Antistreptolysin O (ASO) MonoReagent Procedure

**Intended Use**

In vitro diagnostic reagents for the quantitative determination of Antistreptolysin O (ASO) in human serum by means of particle-enhanced turbidimetric immunoassay.

**Background**

Immunological testing for specific antibodies to streptococcal metabolites provides important information regarding a prior streptococcal infection. Antibodies are formed against both the pathogen itself and its metabolic products. An example for the latter is the antibody against streptolysin O, an enzyme secreted by betahaeamolytic streptococci of the Landfield Group A. Antistreptolysin O (ASO) testing is thus used for the diagnosis of non supplicative complications of the infections caused by these pathogens: acute rheumatic fever or acute poststreptococcal glomerulonephritis. In the determination of antibodies to various streptococcal exoenzymes preference is to be given to anti-streptolysin O, since this sensitive parameter is found to be elevated in about 80 to 85% of cases.

**Test Principle**

The present ASO test is based upon the reactions between antibodies against streptolysin O (ASO) and latex particles bound streptolysin O. ASO values are determined photometrically.

**Reagents**

- **Buffer**
  Phosphate buffer, pH: 7,0, containing protein stabilizers and 0.09% sodium azide as preservative.
- **Latex reagent**
  polystyrene particles bound Streptolysin in a glycin buffer (0.1 M, pH: 8.2), containing NaCl (0.15M) and bovine serum albumin (0.5%). Preservative: Sodium azide 0.075%.
- **Calibrator**
  Human - based reference fluid. Preservative: sodium azide, 0.075%.

All raw materials of human origin must be handled with precaution. Sodium azide can form explosive azides with lead and copper plumbing.

**Precautions and Warnings**

For in vitro diagnostic use only. Do not pipette by mouth. Reagents containing sodium azide must be handled with precaution. Sodium azide can form explosive azides with lead and copper plumbing. Since absence of infectious agents cannot be proven, all specimens and reagents obtained from human blood should always be handled with precaution using established good laboratory practices.

**Material Required**

- Spectrophotometric analyzer.
- Saline solution.
- Controls.

**Storage and Stability**

Reagents in the original vial is stable to the expiration date on the vial label when capped and stored at (2 - 8°C). Immediately following the completion of an assay run, the reagent vial should be capped until next use in order to maximize curve stability. Once opened the reagent can be used within 1 month if stored tightly closed at (2 - 8°C) after use. Do not freeze reagents.

The ASO latex reagent should have a white, turbid appearance free of granular particulate. Visible agglutination or precipitation may be a sign of deterioration, and the reagent should be discarded.

The ASO buffer reagent should be clear and colourless. Any turbidity may be a sign of deterioration and reagent should be discarded. WR is stable for up to one month at 4°C. It is recommended that each Laboratory prepares a fresh Working Reagent based on its workload.

**Specimen Collection and Preparation**

Serum specimens should be collected by venipuncture following good laboratory practices. Suitable assay specimens are human serum samples, as fresh as possible (stored up to 2 days at 2 - 8 °C) or deep-frozen. Any additional clotting or precipitation which occurs due to the freeze/thaw cycle should be removed by centrifugation prior to assay. Heavily lipemic sera may lead to a non-specific reaction due to chylomicrons. Lipemic specimens, or turbid frozen specimens after thawing, must be clarified before the assay by high-speed centrifugation (15 min at approx. 15:000 rpm).

**Reagent Preparation**

Working Reagent is prepared with 1 part of Latex Reagent and 9 parts of Buffer Reagent. Prepare a fresh WR based on its workload. Shake gently the reagents before pipetting.

**Procedure**

<table>
<thead>
<tr>
<th>Calibrator</th>
<th>Sample</th>
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<tbody>
<tr>
<td>Calibrator</td>
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<td>-</td>
</tr>
<tr>
<td>Sample</td>
<td>5 μl</td>
<td>-</td>
</tr>
<tr>
<td>Distilled Water</td>
<td>500 μl</td>
<td>500 μl</td>
</tr>
<tr>
<td>Work. Reagent</td>
<td>500 μl</td>
<td>500 μl</td>
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</tbody>
</table>

Mix and measure absorbance immediately (A1) incubate 2 min (37°C), after incubation read absorbance (A2).

**Calculation**

Plot the calibration curve and the sample concentration is obtained by interpolation the sample absorbance (A2-A1) in the calibration curve. If an one point calibration:

\[
\frac{(A2-A1)_{\text{sample}} - (A2-A1)_{\text{blank}}}{(A2-A1)_{\text{calibrator}} - (A2-A1)_{\text{blank}}} \times \text{Calibrator concentration}
\]
Linearity

Up to 940 IU/mL

Calibration and Quality Control

| Calibrator 1 | 100 µl of Spectrum ASO Calibrator* |
| Calibrator 2 | 100 µl of Calibrator 1 + 100 µl of Saline Solution |
| Calibrator 3 | 100 µl of Calibrator 2 + 100 µl of Saline Solution |
| Calibrator 4 | 100 µl of Calibrator 3 + 100 µl of Saline Solution |
| Calibrator 5 | 100 µl of Saline Solution |

(*) See values on the label or on the insert. Multiply by the appropriate factor.

For quality control use Spectrum Control or other suitable control material. The control intervals and limits must be adapted to the individual laboratory requirements. Values obtained should fall within established limits. Each laboratory should establish corrective measures to be taken if values fall outside the limits. Control must be assayed and evaluated as for patient samples.

Expected Values

Each laboratory should establish an expected range for the geographical area in which it is located.

Values < 250 IU/ml are within the normal range. Children could have greater values.

References


ORDERING INFORMATION

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<thead>
<tr>
<th>CATALOG NO.</th>
<th>QUANTITY</th>
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