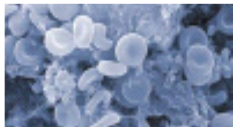


Dr Imad Aboukhamis
Ph-D France

- Antiphospholipid Syndrome
- متلازمة الفوسفوليبيد

Main clinical features

Thrombotic eventsarterial, venous or small vessel thrombosis in any tissue or organ

يحدث تختثروريدي أو شرياني في أي نسيج أو عضو

Recurrent pregnancy loss fetal death, premature birth, spontaneous abortion

يحدث فقدان حمل متكرر،وفاة الجنين والولادة المبكرة والإجهاض التلقائي

Thrombocytopenia a decrease in the number of blood platelets

نقص الصفيحات الدموية

Epidemiology

Sex ratio	pAPS	(F:M) 5:1
	sAPS	(F:M) 3:1
Age at onset	34 years +/- 13	
Diagnosis	42 years +/- 14	
APS categories	53 % primary APS	
	47 % disease associated APS	
		36 % SLE
		5 % SLE-like
		6 % other AID

Prevalence and risk groups
الانتشار و مجموعات الخطورة

- primary APS (PAPS) (53%)
- secondary APS (47%)
  - 37% associated with SLE or SLE-like syndrome
- Females are more frequently affected than males
  - يتكرر عند الإناث أكثر من الذكور
  - mainly second and third decades of life
  - يصيب الافراد في العقدين الثاني والثالث من العمر

Thrombophilia Screening

Inherited

1. Hyperhomocysteinemia (C677T) mutation
2. FV Leiden mutation (A506G) mutation
3. Mutation in prothrombin (G 20210 A)
4. Prothrombin II (PTII) mutation
5. Protein S or protein C deficiency

Acquired

1. Lupus anticoagulant
2. Cardiolipin and β2-Glycoprotein I Antibodies

Positive Diagnosis if at least 1 clinical and 1 laboratory criteria are met

Clinical criteria

- Vascular thrombosis
- Pregnancy morbidity
  - Unexplained fetal death beyond 10w
  - Premature birth before 34w
  - 3 or more unexplained spontaneous Ab before 10w

Lab criteria

- Anti-Cardiolipin IgG or IgM positive on two occasions in 12 weeks
- Anti-β_2-Glycoprotein I IgG or IgM positive on two occasions in 12 weeks
- LA on 2 occasions at least 12 weeks apart

International consensus statement on an update of the classification criteria for definite antiphospholipid syndrome (APS)

Replacing the 1999 published Sapporo classification criteria

استبدال معايير تصنيف سابورو المنشورة لعام 1999

Divided in „Clinical“ and „Laboratory“ criteria

مقسمة إلى معايير "سريرية" و "مختبرية"

Anti-β2-glycoprotein I IgG and IgM added to laboratory criteria

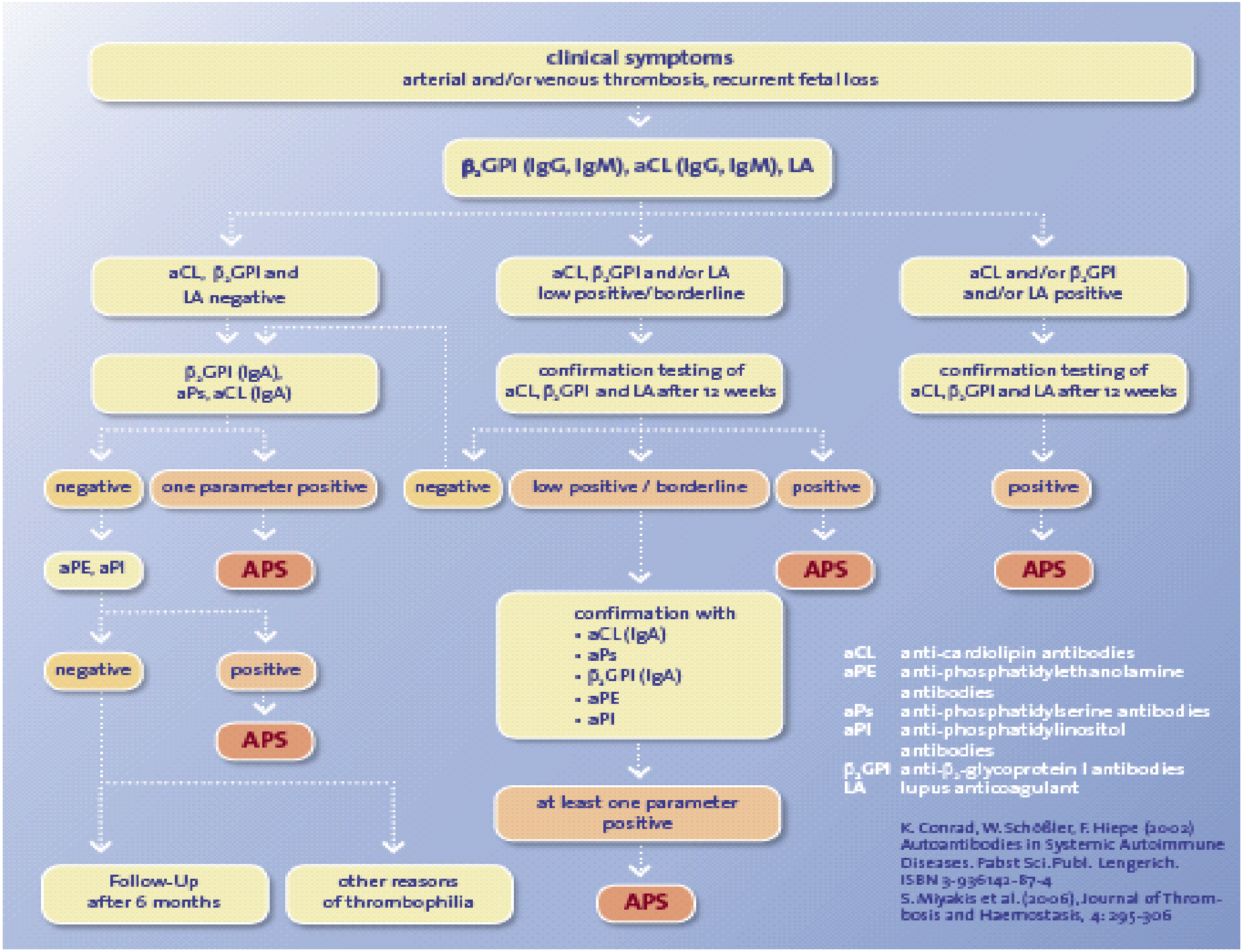
اضافة اضداد B2-glycoprotein I IgG و IgM إلى معايير المختبر

Tests should be repeated after 12 weeks

يجب تكرار الاختبارات بعد 12 أسبوعًا

Coexisting inherited or acquired factors for thrombosis are no reason to exclude Thrombophilia Screening including APS tests

رغم تشارك العوامل الموروثة أو المكتسبة للتخثر يجب عدم استبعاداختبارات APS في Thrombophilia


clinical symptoms
arterial and/or venous thrombosis, recurrent fetal loss
β2GPI (IgG, IgM), aCL (IgG, IgM), LA
aCL, β2GPI and LA negative
aCL, β2GPI and/or LA low positive/borderline
aCL and/or β2GPI and/or LA positive
β2GPI (IgA), aPs, aCL (IgA)
confirmation testing of aCL, β2GPI and LA after 12 weeks
confirmation testing of aCL, β2GPI and LA after 12 weeks
negative
one parameter positive
negative
low positive / borderline
positive
positive
aPE, aPI
APS
APS
APS
confirmation with
• aCL (IgA)
• aPs
• β2GPI (IgA)
• aPE
• aPI
negative
positive
APS
at least one parameter positive
Follow-Up after 6 months
other reasons of thrombophilia
APS
aCL anti-cardiolipin antibodies
aPE anti-phosphatidylethanolamine antibodies
aPs anti-phosphatidylserine antibodies
aPI anti-phosphatidylinositol antibodies
β2GPI anti-β2-glycoprotein I antibodies
LA lupus anticoagulant
K. Conrad, W. Schößler, F. Hiepe (2002) Autoantibodies in Systemic Autoimmune Diseases. Pabst Sci. Publ. Lengerich. ISBN 3-936142-87-4
S. Miyakis et al. (2006), Journal of Thrombosis and Haemostasis, 4: 295–306

APS in pregnancy

Obstetic manifestations in Antiphospholipid syndrom

n = 590 pregnant woman*

- Pre-eclampsia 9.5 %
- Eclampsia 4.4 %
- Placental abruption 2.0 %

n = 1580 pregnant woman*

- Fetal loss < 10 week 35.4 %
- Fetal loss ≥ 10 week 16.9 %
- Premature birth 10.6 %

*reference 2

Causes for Fetal Loss أسباب فقدان الجنين

Numerous causes for recurrent fetal loses are known, like genetic disorders, endocrine factors, anatomic anomalies, immunologic causes

الأسباب المعروفة لفقدان الجنين المتكرر معروفة ، مثل الاضطرابات الوراثية وعوامل الغدد الصماء والشذوذ التشريحي والأسباب المناعية

Miscarriages triggered by APS occur later, beginning with the 16th week of pregnancy

تحدث حالات الإجهاض لاحقًا ، بدءًا من الأسبوع السادس عشر من الحمل

Antiphospholipid antibody titers interfere directly with the proliferation and differentia-tion as well as the invasion of the syncytiotrophoblasts

- يتعارض عيار أضداد الفوسفوليبيد المضاد مباشرة مع الانتشار والتفاضل وكذلك غزو الأرومة الغازية للخلايا المخلوية

APS related complications in vitro Fertilization

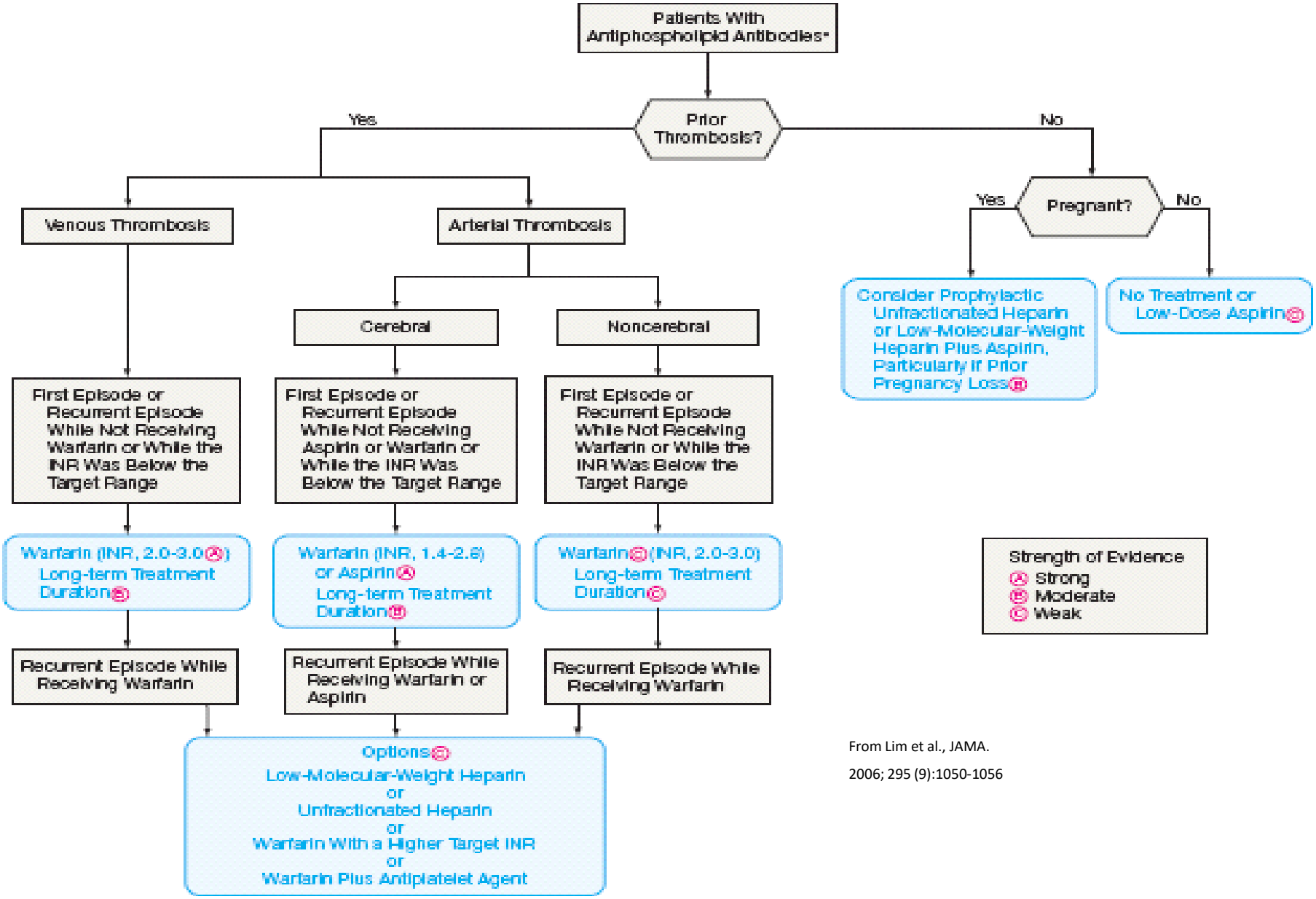
المضاعفات ذات الصلة في الاخصاب بالانبوب

Study Design:

1. At least 3 fetal losses < 10 week (n=97) or
2. IVF with at least 10 failed transfers (n=105)
3. Tested for Lupus-Antikoagulans
4. Anti-Cardiolipin-Abs IgG and IgM,
5. Antiphosphatidylserine-Abs
6. Antiphosphatidylethanolamin-Abs,
7. Antiphosphatidylinositol-Abs
8. β2-Glycoprotein-I-Abs, ANA

84 (23 %) positive for at least one autoantibody
Highest Correlation with β2GP I-Abs and ANA

Antiphospholipid Syndrome



From Lim et al., JAMA.

2006; 295 (9):1050-1056

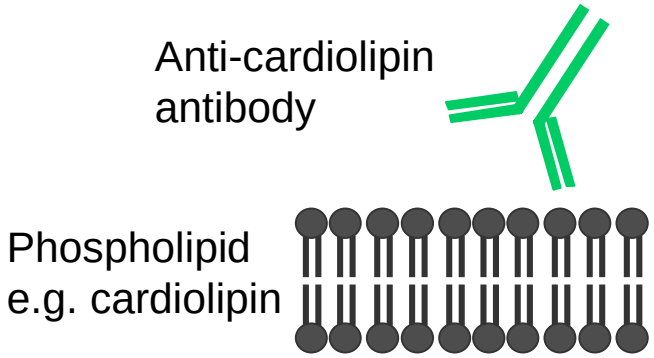
Antiphospholipid Syndrome

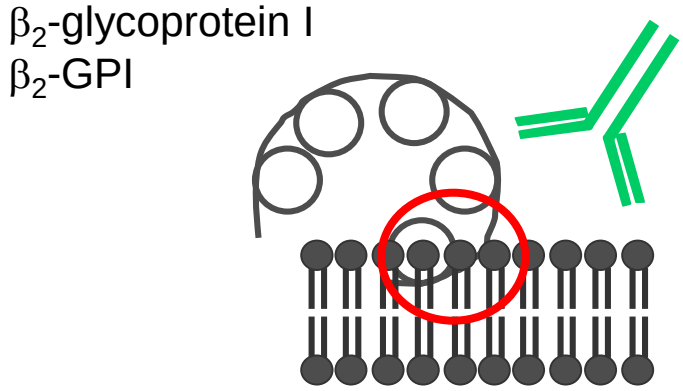
- Cardiolipin-Antibodies
- Cardiolipin Antibodies Screen/ Combi
- IgG / IgM / IgA
  - Cardiolipin is immobilized in a biological active vesicle حويصلة like structure
  - Presentation together with human β_2-Glycoprotein I

  as cofactor on the microtiter plate and in the dilution buffer
  - Calibration and validation by use of Harris sera and Koike preparations
  - Included in the revised international classification criteria for APS

Antiphospholipid Syndrome

Possible antigenic structures of antiphospholipid antibodies



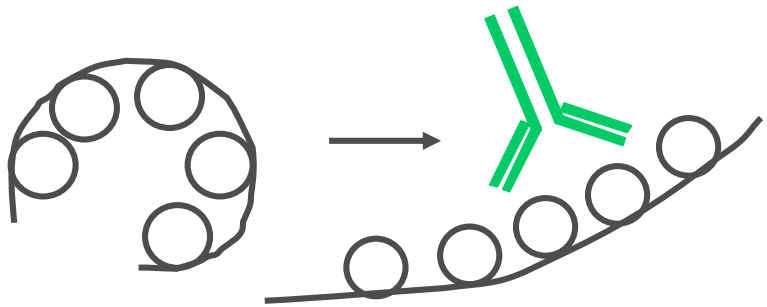


First description of anti-cardiolipin
antibodies: ELISA with cardiolipin coated to wells

β_2-glycoprotein I has been identified
as protein cofactor required for binding
of antibodies to cardiolipin (common epitope)

Antiphospholipid Syndrome

Possible antigenic structures of antiphospholipid antibodies



Anti-phospholipid/cardiolipin antibodies can bind to $ß_2$-GPI alone but epitopes are hidden inside the molecule

Binding is only possible upon conformational change of $ß_2$-GPI

Antiphospholipid Syndrome

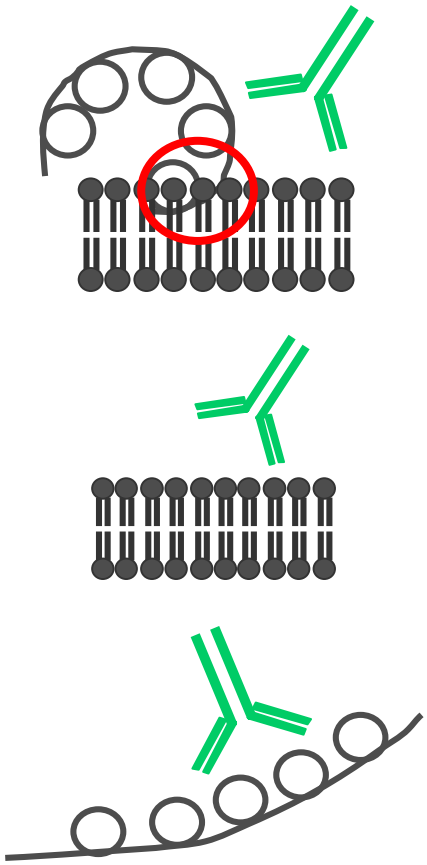
Possible antigenic structures of antiphospholipid antibodies

Conformational change is induced by binding to negatively charged cardiolipin or negatively charged (irradiated) ELSA plate

These anti-$ß_2$-GPI antibodies are regarded as being more specific than anti-cardiolipin antibodies but they lack sensitivity

Antiphospholipid Syndrome

Majority of commercially available Cardiolipin ELISA tests employ Cardiolipin plus ß$_2$-GPI as cofactor



This allows simultaneous detection of:

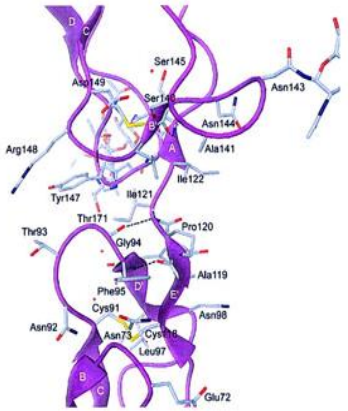
- ß$_2$-GPI dependent anti-cardiolipin antibodies
- Anti-cardiolipin antibodies
- Anti-ß$_2$-GPI antibodies

Antiphospholipid Syndrome

- Cardiolipin-Antibodies
- Calibration against all available international reference preparations

  - Harris sera
    Calibration in GPL-U/ml, MPL-U/ml, APL-U/ml
  - Koike preparations
    monoclonal antibodies, named HCAL and EY2C9
    Calibration in ng/ml

Antiphospholipid Syndrome



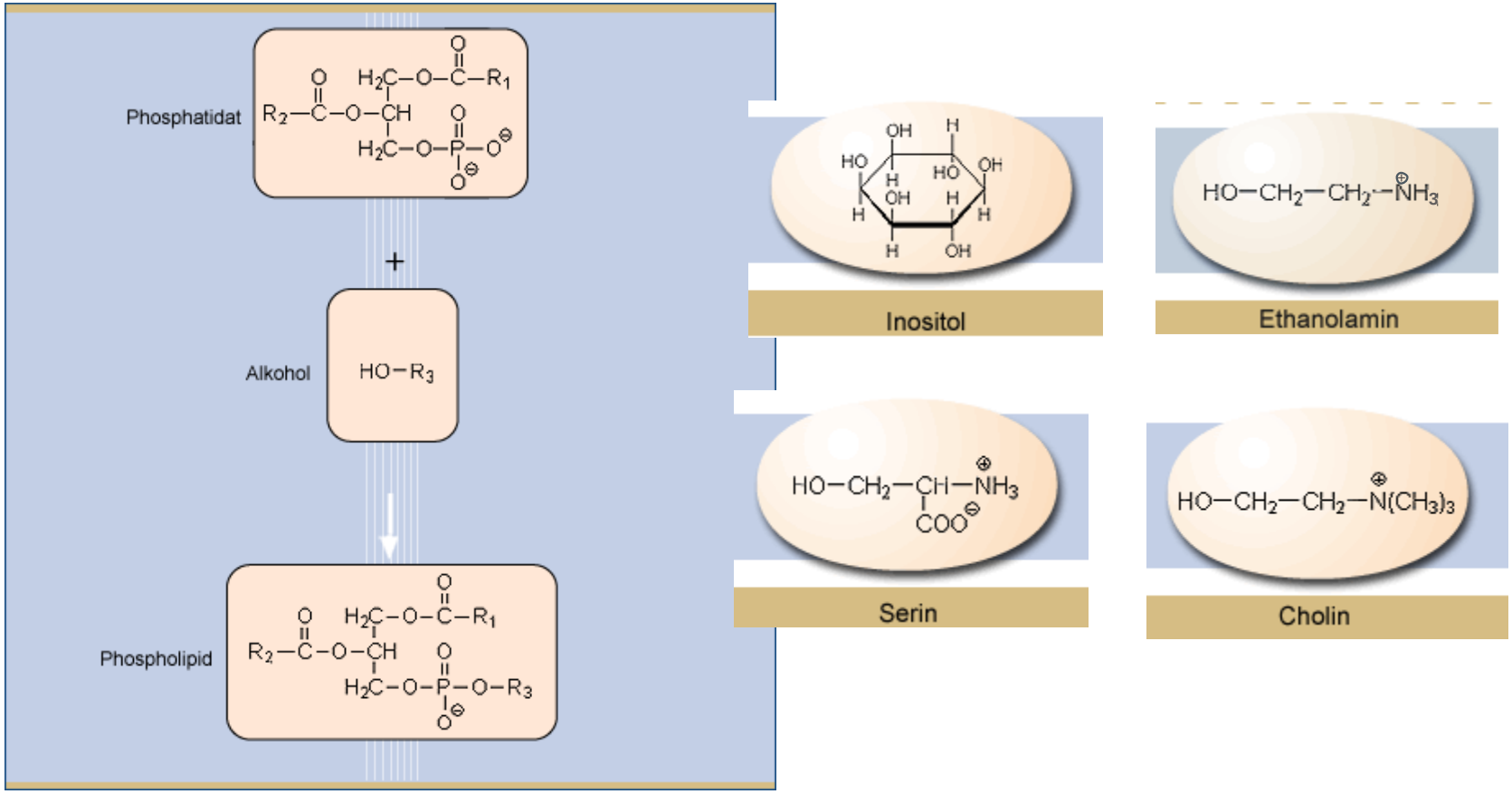
- β2-Glycoprotein I-Antibodies
- β2-Glycoprotein I Antibodies Screen

IgG IgM / IgA

  - human native antigen
  - Calibration and validation by use of Harris sera and Koike preparations
  - Included in the revised international classification criteria for APS

Antiphospholipid Syndrome

PHOSPHOLIPIDS



Antiphospholipid Syndrome

Antiphospholipid antibodies are a heterogeneous family of antibodies that react with:

Phospholipids

Cardiolipin
Phosphatidylcholine
Phosphatidylethanolamine
Phosphatidylinositol
Phosphatidylserine etc …

Phospholipid-binding proteins (Proteins involved in coagulation)

$ß_2$-Glycoptotein I
Thrombin
Prothrombin
Annexin V etc …

Complexes of proteins and phopsholipids

Antiphospholipid Syndrome

- Phosphatidylserine-Antibodies
- Phosphatidylserine-Antibodies Screen IgGAM
  - High correlation to lupus anticoagulans
  - SLE patients frequently show positive for aPS Abs (mainly IgG and/or IgA)
  - In thrombotic-free patients the coincidence of aCL Abs **and** aPS Abs increases the risk for thrombosis by 30 %
  - May be the only autoantibody present in APS (5-10 %)