Autoantibodies against phospholipids: state-of-the-art ELISA for diagnosis of the anti-phospholipid syndrome (APS)

Phospholipids are major components of cell membranes and play an important role in blood clotting. They are composed of phosphatidyl units (fatty acid–glycerol–phosphate), to which polar groups are coupled (e.g., serine, glycerol, choline). Antibodies against phospholipids (APLA) are directed against various antigens (e.g. phosphatidylserine, -inositol, -glycerol, -ethanolamine or -choline), and above all against cardiolipin.

Cardiolipin is a component of the inner mitochondrial membrane. Due to the high level of structural homology, almost all APLA are detected by assaying for anti-cardiolipin autoantibodies (ACA). Antibodies against other phospholipids are virtually never detected in ACA negative sera.

APLA can interfere with the balance between procoagulant and anticoagulant factors and thereby disrupt the physiological process of blood clotting. APLA are associated with a diverse range of clinical symptoms, collectively termed the anti-phospholipid syndrome (APS):

- Vascular thrombosis (arterial, venous, small vessels)
- Pregnancy complications (e.g. early or still birth, eclampsy)
- Lupus anticoagulant (LA)

APS occurs in isolation (primary APS) or in combination with other autoimmune diseases (secondary APS), most frequently systemic lupus erythematosus (SLE). In rare cases an accelerated and often fatal form of APS with multiple organ failure can develop (catastrophic APS).

According to diagnostic criteria defined in the 2004 international Consensus Workshop (Miyakis et al. 2006), Thromb. Haemost. 4:295), APS can be considered proven if at least one clinical and one serological criteria is met:

**APS criteria (Consensus Workshop 2004)**

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<th>Clinical criteria</th>
<th>Serological criteria</th>
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<td>Vascular thrombosis (arterial, venous, small vessels)</td>
<td>Antibodies against cardiolipin (IgG/IgM)</td>
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<tr>
<td>Pregnancy complications (e.g. early or still birth, eclampsy)</td>
<td>Antibodies against β2-glycoprotein (B2GP1) antibodies (IgG/IgM) or lupus anticoagulant (LA)</td>
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The determination of at least one of the following parameters is recommended for the serological diagnosis of APS: ACA (IgG/ IgM), Anti-β2-glycoprotein (B2GP1) antibodies (IgG/IgM) or lupus anticoagulant (LA). The antibody determination must be repeated after 12 weeks, since the serological APS criteria are only fulfilled after two positive results. By investigating ACA and anti-B2GP1 antibodies in parallel, the serological hit rate can be increased to 100%.

20-40% of SLE patients exhibit ACA, particularly when typical APS symptoms are present. ACA also occur in infections, for example lues or viral hepatitis, as well as in 1-5% of apparently healthy people. The prevalence of ACA in infectious diseases and blood donors is 4-20% of sera from patients with viral hepatitis and from blood donors reacted positively. The prevalences of ACA determined in SLE and APS correspond to data in current literature: With the state-of-the-art test systems from EUROIMMUN the detection of antibodies against cardiolipin and against β2-glycoprotein is of great significance for the diagnosis of APS.