Sindrome da anticorpi antifosfolipidi: la moderna gestione del paziente

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THE IDENTIFICATION of THE ANTIPHOSPHOLIPID SYNDROME

Avortments à répétition, thromboses et anticoagulant circulant antithromboplastin
Soulier, J.P., Boffa, M.C.

Thrombosis, abortion, cerebral disease, and the lupus anticoagulant
Hughes, G.R.V.

WITHIN OTHER AUTOIMMUNE DISEASES (OFTEN SLE)

PERIPHERAL VASCULAR SYNDROMES ASSOCIATED WITH SYSTEMIC LUPUS ERYTHEMATOSUS
ALARCON SEGOVIA D, OSMUNDSON PJ. L.
International consensus statement on an update of the preliminary classification criteria of the antiphospholipid syndrome

Clinical criteria

1. Vascular thrombosis

One or more clinical episodes of arterial, venous, or small vessel thrombosis, in any tissue or organ. Thrombosis must be confirmed by imaging or Doppler studies or hystopathology, with the exception of superficial venous thrombosis. For hystopathologic confirmation, thrombosis should be present without significant evidence of inflammation in the vessel wall.

2. Pregnancy morbidity

(a) One or more unexplained deaths in the second or third week of gestation, with normal fetal examination of the fetus, or

(b) One or more premature births before the 32nd week of gestation because of: (a) eclampsia, definitions, or (b) recognized feature

(c) Three or more unexplained consecutive pregnancies, with maternal anatomic or chromosomal causes excluded.

Laboratory Criteria

POSITIVITY MUST BE CONFIRMED AT LEAST 12 WEEKS APART

1. Lupus anticoagulant (LA) present in plasma, on two or more occasions at least 12 weeks apart, detected according to the guidelines of the International Society on Thrombosis and Hemostasis (Scientific Subcommittee on LAs/phospholipid-dependent antibodies).

2. Anticardiolipin (aCL) antibody of IgG and/or IgM isotype in serum or plasma, present in medium or high titer (i.e. >40 GPL or MPL, or >the 99th percentile), on two or more occasions, at least 12 weeks apart, measured by a standardized ELISA

3. Anti-b2 glycoprotein-I antibody of IgG and/or IgM isotype in serum or plasma (in titer >the 99th percentile), present on two or more occasions, at least 12 weeks apart, measured by a standardized ELISA, according to recommended procedures

Investigators are strongly advised to classify APS patients according to single or multiple positivity!

1-Diagnosi precoce:
• Interpretare gli anticorpi
• Riconoscere le manifestazioni cliniche

2-Terapia:
• Profilassi primaria
• Profilassi recidive
Non all the patients with aPL have thrombosis or pregnancy losses.

Many patients with thrombosis or miscarriage do not have aPL.

INTERNATIONAL CONSENSUS ON THE CONCEPT OF MULTIPLE POSITIVITY

2006

Update of the guidelines for lupus anticoagulant detection
Official communication of SSC

A LA result should be always be considered in the context of a full laboratory aPL profile (aCL and anti-b2GPI).
Medium high-titre of IgG aCL, anti-b2GPI and positive LA identify patients at high thrombosis risk.
Isolated LA positivity is more frequent in subjects without clinical events

2009

Investigators are strongly advised to classify APS patients in studies into one of the following categories:
Ia: Anti-cardiolipin antibody present alone
Ib: Lupus Anticoagulant present alone
Ic: Anti-Beta-2 glycoprotein-I antibody present alone
II: More than one Laboratory criteria present (any combination).

More positive tests more risk !!!

In experimental animals, the presence of antiphospholipid antibodies can enhance thrombus formation and cause pregnancy failure. Shoenfeld Y and Blank M 1992; Pierangeli S, Harris N, 1994-98.


**3.**

**Antiphospholipid Antibodies Are Pathogenic**

...BUT anti-β2GPI are detected in:

- Patients affected by systemic autoimmune diseases, without thrombotic events.
- Healthy adult individuals.
- Healthy pre-school children.

Anti β2GPI in healthy subjects

- n= 100 adults
- n= 13 1-3 yrs old children
- n= 17 >3 yrs old children
- n= 10 <1 yr old children

Graph showing IgG anti β2glycoprotein I levels for different age groups.
Anti β2GPI antibodies...are always the same?
Anti β2GPI antibodies and autoimmunity

Andreoli L, Chighizola C et al. Arthritis & Rheumatology 2015; 2196-2204
## ANTIPHOSPHOLIPID ANTIBODY PROFILE

<table>
<thead>
<tr>
<th>Isotype</th>
<th>Anti-cardiolipin antibody</th>
<th>Anti-B2GPI Antibody</th>
<th>Anti-Domain Antibody</th>
<th>Lupus Anticoagulant</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgG, IgM (IgA)</td>
<td>IgG, IgM (IgA)</td>
<td>Domain I/Domain IV-V</td>
<td>/</td>
<td></td>
</tr>
<tr>
<td>Titre</td>
<td>&gt;40 GPL/MPL</td>
<td>low, medium, high</td>
<td>low, medium, high</td>
<td>?+/-</td>
</tr>
<tr>
<td>Interpretation of isolated positivity</td>
<td>+</td>
<td>++</td>
<td>??</td>
<td>+++</td>
</tr>
</tbody>
</table>
217 prospectively followed pregnancies in patients with aPL

Averse pregnancy outcome occurred in 33 (15%) pregnancies:
14 (42%) in triple positive,
5 (15%) in double positive,
14 (42%) in single positive patients.

N.B.: 15 (45%) occurred in LA negative pregnancies.

<table>
<thead>
<tr>
<th>LABORATORY FEATURES (aPL PROFILE)</th>
<th>Adverse pregnancy outcome (n=33)</th>
<th>Favourable pregnancy outcome (n=184)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single aPL positivity</td>
<td>14/33 (42%)</td>
<td>98/184 (53%)</td>
</tr>
<tr>
<td>Double aPL positivity</td>
<td>5/33 (15%)</td>
<td>51/184 (28%)</td>
</tr>
<tr>
<td>Triple aPL positivity</td>
<td>14/33 (42%)</td>
<td>35/184 (19%)</td>
</tr>
</tbody>
</table>
Low-titre aCL/aβ(2)GPI positivity (>95(th) < 99(th) percentile) was considered positive for obstetric but not for thrombotic APS.

Twenty-six women with purely obstetric APS had persistent low-titre aCL and/or aβ(2)GPI.

Our data suggest that... **low-titre antibodies should be included in the diagnosis of obstetric APS.**

**Main and associated Clinical Features**

- Recurrent arterial/venous thrombosis
- Recurrent pregnancy loss
- Throbocytopenia
- Livaedo reticularis
- Leg ulcers
- Headache
- Chorea, epilepsy
- Cognitive disorders
- Heart valve lesions
- Haemolytic anemia
- Pulmonary hypertension

**Clinical features associated to aPL in SLE**

- Recurrent fetal loss
- Venous thrombosis
- Arterial thrombosis
  - Leg ulcers
  - Livaedo reticularis
  - Haemolytic anemia
  - Throbocytopenia

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GRV Hughes  BMJ 1988
GRV Hughes Lancet 1993

Antiphospholipid Syndrome
Clinical and Immunologic Manifestations and Patterns of Disease Expression in a Cohort of 1,000 Patients

The Euro-Phospholipid Project

<table>
<thead>
<tr>
<th>Clinical features at disease onset</th>
<th>No. (%) of patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deep vein thrombosis</td>
<td>317 (31.7)</td>
</tr>
<tr>
<td>Thrombocytopenia (&lt;100,000 plts/µl)</td>
<td>219 (21.9)</td>
</tr>
<tr>
<td>Livedo reticularis</td>
<td>204 (20.4)</td>
</tr>
<tr>
<td>Stroke</td>
<td>131 (13.1)</td>
</tr>
<tr>
<td>Superficial thrombophlebitis</td>
<td>91 (9.1)</td>
</tr>
<tr>
<td>Pulmonary embolism</td>
<td>90 (9.0)</td>
</tr>
<tr>
<td>Fetal loss</td>
<td>83 (8.3)</td>
</tr>
<tr>
<td>Transient ischemic attack</td>
<td>70 (7.0)</td>
</tr>
<tr>
<td>Hemolytic anemia</td>
<td>66 (6.6)</td>
</tr>
<tr>
<td>Skin ulcers</td>
<td>39 (3.9)</td>
</tr>
<tr>
<td>Epilepsy</td>
<td>34 (3.4)</td>
</tr>
<tr>
<td>Pseudovasculitic skin lesions</td>
<td>26 (2.6)</td>
</tr>
<tr>
<td>Myocardial infarction</td>
<td>28 (2.8)</td>
</tr>
<tr>
<td>Amaurosis fugax</td>
<td>28 (2.8)</td>
</tr>
<tr>
<td>Digital gangrene</td>
<td>19 (1.9)</td>
</tr>
</tbody>
</table>
Recommendation for future classification

- APS Nephropathy: Strongly recommended
- Valve hearth Lesions: Recommended
- Superficial venous thrombosis: Recommended
- Livedo reticularis: Recommended
- Thrombocytopenia: Recommended
- Chorea: Recommended
- Longitudinal myelitis: Recommended
- Seronegative APS: Recommended
- Seizures: Not recommended
- Migraine: Not recommended

Standard of care of well defines APS

(prophylaxis of recurrence)

Venous thrombosis: long term oral anticoagulation with target INR 2-3.
Arterial thrombosis: long term anticoagulation with target INR 3-4; antiplatelet alone; long term anticoagulation with target INR 2-3; long term anticoagulation with target INR 2-3 + antiplatelet.
Obstetric APS: LDA + LMWH

aPL Carriers
Primary prophylaxis of thrombosis
Primary prophylaxis of pregnancy loss

Absence of aPL
Seronegative APS
Classical APS that become aPL negative after time and treatments

Refractory APS
Thrombosis recurrence in standard treatment
Pregnancy loss in standard treatment

Reports on Thrombosis Recurrence

95 patients with primary APS
Mean follow-up time 4.5 years (0.3-26)

Breslow test 4.7, p=0.02

177 patients (56% primary) with vascular APS
Median follow-up time 6.5 years (1-27)

Log-rank test (p < 0.001).


Longterm Outcome of Patients with Primary Antiphospholipid Syndrome

84 PAPS patients with thrombotic history, median follow-up time: 17.66 yrs (range 15-30)

Median free time from events:
- in oral anticoagulant (OA) 221 months
- no oral anticoagulant (NO) 198 months

24 severe bleeding episodes were observed in 18 patients, all treated with oral AC (23%), 4 having more than 1 bleeding episode. Genital tract (metrorrhagia) was the most frequent affected site (29%), followed by cerebral (23%), gastro-intestinal (GI; 17%), ENT (14%), etc.

The new oral anticoagulants

The reported safety of oral anticoagulants combined with an increase in patients’ wellbeing (reduced need of monitoring) are important arguments in favour of these drugs in the secondary prevention of venous thromboembolism.

-Urbanus R. Rivaroxaban to treat thrombotic antiphospholipid syndrome. Lancet Haematology 2016; 3 September
The new oral anticoagulants: clinical studies in APS


26 pts with thrombotic APS (11 primary); 11 with arterial events.
11 pts in dabigatran
15 pts in rivaroxaban

Discontinuation of the treatment in 4 pts (15%) after a median follow-up of 19 months; one patient had relapse of arterial thrombosis, two developed bleeding events, and one recurrent migraine.

Rivaroxaban v.s. warfarin in pts. with thrombotic APS, with or without SLE: a randomised, controlled, open-label, phase 2/3, non-inferiority trial.
Available data: 56 pts in warfarin (INR 2.5) and 54 patients rivaroxaban (20 mg daily).

At day 42:
thrombin generation (assessment of the anticoagulant effects of warfarin and rivaroxaban).
The overall thrombogram indicated no increase in thrombotic risk with rivaroxaban.

At day 210:

**Study limitations:**
- small number of pts included
- <20% triple-positive aPL
- excluded pts with arterial thrombosis or recurrence in warfarin
- too short clinical follow-up

-Cohen H et al. Rivaroxaban versus warfarin to treat patients with thrombotic antiphospholipid syndrome, with or without systemic lupus erythematosus (RAPS): a randomised, controlled, open-label, phase 2/3, non-inferiority trial. Lancet Haematology 2016; Vol. 3 September.

The new oral anticoagulants: Ongoing clinical trials in APS

(1) Rivaroxaban in APS Pilot Trial—a Multicenter Feasibility Study of Rivaroxaban for Patients with APS and Prior Arterial or Venous Thrombosis (ClinicalTrials.gov Identifier: NCT02116036);

(2) TRAPS—Rivaroxaban in Thrombotic APS Trial Update—a Prospective, Randomized Clinical Trial Comparing Rivaroxaban vs Warfarin in High Risk Patients With APS (ClinicalTrials.gov Identifier: NCT02157272);

(3) ASTRO APS—Apixaban for the Secondary Prevention of Thromboembolism: A Prospective Randomized Outcome Pilot Study Among Patients with APS (ClinicalTrials.gov Identifier: NCT02295475).
Treatment of «non-criteria» clinical manifestations of APS

<table>
<thead>
<tr>
<th>Manifestations</th>
<th>Treatment</th>
<th>Evidence</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hematological manifestations</strong></td>
<td>Thrombocytopenia and hemolytic anemia: Steroids, IVIG, immunosuppressant,</td>
<td>Case series and 1 open label study</td>
</tr>
<tr>
<td>- Thrombocytopenia</td>
<td>rituximab, splenectomy. Primary thromboprophilaxis (HCQ, LDA) in case of</td>
<td></td>
</tr>
<tr>
<td>- Hemolytic anemia</td>
<td>significant serology.</td>
<td></td>
</tr>
<tr>
<td><strong>Neurologic manifestations</strong></td>
<td>Chorea, myelitis, multiple sclerosis-like disease: Steroids and</td>
<td>Case series</td>
</tr>
<tr>
<td>- Chorea</td>
<td>immunosuppressive agents (&gt;in SLE-APS) + anticoagulat (&gt;in patients with</td>
<td></td>
</tr>
<tr>
<td>- Myelitis</td>
<td>multiple sclerosis like disease and myelitis). Antidopaminergic drugs or</td>
<td></td>
</tr>
<tr>
<td>- Multiple sclerosis-like disease</td>
<td>dopamine-depleting agents in chorea.</td>
<td></td>
</tr>
<tr>
<td><strong>Heart valve disease</strong></td>
<td>Oral anticoagulant for symptomatic patients</td>
<td>Case series</td>
</tr>
<tr>
<td></td>
<td>Careful monitoring of anticoagulation for heart valve surgery</td>
<td></td>
</tr>
<tr>
<td><strong>aPL-associated nephropathy</strong></td>
<td>SLE nephritis: HCQ and/or antiplatelet/anticoagulant</td>
<td>Non randomized controlled studies</td>
</tr>
<tr>
<td></td>
<td>Isolated aPL-nephropathy: antiplatelet/anticoagulant</td>
<td>Case series</td>
</tr>
<tr>
<td></td>
<td>All the patients: Angitensin-converting enzyme inhibitors and angitensin-</td>
<td>Clinical judgement</td>
</tr>
<tr>
<td></td>
<td>receptor blockers</td>
<td></td>
</tr>
</tbody>
</table>
HYDROXYCHLOROQUINE (HCQ) and APS

Synthetic antimalarial drug with immunosuppressive properties in systemic autoimmune diseases

- Antiplatelet
- Hypoglycemic
- Cholesterol metabolism

Block TNF-α
- Thrombosis
- Survival
- Binding of anti-β2GPI to phospholipid bilayers

↓ Activation TLR 3, 7 e 9
↓ Proinflammatory cytokines


EFFECTS ON aPL IN PATIENTS WITH SLE

↓ aPL titres

Broder A, J Rheumatol 2013

↓ binding of anti-β2GPI to phospholipid bilayers

Rand JH, Blood 2008

What do we see in patients with primary APS?
**114 patients (from Brescia, Milan and Padua)**
Retrospective, propensity score-matched cohort study from 1992 to 2016 with mean follow-up of 76 months (±48 SD).

**Inclusion criteria**
- APS classification according to the 2006 criteria by Miyakis et al.
- Treatment with HCQ for at least 12 consecutive months

**Exclusion criteria**
- Treatment with other immunosuppressive drugs
- Concomitant systemic autoimmune disease

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**HCQ EXPOSED**
- 57 pts.
- 45% SINGLE aPL POSITIVE
- 41% DOUBLE aPL POSITIVE
- 14% TRIPLE aPL POSITIVE

**HCQ NOT EXPOSED**
- 57 pts.
- 40% SINGLE aPL POSITIVE
- 49% DOUBLE aPL POSITIVE
- 11% TRIPLE aPL POSITIVE

aPL TITERS: START vs. END OF FOLLOW-UP

**HCQ EXPOSED**

**HCQ NOT EXPOSED**

Level of aPL positivity
-negative: blank or white, low:
-gray, medium–high
titer: dark gray or black) at the beginning and at the end of the follow-up.

## THROMBOTIC EVENTS DURING THE FOLLOW-UP

<table>
<thead>
<tr>
<th>Group of patients</th>
<th>Site of thrombotic onset</th>
<th>Site of first thrombotic recurrence</th>
<th>Therapy at the time of recurrence</th>
<th>Trigger</th>
<th>aPL profile</th>
<th>Other cardiovascular risk factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arterial (MI)</td>
<td>Venous (DVT)</td>
<td>LMWH+HCQ</td>
<td>Puerperium</td>
<td>Double positivity</td>
<td>Smoking, inherited thrombophilia</td>
<td></td>
</tr>
<tr>
<td>Arterial (stroke)</td>
<td>Venous (porta)</td>
<td>HCQ</td>
<td>Not identified</td>
<td>Single positivity</td>
<td>Arterial Hypertension, inherited thrombophilia</td>
<td></td>
</tr>
<tr>
<td>Venous (DVT)</td>
<td>Small vessels (MI)</td>
<td>OA+HCQ</td>
<td>No identified</td>
<td>Triple positivity</td>
<td>Inherited thrombophilia</td>
<td></td>
</tr>
<tr>
<td>Venous (DVT)</td>
<td>Small vessels (CAPS)*</td>
<td>OA+HCQ</td>
<td>Infection</td>
<td>Double positivity</td>
<td>Arterial Hypertension</td>
<td></td>
</tr>
<tr>
<td>Obstetrical onset</td>
<td>Arterial (stroke)</td>
<td>HCQ</td>
<td>Not identified</td>
<td>Single positivity</td>
<td>Arterial Hypertension, inherited thrombophilia, Hypercholesterolemia</td>
<td></td>
</tr>
</tbody>
</table>

### HCQ exposed
7 events

### HCQ not exposed
7 events

THROMBOTIC EVENTS DURING THE FOLLOW-UP

HCQ exposed

1,16% 3,19-4,4% 1,71%

HCQ not exposed

Annual incidence thrombotic recurrence

~0% 1-3% 1,14%

Annual incidence of arterial recurrence


Rituximab and APS


B cell inhibition may have a role in difficult-to-treat APS patients, possibly in those with hematologic and microthrombotic/microangiopathic manifestations.


Combine treatment that includes anticoagulation with:
- heparin,
- high dose steroids,
- plasma exchange and/or intra-venous immunoglobulins

In refractory patients: rituximab and eculizumab are good alternatives.

Eculizumab, a humanized monoclonal antibody against complement protein C5, is currently approved for the treatment of paroxysmal nocturnal hemoglobinuria. It is able to reduce intravascular hemolysis and control complement mediated damage.
**FUTURE TREATMENTS**

**Novel therapeutic targets**

Antibodies against **D1 of β2GPI** have been shown to be pathogenic in animal models and have been found associated with obstetrical APS in humans. Tolerogenic dendritic cells specific for β2GPI D1, lowering antibody titre, were able to lower the rate of fetal loss.

Andreoli L. et al Arthritis Rheum. 2015  

Synthetic **peptide TIFI**, that mimic the phospholipid binding site of β2GPI (domain 5) was shown able to abrogate the aPL mediated angiogenesis inhibition at endometrial level.


**Toll like receptor 4** was shown able to mediate the aPL impairment of trophoblast fusion and differentiation. HCQ could to reduce Toll lik receptor 4 mRNA and protein expression and to restore trophoblast function.

Marchetti T. et al., J Thromb Haemost. 2014

Revised in: Ostensen M. Autoimmunity Reviews 2015
1-Diagnosi precoce:
• Stratificazione del rischio!
• Riconoscere non soltanto trombosi e patologia della gravidanza!

2-Terapia:
• Non solo anticoagulanti!
Grazie a tutti per la attenzione !!!

Brescia Rheumatology Unit