A Comparison of Measured and Predicted Haemoglobin Genotype in a Nigerian Population in Bonny, Rivers State, Nigeria

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ABSTRACT

Cellulose acetate electrophoresis technique has been used to estimate haemoglobin genotype and the predicted values from the estimate in accordance with the population genetics studies (Fleming and Lehman 1982) were evaluated in 850 subjects (386 males and 464 females) selected randomly from Bonny, in Rivers State, Nigeria of ages between 3 and 77 years. There was significant differences (P < 0.001) between the measured and the predicted haemoglobin genotype values suggesting that the predicted cannot be substituted for the measured. Two further haemoglobinopathies, HbSC and HbCC were
predicted to be the expected haemoglobin genotype of newborn into the population in the next decade suggesting that the incidence of abnormal haemoglobin in the population may arise from the complex interaction between the genetic constitution of the individual and some unidentified environmental factors. @ JASEM

The percentage of person with one of two types of haemoglobin genes – normal haemoglobin (HbAA) and abnormal haemoglobin (sickle cell trait, HbAS, sickle cell disease HbSS etc) in a community is an indication of Health awareness of its population. Knowledge and care of whether one is a carrier or a sickler or have normal haemoglobin is therefore the responsibility of the person and the entire community. Available reports indicate important differences in the distribution of haemoglobin genotypes between Caucasians and Africans (Schnelder et al 1976, Graham 1988, Reid and Famodu, 1988, Fleming and Lehman 1982). The most prominent feature is the prevalence of sickle cell trait/sickle cell disease among Africans than Caucasians and this correlates with falciparium malaria parasite transmission theory (Allison, 1964). There is however, a limited number of information of studies which described the distribution of haemoglobin genotypes among Nigerian population, in particularly, for a ethnic group in the swampy environment of the Niger Delta. The present report is, therefore, a study designed to find out the frequency of haemoglobin genotype in the population of Bonny ethnic group in the Niger Delta from which further information about the pattern of distribution of normal and abnormal haemoglobin genotypes for Nigerian may emerge. The Hardy-Weinbery law (Fleming and Lehman, 1982) has been used to predict the expected distribution of haemoglobin genotype of newborn into the population in the next decade.

MATERIALS AND METHODS

Subjects were selected randomly and drawn among Bonny ethnic groups located on an island south of Nigeria in the Niger Delta at the estuary of the Atlantic Ocean, and a former seaport during the slave trade era. Currently, it is one of the largest of oil exporting terminals in African and also the site of Nigeria liquefied Natural Gas Plants. A total of 850 subjects (386 males and 464 females) with age range of to 77 years were used in the determination of haemoglobin genotype.

Blood samples were collected by venopucture in adults and by finger or heel pricking in children into EDTA anti-coagulant bottles. The sickling test were carried out at a slightly acidic pH of 6.8 (Fleming and Lehman, 1982) to observe the presence of sickle cells under reduced oxygen tension. The electrophoretic method described by Fleming and Lehman (1982) and Graham (1988) was used as a
confirmatory test.

For the study of the haemoglobin electrophoresis, a small quantity of haemolysate of venous blood from each subject was placed on the cellulose membrane and carefully introduced into the electrophoretic tank containing Tris-EDTA-Borate suffer 89 as described by Fleming and Lehman, (1982). The electrophoresis was then allowed to run for 15 to 20 minutes at an emf of 160V. The results were read immediately. Hamolysates from blood samples of known haemoglobin (BDH & Sigma, London) were run as controls.

RESULTS AND DISCUSSION

The percentage of the various haemoglobin genotypes obtained in this study are shown in Table 1. Of the 850 subjects screened, 73% are HbAA, 22% HbAS and 4% are HbSS. From population genetics studies (Fleming and Lehman, 1982), if the incidence of sickle cell trait (HbAS) is known, the proportion of infants which would be born into the population with each haemoglobin genotype HbAA, HbAS, HbSS etc could be predicted. When the percentage genotype of the population are expressed as a proportion of 1.0 (Fleming and Lehman, 1982) the gene frequencies would be HbAA = 73, HbAS = 72, HbSS, = 4, HbAC = 1.

\[
\begin{align*}
\text{HbA} &= 73 + 11 + 0 + 0.5 = 84.5 \\
\text{HbS} &= 0 + 11 + 4 + 0.0 = 15.0 \\
\text{HbC} &= 0 + 0 + 0 + 0.5 = 0.5 \\
\end{align*}
\]

Given that a, s and c represent HbA, HbS and HbC respectively and further expressing them as a proportion of 1.0, gives HbA = a = 0.845, HbS = s = 0.15 and HbC = c = 0.005. And according to Hardy-Weinbrey law (Fleming and Lehman, 1982) the distribution of genotype of the newborn into the population in the next decade would be obtained as follows:

\[
(a + s + c)^2 = a^2 + 2as + s^2 + 2ac + 2sc + c^2 = 1.0
\]

Column 4 of table 1 shows the predicted values of the various haemoglobin genotypes and showed a further two haemoglobinopathies being introduced into the population as a result of gene recombination. The predicted HbAS values was significantly higher than the measured value. On the other hand, the measured valued for HbSS was significantly higher than the predicted value (P > 0.001).
### TABLE 1: PERCENTAGE DISTRIBUTION OF MEASURED AND PREDICTED HAEMOGLOBIN GENOTYPES IN THE POPULATION

<table>
<thead>
<tr>
<th>haemoglobin genotype</th>
<th>Number</th>
<th>Measured Value (%)</th>
<th>Predicted value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbAA</td>
<td>622</td>
<td>73</td>
<td>71.4</td>
</tr>
<tr>
<td>HbAS</td>
<td>190</td>
<td>22</td>
<td>25.35</td>
</tr>
<tr>
<td>HbSS</td>
<td>30</td>
<td>4</td>
<td>2.25</td>
</tr>
<tr>
<td>HbAC</td>
<td>8</td>
<td>1</td>
<td>0.85</td>
</tr>
<tr>
<td>HbSC</td>
<td>-</td>
<td>-</td>
<td>0.15</td>
</tr>
<tr>
<td>HbCC</td>
<td>-</td>
<td>-</td>
<td>0.0025</td>
</tr>
</tbody>
</table>

### TABLE 2. FREQUENCY OF Haemoglobin Genotype in Males and Females

<table>
<thead>
<tr>
<th>Sex</th>
<th>HbAA n</th>
<th>HbAA %</th>
<th>HbSS n</th>
<th>HbSS %</th>
<th>HbAS n</th>
<th>HbAS %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>277</td>
<td>44.53</td>
<td>91</td>
<td>47.89</td>
<td>14</td>
<td>46.7</td>
</tr>
<tr>
<td>Female</td>
<td>345</td>
<td>55.47</td>
<td>99</td>
<td>52.11</td>
<td>16</td>
<td>53.3</td>
</tr>
</tbody>
</table>
Table 2 shows the frequency of haemoglobin genotype in males and females. The percentage of females that are HbAA, HbAS and HbSS are more than the corresponding percentage of males. This represents differences of 44.3%, 17.0% and 26.5% respectively (P>0.001). The ratio of HbAA to HbAS was 3:1, HbAA to HbSS, 20:1 AND HbAA to HbAC 78:1. The ratio of HbAS to HbSS was 6:1, and HbAS to HbAC 24:1 and the ratio of HbSS to HbAC was 4:1.

From our study, the predicted values of the abnormal haemoglobin genotype sickle cell anaemia (HbSS) was less than the measured values with a mean difference of 121.5% and both measured and the predicted values were lower than the range 30 to 46% generally reported for Africans (Allison 1964, Richard 1975, Lewis 1970). In the U.S.A. 9% of the black population are HbSS (Richard, 1975). A possible explanation for this significant observed low incidence in HbSS might be attributed to environmental factors which include: Improved socio-economic conditions, better nutrition, blood type compatibility, absence and/or presence of malaria parasite infection, all of which tend to influence the importance that a person places on his health, and the degree of care that he may seek to protect it. Alternatively, the lower the socio-economic and the educational level and the difficulty of seeking medical advice and care of people of rural dwellers, to say, might influence the incidence of abnormal haemoglobin in the population. Furthermore there was significant differences (P < 001) between the predicted and the measured haemoglobin genotype. This might suggest that the predicted values might be are over or under simplification of the result. However, two further haemoglobinopethis HbSC and HbCC were predicted using the population genetic equation of (Fleming and Lehman 1982) to be the expected haemoglobin genotype of new born into the population in the next decade. This might suggest that the incidence of the abnormal haemoglobin in the population may arise from a complex interaction between the genetic constitution of the individual and some unidentified environmental factors.

The observed high incidence of HbAA and HbAS in the population, though the frequency of HbAA being significantly higher than that for HbAS, is in agreement with previous reports that the normal haemoglobin (HbAA), range from 55 to 75% (Fleming and Lehman, 1982), and the sickle cell trait (HbAS) 20 to 30% in Nigeria (Reid and Famodu, 1988) and 20 to 40% in Africa (Fleming and Lehman, 1982). The present study has for the first time also established the ratio for each of the haemoglobin genotypes.

The study also showed percentage distribution of persons for haemoglobin genotype by sex and found that there were significance difference between males and females in the distribution of HbAA, HbAS and HbSS (P<0.001). The difference might be a reflection that females generally are known to be home bound than
males, but the main reason for this disparity between the number of males and females examined was that men are less likely to be at home during the daytime and perhaps another reason may simply be increased frequency of clinic attendance by females compared to males.

It is noteworthy to highlight problems encountered and this was the belief and attitude of the people towards the use of blood. It was difficult to convince the people that their blood was not being used for ritual purposes rather for the determination of the state of their well being. Our experience suggests that mass literacy campaign was required to educate people on the need to donate blood as well have their blood examined because ignorance of the importance of this study (as well as proper matching, HIV/AIDS) could lead to genetic incompatibility in marriages involving individuals carrying the sickle cell trait an/or suffering from sickle cell anaemia and consequently a congenital abnormal offsprings.

REFERENCES


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