**Assay kit for the assessment of Lp(a) levels in serum or plasma with minimum apo (a) size related bias**

Lp(a) is a complex macromolecule synthesized in the liver, it presents similarities with Low Density Lipoprotein (LDL) as it possesses a cholesterol-phospholipid core and a closely associated protein, apoB100. Lp(a) differs however from LDL in that each lipoprotein particle contains one copy of the glycoprotein apolipoprotein (a) [apo (a)] covalently bound to apoB100 by a single disulfide bond [1]. Studies indicate the association of elevated levels of plasma Lp(a) and risk of coronary heart disease [2,3]. A study reported that elevated plasma Lp(a) and small apolipoprotein (a) increased the risk of recurrent arterial ischemic stroke in children [4]. Moreover, the structural similarities between apo(a) and plasminogen highlighted the importance of Lp(a) in both atherosclerosis and thrombogenesis [5-7]. The intra and inter-individual size heterogeneity of apo(a) is genetically determined, this size variation constitutes a challenge for the immunochemical measurement of Lp(a) in plasma. The use of a 5-point calibrator which take into account the heterogeneity of Lp(a) for each of the levels would reduce result discrepancies. This study reports the performance characteristics of an immunoturbidimetric assay for the determination of Lp(a) in serum or plasma with minimum apo(a) size related bias.

**Methodology**

In this immunoturbidimetric assay agglutination occurs due to an antigen-antibody reaction between Lp(a) in a sample and anti-Lp(a) antibody adsorbed to latex particles. The agglutination is detected as an absorbance change at 700 nm proportional to the concentration of Lp(a) in the sample. The reagents are stable and ready to use. Lp(a) Calibrator Series, Lp(a) Control and Lipid Controls were used (Randox Laboratories Limited, Crumlin, UK). The assay is applicable to a variety of clinical chemistry analysers, for the results of this study the RX Daytona Plus analyser was used (Randox Laboratories Limited, Crumlin, UK).

**Results**

**Assay range**

This immunoturbidimetric assay presented a reportable range of 3 to 106 mg/dL.

**Sensitivity**

The Limit of Quantitation (LOQ), the Limit of Detection (LOD) and the Limit of Blank were determined consistent with CLSI guidelines EP17-A (table 1).

**Prozone**

Antigen excess effects were not noted until Lp(a) levels approached 493 mg/dL.

**Within run and total precision**

Within precision and total precision, as expressed as CV(%), were <4.00 and <4.5 respectively (Table 2).

**Serum/plasma comparison**

The assay was used to compare serum to plasma samples (n=56) collected into tubes containing Li heparin, Na EDTA, K EDTA or citrate. The data was subjected to linear regression analysis and in all cases the correlation coefficient (r) was > 0.996.

**Conclusion**

The immunoturbidimetric assay reported here for the determination of Lp(a) in serum/plasma with minimum apo (a) size related bias showed optimal analytical performance. The assay utilises ready-to-use stable reagents which facilitates the application in test settings by simplifying the experimental procedure and reducing handling errors. Its applicability to different automated analysers ensures the reliability, the accuracy of the measurements and facilitates the testing procedure. This represents an excellent analytical tool to facilitate clinical investigation.

**References**


Randox Laboratories Limited, Diamond Road, Crumlin, County Antrim, N. Ireland, BT29 4QY, UK
www.randox.com

---

**Table 1 Lp(a) immunoturbidimetric assay on the RX Daytona Plus: sensitivity.**

<table>
<thead>
<tr>
<th></th>
<th>Level 1</th>
<th>Level 2</th>
<th>Level 3</th>
<th>Level 1</th>
<th>Level 2</th>
<th>Level 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Limit of Quantitation (mg/dL)</td>
<td>3.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Limit of Detection (mg/dL)</td>
<td>2.57</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Limit of Blank (mg/dL)</td>
<td>0.45</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Table 2 Lp(a) immunoturbidimetric assay on the RX Daytona Plus: precision.**

<table>
<thead>
<tr>
<th>Mean (mg/dL)</th>
<th>Within run precision</th>
<th>Total precision</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Level 1</td>
<td>Level 2</td>
</tr>
<tr>
<td>SD</td>
<td>0.636</td>
<td>0.665</td>
</tr>
<tr>
<td>CV (%)</td>
<td>3.66</td>
<td>2.35</td>
</tr>
<tr>
<td>n</td>
<td>79</td>
<td>80</td>
</tr>
</tbody>
</table>