood service could allow for the collection of needed blood components other than red cell concentrate by apheresis or processing of collected whole blood from such donors with adequate blood haemoglobin level. In the absence of blood components preparation, blood donors with sickle cell disease Hb patterns should be permanently deferred.

CONCLUSION

We conclude from this study that blood donors with various types of Hb protein electrophoretic combinations could meet donor suitability criteria including heterozygous and rarely homozygous sickle cell diseases. The determination of the haemoglobin protein electrophoretic pattern of blood units for transfusion could enhance selective blood issuing based on the recipient's haemoglobin type. This study justifies the need to determine the haemoglobin type of all safe blood units, withdrawal of units with sickle cell disease electrophoretic patterns and permanent deferral of such whole blood donors except for components preparation.

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REFERENCE


