New HemosIL™

Screening Assays for the Diagnosis of Lupus Anticoagulant (LA)

Enhance LA Screening with a Complete Panel of Fully Automated Assays
Screening Assays for the Diagnosis of Lupus Anticoagulant (LA)

**Antiphospholipid Syndrome (APS)**

APS is an autoimmune disorder in which antiphospholipid (aPL) antibodies are associated with venous and arterial thrombosis, recurrent fetal loss, repeated unexplained spontaneous abortion or premature birth.

The laboratory criteria used to support the diagnosis of APS are based on the presence of LA, Anticardiolipin antibody (aCL) or anti-β2-Glycoprotein-I antibody.

LA is a class of auto-antibodies which may be directed toward different human proteins: β2-glycoprotein-I and Prothrombin primarily, but also Protein C, Protein S and Annexin V.

The in vitro effect of LA is based on the formation of complexes between the auto-antibodies and proteins. The formation of these complexes is enhanced in the presence of phospholipids.

For this reason, LA testing is mainly based on the prolongation of clotting times of different assays such as diluted PT and APTT, dRVVT or KCT.

These assays use very low phospholipid concentrations, which makes them sensitive to anti-phospholipid antibodies. Due to the heterogeneity of plasma auto-antibodies and the variable sensitivity of assays, the approach to LA diagnosis is based on multiple tests.

**LA Testing**

Laboratory diagnosis of LA is a complex procedure, based on the following criteria:

- Prolongation of phospholipid-dependent assays
- Exclusion of Factor deficiencies
- Phospholipid-dependence
- No effect from Factor inhibitors

To demonstrate phospholipid dependence, the most recent SSC-ISTH recommendations on LA require the use of two different phospholipid-dependent clotting assays, based on different methodologies.

**General Screening**

LA may be detected by performing general screening assays, such as PT and APTT. However, PT and APTT reagents are generally not designed to be sensitive to all types of LA, and because of the variety and heterogeneity of plasma anti-phospholipid antibodies, they cannot be used alone for the diagnosis of LA.

In addition, prolongation of clotting times above the normal range may have many causes, including the presence of Factor deficiencies, Factor inhibitors or anticoagulants. Samples demonstrating prolonged clotting time should be thoroughly assessed before proceeding with LA investigation.
Silica Clotting Time

- Screen (low phospholipid concentration) and Confirm (high phospholipid concentration) in the same kit
- Liquid formulation, easy to use, fully automated
- Suitable for mixing studies and oral anticoagulant-treated patient samples
- Sensitive to LA
- Sensitive to anti-β₂-Glycoprotein-I antibodies

LAC Screen and LAC Confirm

- Based on dRVVT, is the most common Screening and Confirmatory test for LA in the laboratory
- Easy to use, fully automated

SCT and LAC Screen/Confirm

- Cover the maximum spectrum of antiphospholipid antibodies
- Results for both are expressed as Normalized Ratio
- In accordance with the most recent SSC-ISTH recommendations on LA screening
- The combination of SCT and LAC Screen and Confirm are more informative and more likely to differentiate LA from anti-FVIII inhibitors, than either test alone
- The combination of SCT and LAC Screen and Confirm had the highest sensitivity in the detection of LA in patients who met the clinical criteria for APS
Screening Algorithm for LA with HemosIL Assays

The use of both SCT and LAC Screen and Confirm assays enhances the identification of LA patient samples, due to their different sensitivities to anti-phospholipid antibodies.

The charts below, demonstrate the major analytical steps in the diagnosis of LA.

**LA Investigation**

*N.R.: Normalized Ratio. See package insert for instructions on calculating N.R.*
## HemosIL Silica Clotting Time and LAC Screen and Confirm

<table>
<thead>
<tr>
<th>Part number</th>
<th>0020004800</th>
<th>0020008000 and 0020008200</th>
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<tbody>
<tr>
<td>Stability 2-8 C</td>
<td>20 Days</td>
<td>48 Hours</td>
</tr>
<tr>
<td>Stability on-board</td>
<td>5 Days</td>
<td>3 Days</td>
</tr>
<tr>
<td>Results</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normalized SCT Ratio</td>
<td>Screen Ratio = Confirm Ratio</td>
<td>Normalized LAC Ratio = Screen Ratio = Confirm Ratio</td>
</tr>
<tr>
<td>Precision</td>
<td>Mean (N. Ratio)</td>
<td>CV% Within run</td>
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<tr>
<td>Normal</td>
<td>~1.5</td>
<td>&lt;2.5</td>
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<tr>
<td>Low</td>
<td>~2.5</td>
<td>&lt;4.5</td>
</tr>
<tr>
<td>High</td>
<td>~3.5</td>
<td>&lt;5.5</td>
</tr>
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<td>Expected values</td>
<td>Normalized SCT Ratio</td>
<td>Normalized LAC Ratio</td>
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<tr>
<td>ACL™ Advance/Futura</td>
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<td>0.8 – 1.2</td>
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<tr>
<td>ACL 8/9/10000</td>
<td>0.75 – 1.20</td>
<td>0.8 – 1.2</td>
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In published clinical studies, HemosIL SCT demonstrated a very high sensitivity to Antiphospholipid antibodies in patients with Antiphospholipid Syndrome. The association of total thrombotic events, and specifically of venous thrombosis, with LA detected by the combination of SCT and dRVVT was significantly higher than with LA detected by the single tests alone or by the combination of APTT and dRVVT.(13)

### Summary
- HemosIL SCT was the most sensitive test with APS patients identified by the Sapporo criteria.
- When used in conjunction with the dRVVT, the sensitivity increased further with the APS population.
- Incorporating multiple assays, based on different methodologies, into the analytical test profile, increases the probability of detecting LA.
References

1. Horbach DA, Van Oort E, Donders RCJM, Derksen RHWM, De Groot PG. Lupus anticoagulant is the strongest risk factor for both venous and arterial thrombosis in patients


3. Galli M. Which antiphospholipid antibodies should be measured in the antiphospholipid syndrome? Haemostasis 2000; 30 (Suppl. 2) 57 - 62.


9. Dragoni F, Minotti C, Palumbo G, Faillace F, Redi R, Bongarzoni V, Arvisati G. As compared to kaolin clotting time, silica clotting time is a specific and sensitive method for


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Screening Assays for the Diagnosis of Lupus Anticoagulant (LA)

<table>
<thead>
<tr>
<th>Reagent</th>
<th>Part Number</th>
<th>Kit Configuration</th>
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<tbody>
<tr>
<td>Silica Clotting Time (SCT)</td>
<td>0020004800</td>
<td>SCT Screen 3 x 5 mL</td>
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<tr>
<td></td>
<td></td>
<td>SCT Confirm 3 x 5 mL</td>
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<tr>
<td></td>
<td></td>
<td>SCT CaCl2 3 x 10 mL</td>
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</tbody>
</table>

| LAC Screen                    | 0020008000        | 10 x 2 mL                       |
| LAC Confirm                   | 0020008200        | 10 x 2 mL                       |