SERUM ANTIPHOSPHOLIPID ANTIBODIES AMONG HEALTHY ADULTS IN NORTHEASTERN NIGERIA

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ABSTRACT

Background: Antiphospholipid antibodies have been associated with variety of conditions. There is no standard health associated reference values required for the interpretation of antiphospholipid antibodies result available among adults in North- eastern Nigeria and Nigeria in general. The aim of this study is to determine the normal serum level (cutoff point) in healthy adult Nigerians. The need or otherwise for stratification of the reference values by sex would also be determined.

Methods: Seventy-six healthy adults' medical students and health workers were enrolled in the study over the period of twelve consecutive months. Blood samples were taken for quantification of antiphospholipid antibodies using Enzyme Linked Immunosorbent Assay (ELISA) technique. Apparently healthy adults age between 16-50years who are neither human immunodeficiency virus positive nor VDRL positive were included in the study. Reference values were calculated by parametric method.

Results: The mean serum concentration of IgG Antiα,GP1 for the sample population was 21.43 ± 9.43 U/ml. The mean serum concentration of IgG Antiα,GP1 for males was 18.6 ± 7.6 U/ml while for the female sample population was 22.7±10.2U/ml. There is no gender variation (P>0.05).

Conclusion: The reference value for serum antiphospholipid antibodies in healthy adults in Northeastern Nigeria was 21.43 ± 9.43 U/ml. The reference values should be used in this environment for both sexes. Extension of the scope of the study to cover the entire homogenous society of Northern Nigeria is recommended.

Key words:

INTRODUCTION

A reference value is a value obtained by observation or measurement of a particular type of quantity on a reference individual for the purpose of measurement. The International Federation of Clinical Chemistry demands that reference values be established apriori by a laboratory that serves the reference population. Antiphospholipid antibodies (APA) are a group of heterogenous circulating auto antibodies primarily directed against negatively charged phospholipids compounds. These antibodies share in common reactivity to phospholipids alone or bound to plasma protein co-factor (s) or to the co-factors themselves. The patients with antiphospholipid antibodies may present with a variety of conditions. These include autoimmune diseases such as lupus, lymphoproliferative disease and some forms of cancers. A wide range of neurological disorders like myasthenia gravis, seizures, migraine headaches and transient global amnesia have been associated with APA. Also, APA causes antiphospholipid syndrome (APS), which is a clinical syndrome that presents with abnormal clotting / clots in vein, arteries and capillaries, recurrent spontaneous abortion usually during the 2nd or 3rd trimester, thrombocytopenia, Livedo reticularis (a lacy, mottled appearance to the skin), falsepositive test for syphilis serology and persistently and substantially raised (>2units) circulating (IgG) anticardiolipin antibodies and / or the circulating lupus antibodies detected using prolongation of the activated partial thromboplastin time. Monitoring and management of these disease conditions demand that the concentration of APA of these patients be compared with reference values obtained from the same population. Therefore, the study aims at establishing apriori adult health associated reference values for adults in Northeastern Nigeria using University of Maiduguri Teaching Hospital Laboratory.

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DESIGN
Seventy-six healthy adults' medical students and health workers of University of Maiduuguri Teaching Hospital, in Northeastern Nigeria were recruited over twelve consecutive months after informed patient's consent.
Approval was obtained from the Ethical committee of the University of Maiduuguri Teaching Hospital. Individuals who had history of migraine headache, diarrhea within the last one month, positive VDRL test, HIV positive result, ingestion of drugs known to interfere with the result were excluded.
Blood sample was obtained into plain bottles for the estimation of antiphospholipid antibody using Enzyme linked immuno-sorbent assay [ELISA] technique. Samples were centrifuged immediately after clot retraction, aliquot was used to determine the HIV and VDRL status of the individual and the remaining sample stored at 20° C. Bindazyme MK 040 kit of the binding site Ltd, Birmingham, UK was used to estimate serum concentration of Human Anti α1 GPI IgG. Control sera for antiphospholipid antibodies were used to determine the quality of the assay and only the batch containing control sera results within control limits were accepted for the study.
All data were entered into computer and analyzed by SPSS and EPI info version 6.04. Data were analyzed by Analysis of variance ANOVA, Chi square test and student test were used to compare means.

RESULTS
Seventy-six subjects made up of 46 (60.5%) females and 30 (39.5%) males were studied. The average age was 26.37 ± 3.97 years. The female to male ratio was 1.5:1, with a mean age of 25.98±4.02 yrs and 26.79±3.88 years respectively. The age and sex distribution follow the normal (Gaussian) distribution curve.
The mean serum concentration was 21.43±9.4U/ml, median 20U/ml and with a range of 10-42.3U/ml at confidence interval of 95%. The group peak serum concentration was within the range of 11-20U/ml.
The mean serum concentrations of Anti-α2GPI IgG were 18.6 ± 7.6 and 22.7± 10.2U/ml in males and females respectively. Gender variation was not statistically significant with an F= 3.51, (df) 1 and p value of 0.065).

Table 1 Age and Gender distribution of subjects

<table>
<thead>
<tr>
<th>Age in years</th>
<th>Males</th>
<th>Females</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>16-25</td>
<td>9</td>
<td>25</td>
<td>34</td>
</tr>
<tr>
<td>26-35</td>
<td>19</td>
<td>17</td>
<td>36</td>
</tr>
<tr>
<td>36-45</td>
<td>2</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>30</strong></td>
<td><strong>46</strong></td>
<td><strong>76</strong></td>
</tr>
</tbody>
</table>

DISCUSSION
Antiphospholipids are a heterogeneous family of antibodies that react to negatively charged phospholipids or phospholipid-protein complexes and are associated with diverse clinical situations that cut across all sub specialty of medicine. Determining a reference values is certainly not a cost free operation particularly for different age and sex groups, which may explain the absence of reference values for APA in Northeastern Nigeria, a poor resource setting where there is no functional National Health Insurance Scheme. To the best of our knowledge, APA reference value has never been established by any laboratory in Nigeria, hence the dearth of information on the subject matter. To avoid possible seasonal variation, samples were obtained for a period of 12 months. Subject based reference values uses earlier result(s) from the same individual in certain state of health as a reference. Unlike subject based reference values, population based reference values may require stratification into demographic groups to improve its usefulness. There was no previous study of the normal value of antiphospholipid antibodies in this population for comparison.
The mean value of antiphospholipid antibodies was 21.43 ± 9.43 U/ml. Hence a cut-off point of < 21 U/ml was considered. The value is similar to the value estimated by the manufacturer, 20U/ml. The mean serum concentration was 18.57IU/mL for males and 22.04IU/mL for females, with (p = 0.065). Thus, the difference was not statistically significant and could not satisfy the stratification criteria of Harris and Boyd. Therefore, the established reference values should be used for both male and female adults in Northeastern Nigeria.

CONCLUSION

Antibodies occur in normal healthy adults with the mean concentration of 21.43U/mL and a reference range of 2.6-40.3u/mI is suggested. There is no gender variation in serum concentration of antibodies among healthy adults in North Eastern Nigeria. Extending the scope of the study to cover the homogenous society of Northern Nigeria is recommended.

REFERENCE


