11th Meeting of the European Forum on
Antiphospholipid Antibodies
*Maastricht, The Netherlands*

APSFORUMMAASTRICHT

# Updating of APS pathophysiology: does it impact on our clinical management?

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# Disclosures: Dr. PL Meroni

- consultant to Inova Diagnostics Inc., Termo Fisher Scientific, Pfizer, Abbvie, UCB, MSD.

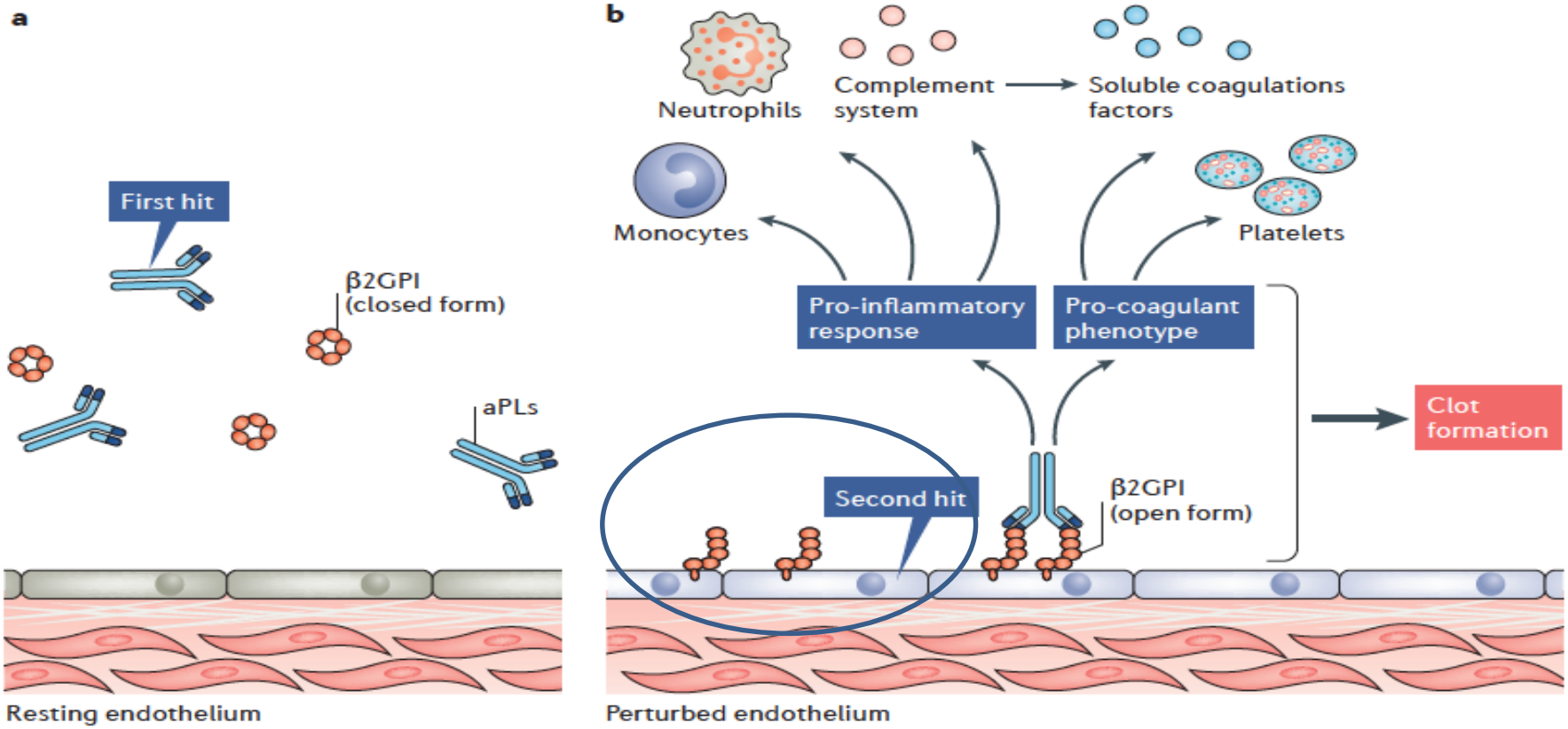
# Outline of presentation

- vascular APS: the culpirit cell
- β2GPI tissue expression as the true 2nd hit
- Complement activation the real new entry
- OBG – APS another disease?
- risk assessment: new approach tomorrow?

# Key points in vascular APS

- Clotting is usually localized in common APS while systemic coagulopathy is present in CAPS only.
- This finding supports an upstream key pathogenic role for the endothelium rather than for other cell types (i.e. Mo, neutrophils, PP) or fluid phase components involved in the coagulation cascade.

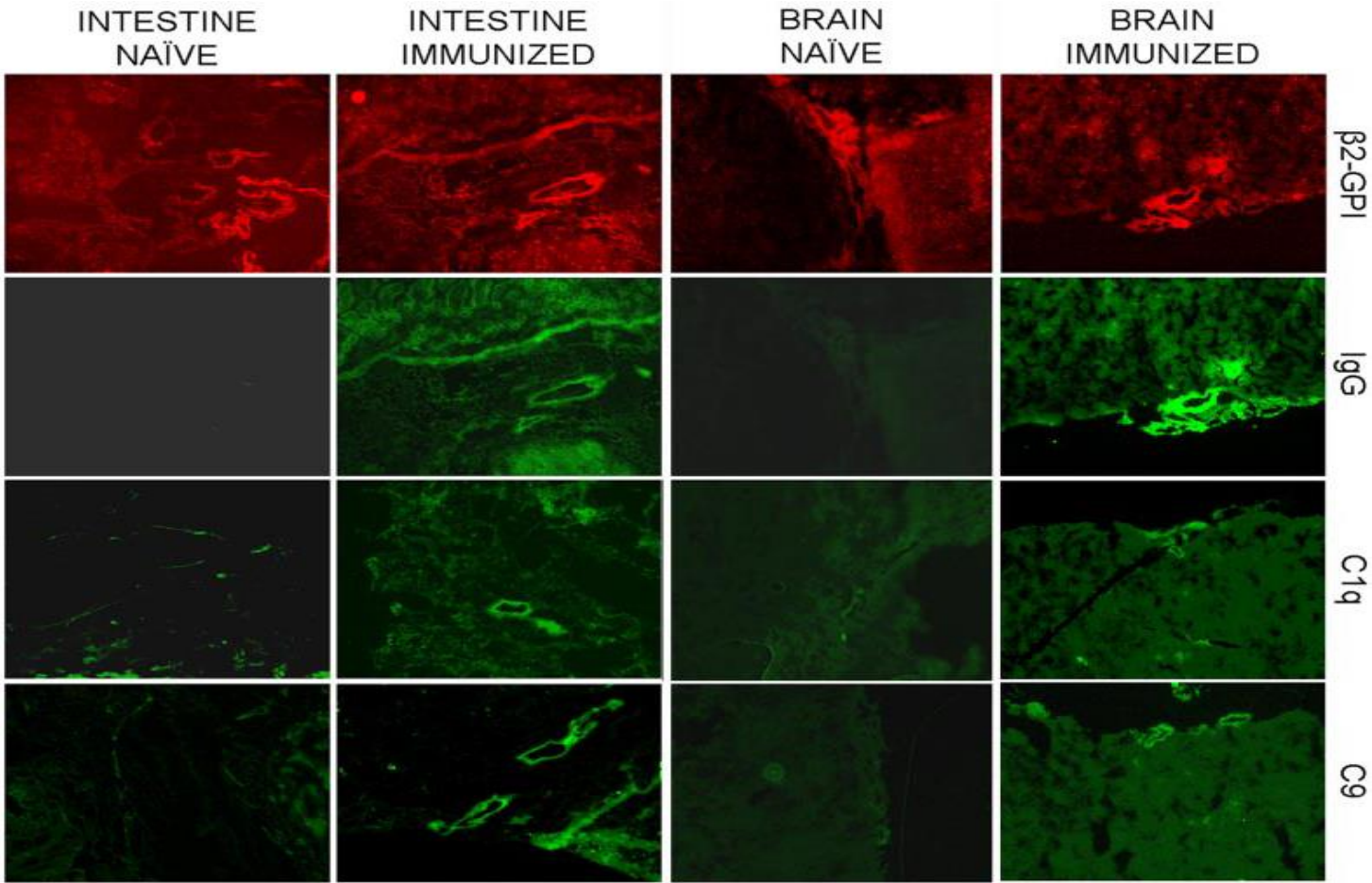
- We still do not know the reasons for the selective arterial (and particular anatomical localizations) or venous vessels involvement.



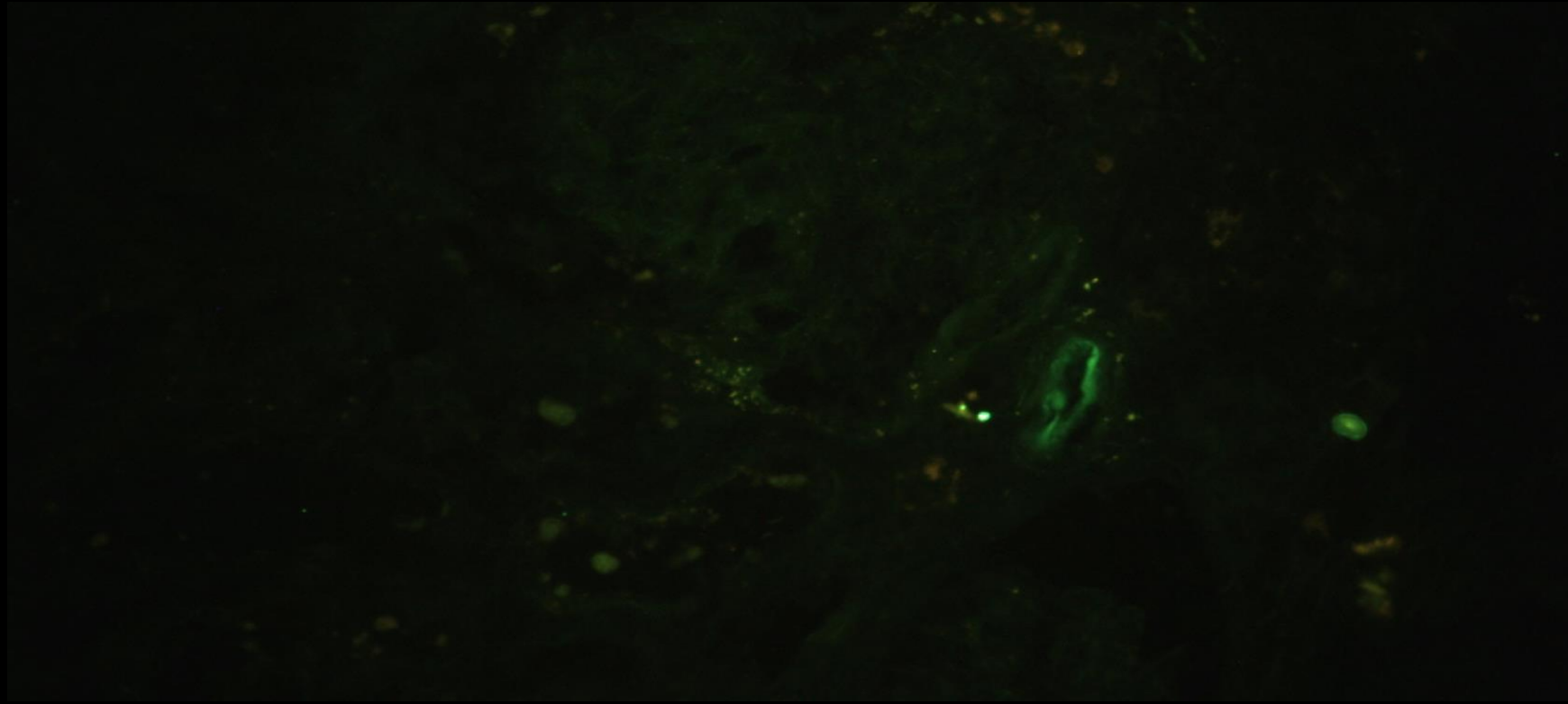

Nature Reviews | Rheumatology

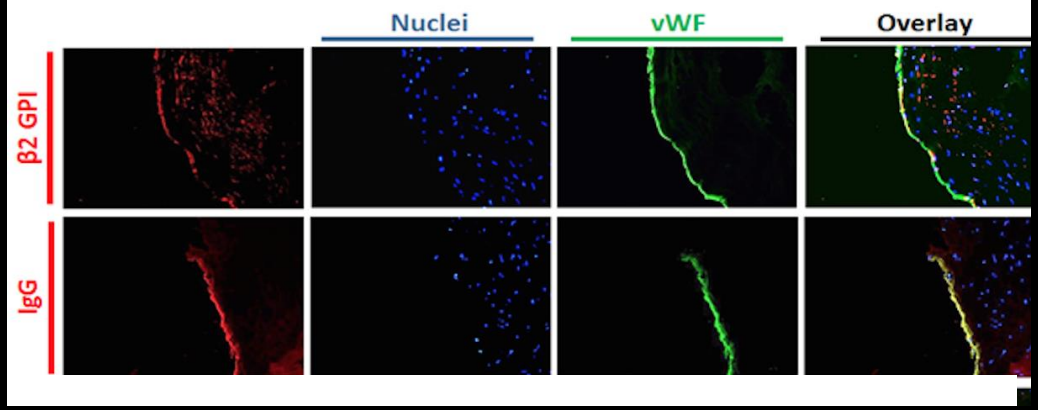
Meroni PL et al *NRR* '18

# β2GPI distribution was similar in naive & immunized mice *BUT* IgG/C1q/C9 colocalized after LPS (Agostinis et al *Blood* '11)

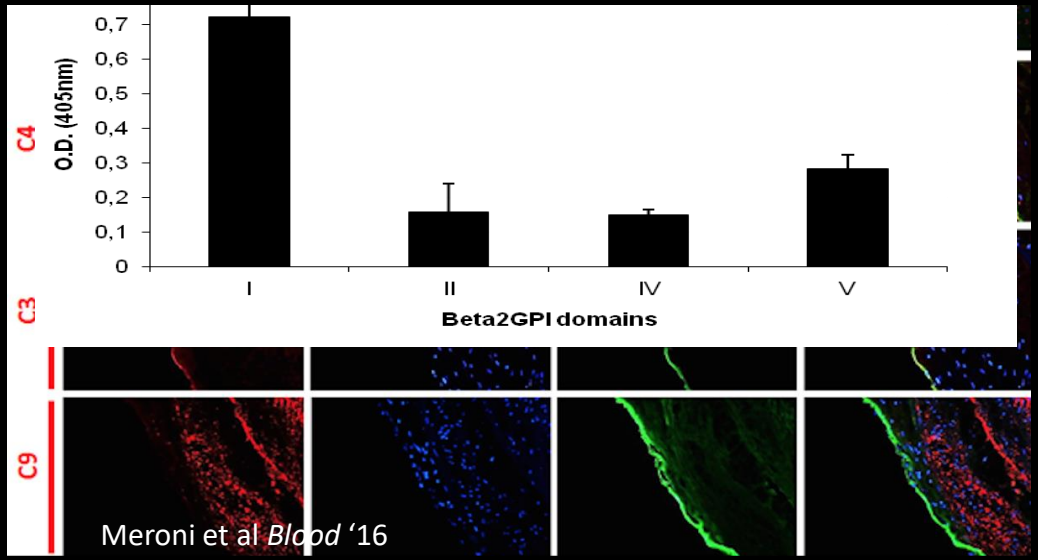


# Normal human kidney stained by anti-hu β2GPI IgG MoAb: NO SIGNAL!

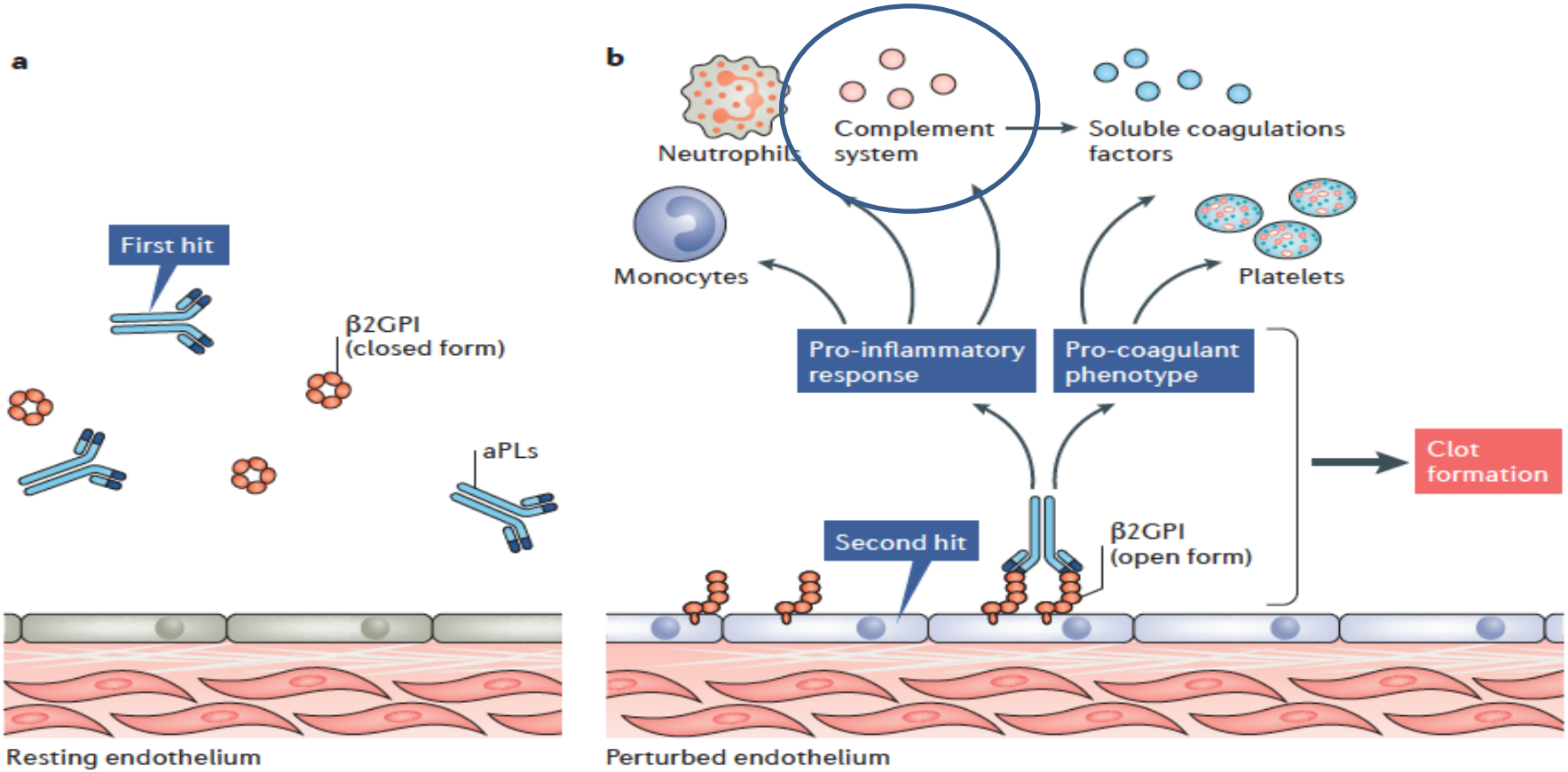





# Tissue β2GPI is stained by MBB2 MoAb that recognizes D1




Meroni et al *Blood* '16




Nature Reviews | Rheumatology

Meroni PL et al *NRR* in press

# Complement in APS models

Blood. **2005** Oct 1;106(7):2340-6. Epub **2005** Jun 14.

## Thrombus formation induced by antibodies to beta2-glycoprotein I is complement dependent and requires a priming factor.

**Fischetti** F[1], **Durigutto** P, Pellis V, Debeus A, Macor P, Bulla R, Bossi F, Ziller F, Sblattero D, Meroni P, Tedesco F.

Arthritis Rheum. 2005 Jul;52(7):2120-4.

## Requirement of activation of complement C3 and C5 for antiphospholipid antibody-mediated thrombophilia.

**Pierangeli** SS[1], **Girardi** G, Vega-Ostertag M, Liu X, Espinola RG, Salmon J.

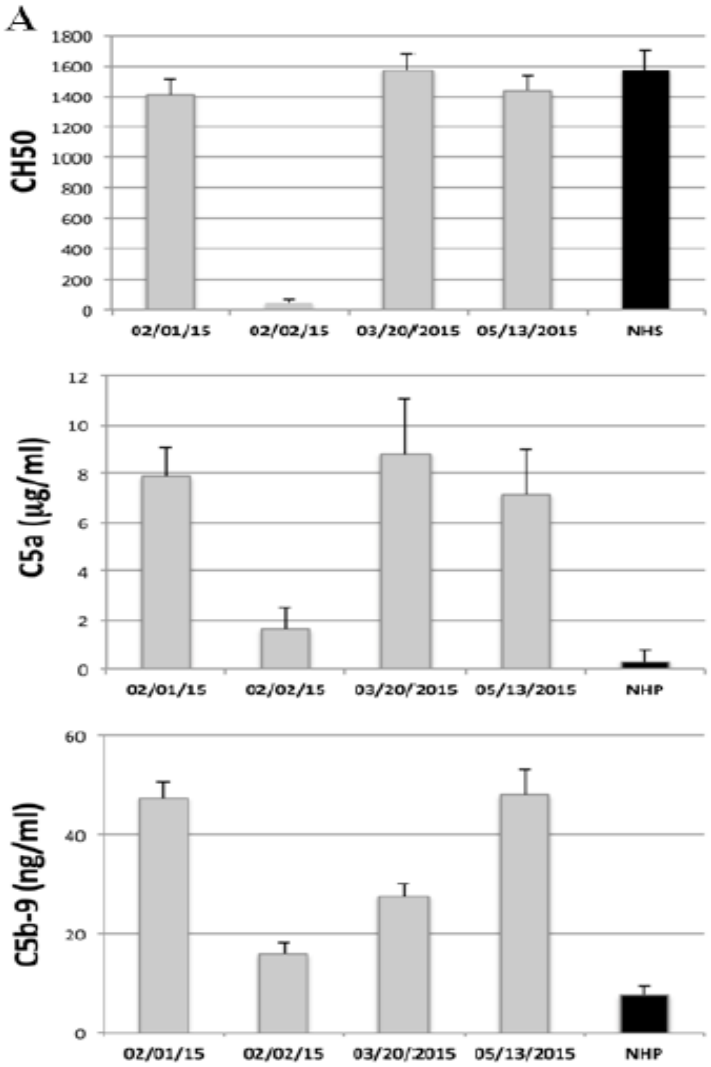
Lupus. 2012 Dec;21(14):1497-505. doi: 10.1177/0961203312458839.

## C6 knock-out mice are protected from thrombophilia mediated by antiphospholipid antibodies.

Carrera-Marín A[1], Romay-Penabad Z, Papalardo E, Reyes-Maldonado E, García-Latorre E, Vargas G, Shilagard T, **Pierangeli** S.

# Complement levels in APS sera

- Hypocomplementemia in up to half of primary APS pts but *no* clear correlation with the thrombotic events (Davis & Brey *CER* '92; Carbone et al *Lupus* ' 99; Ramos-Casals et al *Lupus* ' 04; Oku et al *Ann Rheum Dis* '09)
- Reduced complement levels (not necessarilly pathological low) and increased C3a/C4a levels in primary APS (Oku et al *ARD* '09)
- Significantly increased levels of complement activation products (Fragment Bb & C3a-desArg which correlated among them and with the double/triple aPL positivity) (Devreese et al *Thromb Haemost* '10; Breen et al *Thromb Haemost* '12)

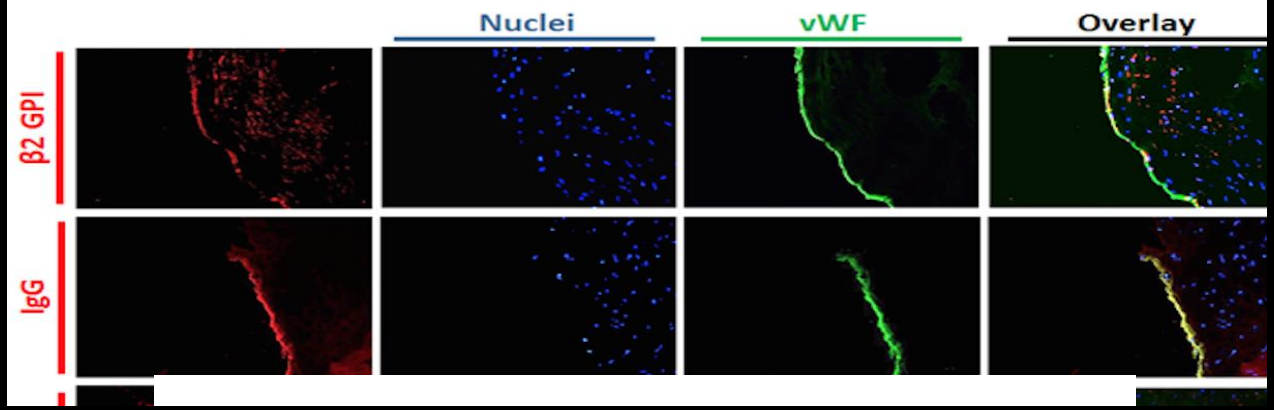



**Complement activation in antiphospholipid syndrome and its inhibition to prevent rethrombosis after arterial surgery**

Pier Luigi Meroni,[1,2] Paolo Macor,[3] Paolo Durigutto,[3] Luca De Maso,[3] Maria Gerosa,[1] Mariano Ferraresso,[1,4] Maria Orietta Borghi,[1,2] Tom Eirik Mollnes,[5-7] and Francesco Tedesco[2]

BLOOD, 21 JANUARY 2016 • VOLUME 127, NUMBER 3

- aPL-mediated complement activation in the arterial wall of PAPS patients.
- Rational for complement inhibition therapy (*eculizumab*) at least in preventing re-thrombosis triggered by vascular surgery

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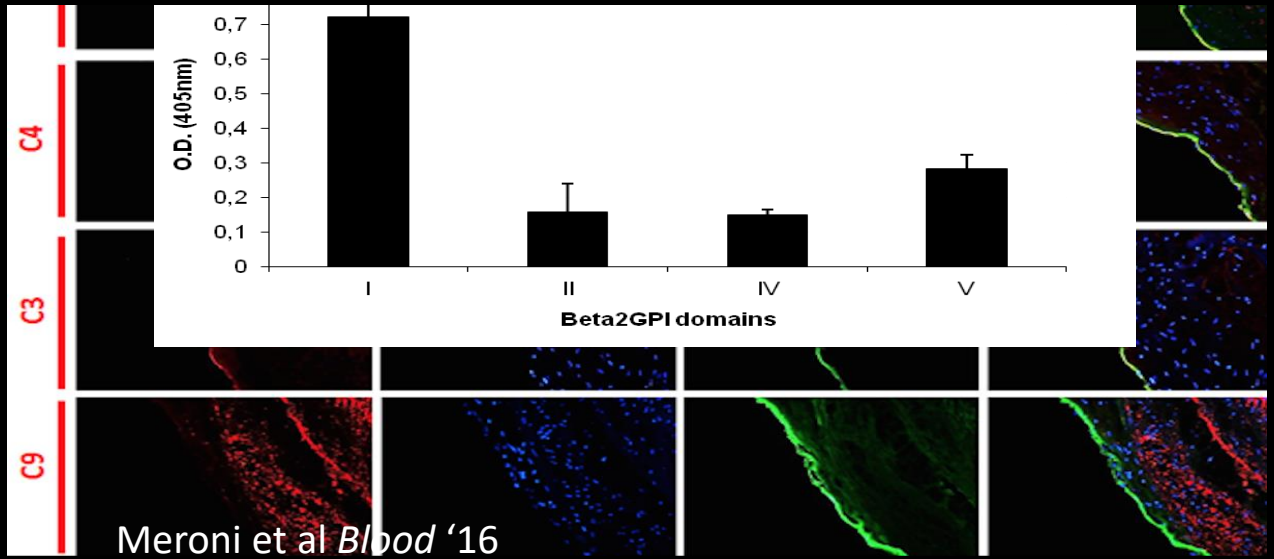
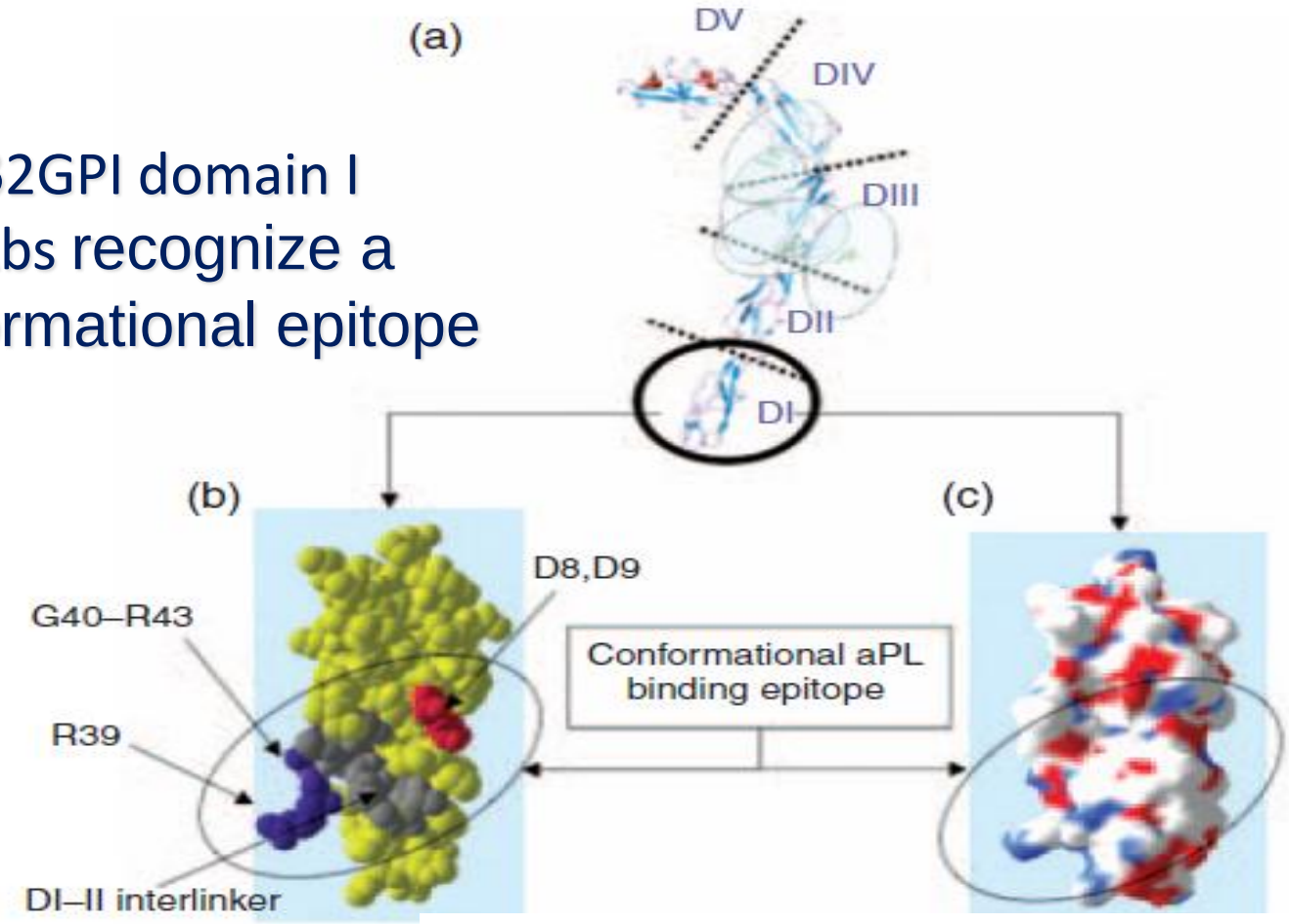



Meroni et al *Blood* '16

# Anti-β2GPI domain I autoAbs recognize a conformational epitope




Iannou et al *A&R* '11

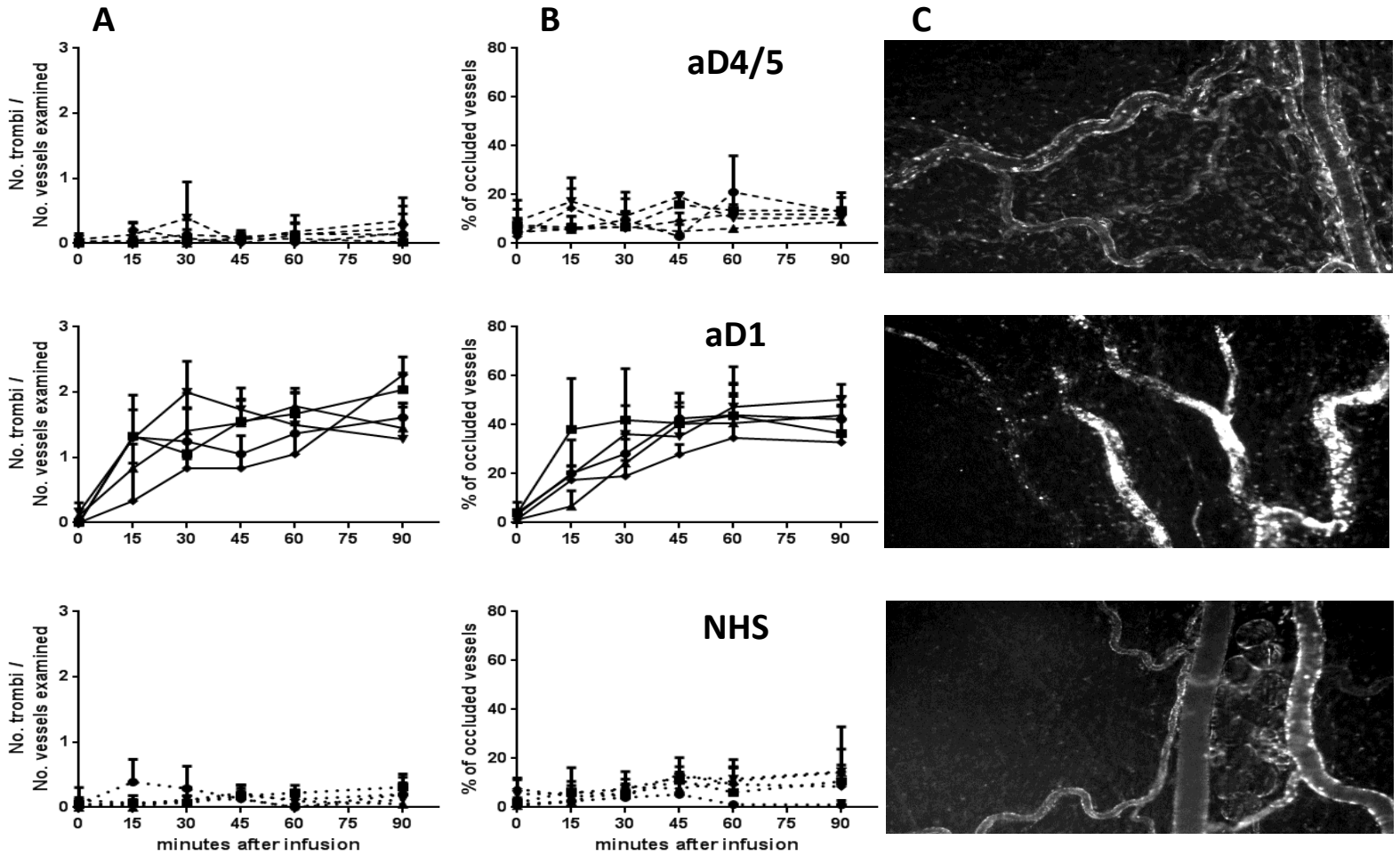
# Anti-domain 1/ 4,5: clinical meaning

- Anti-D1 IgG are present in all the patients with high risk aPL profile (vascular & obstetric)(Radin et al *Sem Thromb Hemost* '18)
- aPL asymptomatic carriers display a preferential polarization profile toward D4,5.

- "pure" anti-D4,5 are rare but usually asymptomatic or with the absence of aCL and/or LA and in the majority of the cases with doubtful APS picture which does not fulfill the classification/diagnostic criteria.

Andreoli et al *AR* '15; Chighizola et al *J Autoimmun* '18

# Anti-D1 but not anti-DIV-V pos sera are thrombogenic in animals




Durigutto et al *in revision*

# Take home messages

- The polarization towards anti-D1 IgG as a fingerprint of systemic autoimmunity and higher risk for clinical manifestations.
- Low anti-D1/D4,5 ratio is preferentially present in asymptomatic carriers and in patients with doubtfull or no APS.

**Should we introduce the $\beta_2$GPI epitope characterization as a test of 2$^{nd}$ level?**

- **Up to 20% of anti-$\beta_2$GPI pos samples are not reactive against D1.**

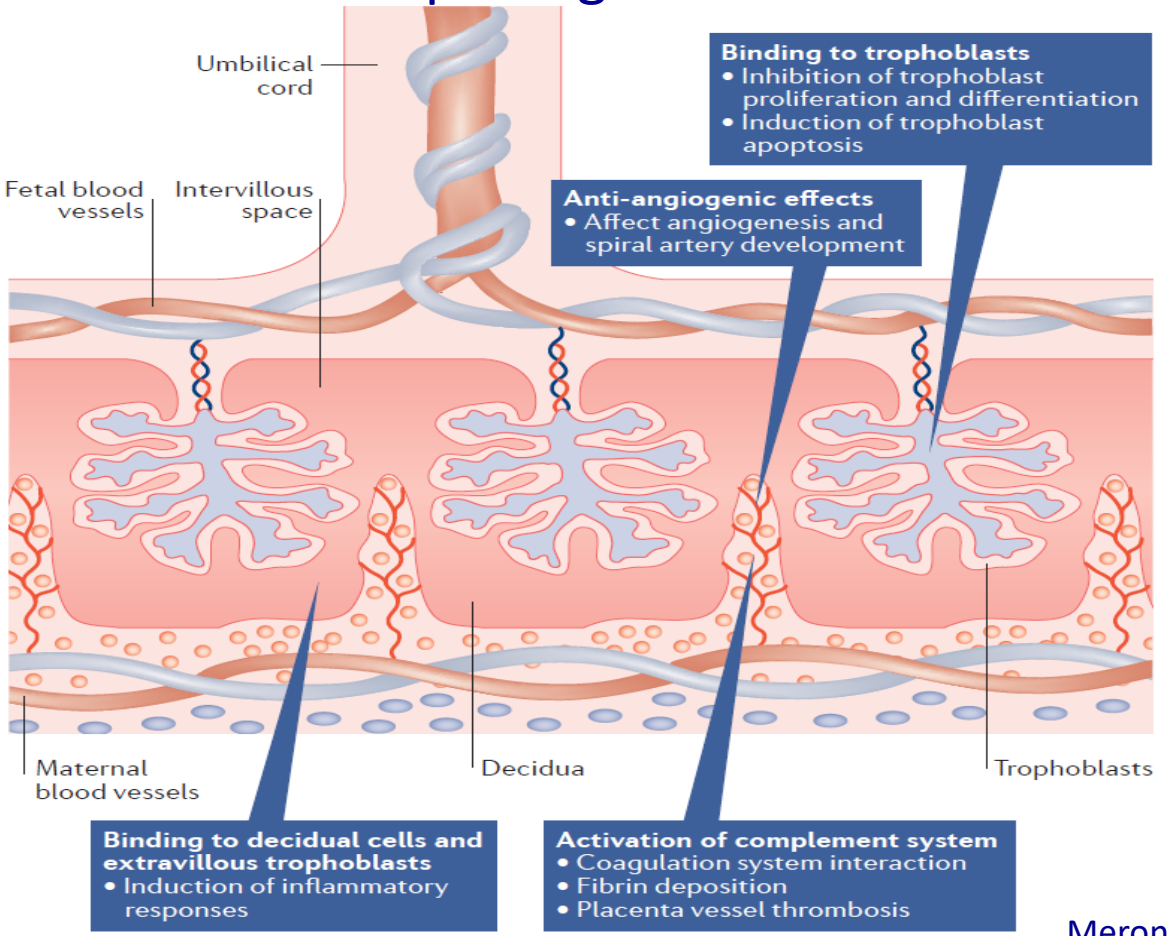
# Obstetric versus vascular APS

- ✓ Although most patients with APS have both vascular & obstetric manifestations, isolated vascular or obstetric variants exist. Obstetric APS is a specific subset within the APS box. Maternal thrombosis & progression to SLE are scarce (Alijotas-Reiget al *Autoimmun Rev* '15)

- ✓ Thrombosis represents the main clinical manifestation of vascular APS, whereas obstetric APS is characterized by defective placentation; non-thrombotic mechanisms might be more important than placental infarction in the pathogenesis of obstetric APS .

Meroni et al *Nat Rev Rheum* '18

# Obstetric APS: pathogenic mechanisms



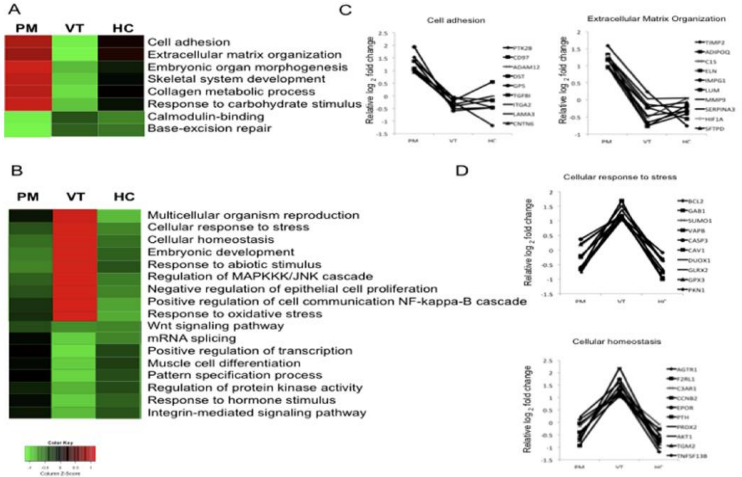

Meroni et al *Nat Rev Rheum* '18

# Obstetric vs vascular APS

- Anti-β2GPI Abs are the main pathogenic aPL mediating the placental damage. *Same pathogenic aPL as in vascular APS?*

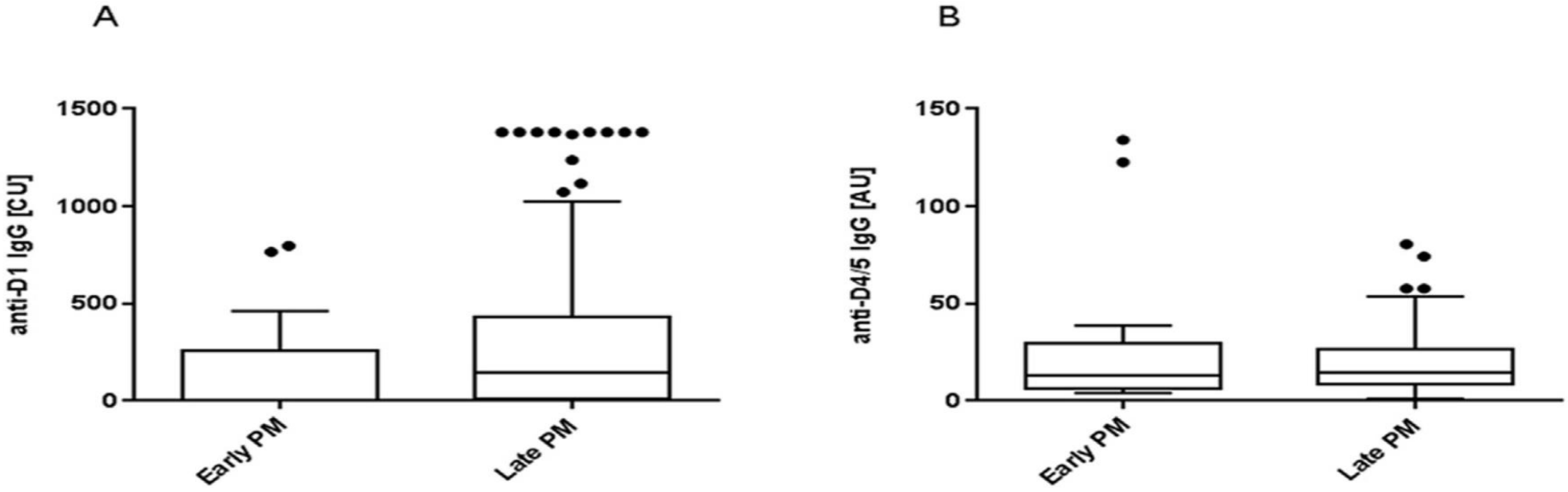
- IgG fractions from pure obstetric APS display different *in vitro* effects on Mo & trophoblast cells

(Lambrianides et al *J Immunol* '10; Poulton et al *Am J Reproduct Immunol* '15; Ripol et al *J Autoimmun* '18)

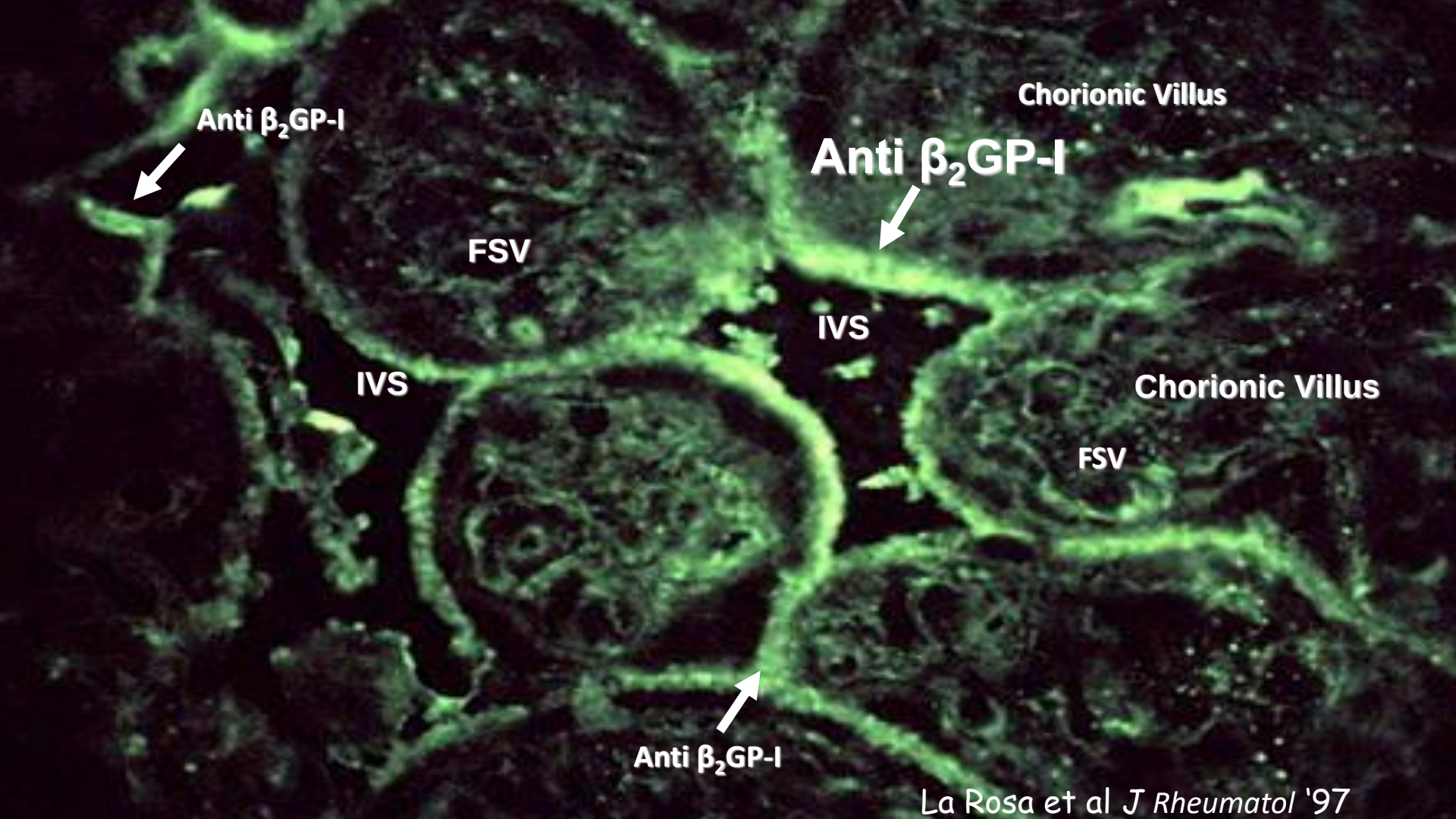



- Passive transfer of aPL IgG in naive pregnant animals does not need the *2° hit* to induce fetal loss (Meroni et al *Nat Rev Rheumatol* '11)
- aPL IgG from pure vascular or OBG APS induce thrombi & fetal loss in animal (Meroni et al *Nat Rev Rheum* '18)

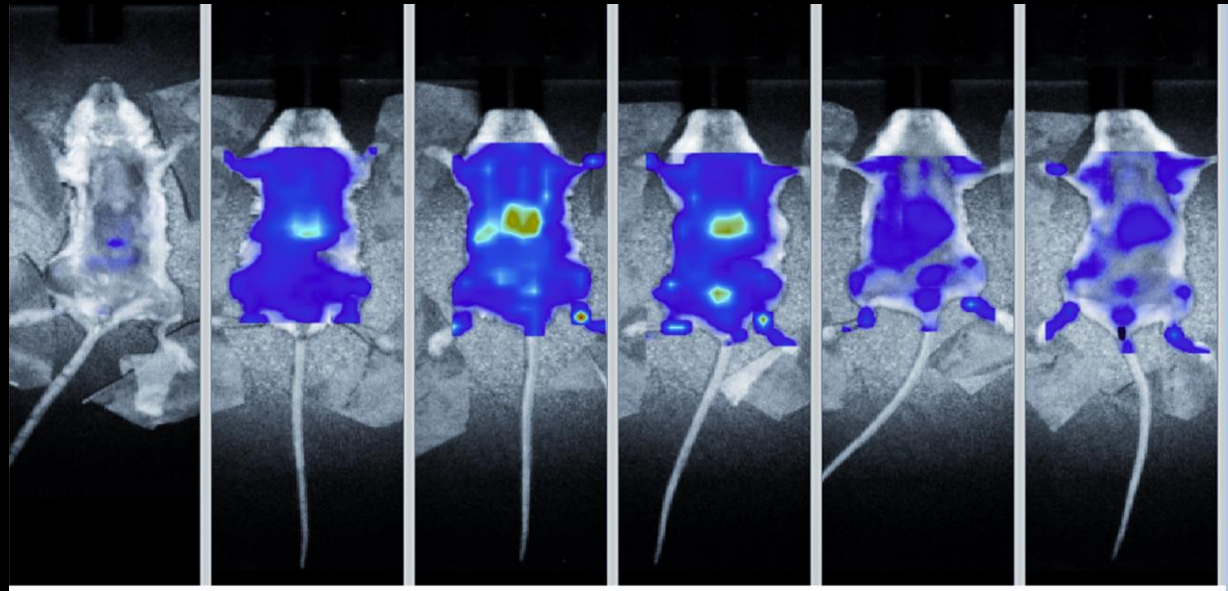
# Levels of anti-D1 and anti-D4/5 IgG antibodies in women with early and late pregnancy morbidity.



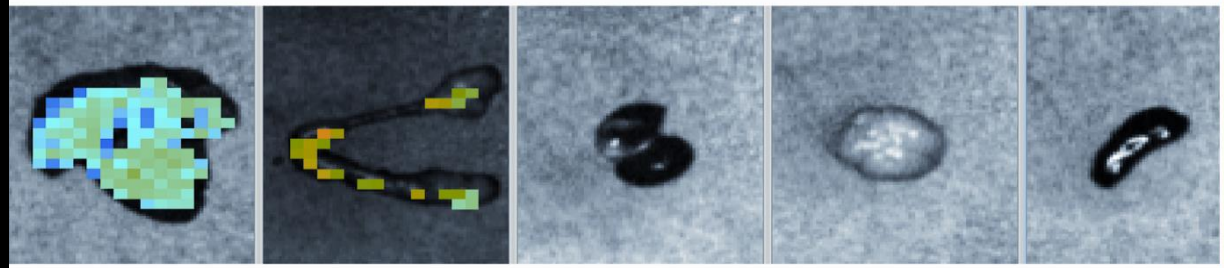

Chighizola et al *J Autoimmun* '18




La Rosa et al *J Rheumatol* '97



prescan 3min 24h 48h 144h 168h



liver uterus kidney brain spleen

Intensity (NC)

6.72e+004

5.04e+004

3.36e+004

1.68e+004

1.67

Lifetime (NS)

2

1.75

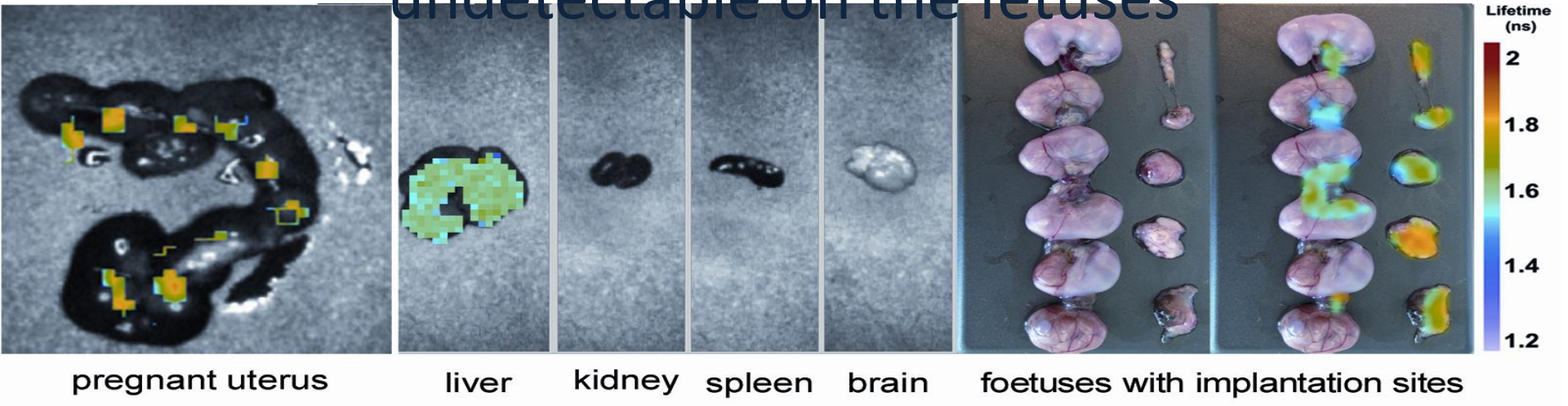
1.5

1.25

1

Agostinis et al *Blood* '11

***Pregnant mice***: Specific β2GPI signal both in the liver &in the uterus. The uterine signal was localized at the implantation sites, but was undetectable on the fetuses




Agostinis et al *Blood* '11

# The obstetric risk and the effect of treatment in women with low titer anti-phospholipid antibodies

- ✓ Low titer aPL increases the probability to have an obstetric complication
  - ✓ The risk increases in case of double aPL positivity
- → Time to reconsider the current classification criteria/treatment for obstetric APS?

Chighizola et al *submitted*

# Women with positive aPL do not carry all the same obstetric risk: *risk profile*

- Double/triple vs single aPL assay positivity
- IgG vs IgM isotype
- Medium/high titers vs low


The Journal of Rheumatology Volume 42, no. 2

Obstetric Antiphospholipid Syndrome: Has the Black Swan Swallowed a Red Herring?

CHRISTINE A. CLARK, KAREN A. SPITZER and CARL A. LASKIN

J Rheumatol 2015;42;155-157

The Journal of Rheumatology Volume 42, no. 2

Obstetric Antiphospholipid Syndrome: Lobsters Only? Or Should We Also Look for Selected Red Herrings?

PIER LUIGI MERONI, CECILIA B. CHIGHIZOLA, MARIA GEROSA, LAURA TRESPIDI and BARBARA ACAIA

J Rheumatol 2015;42;158-160

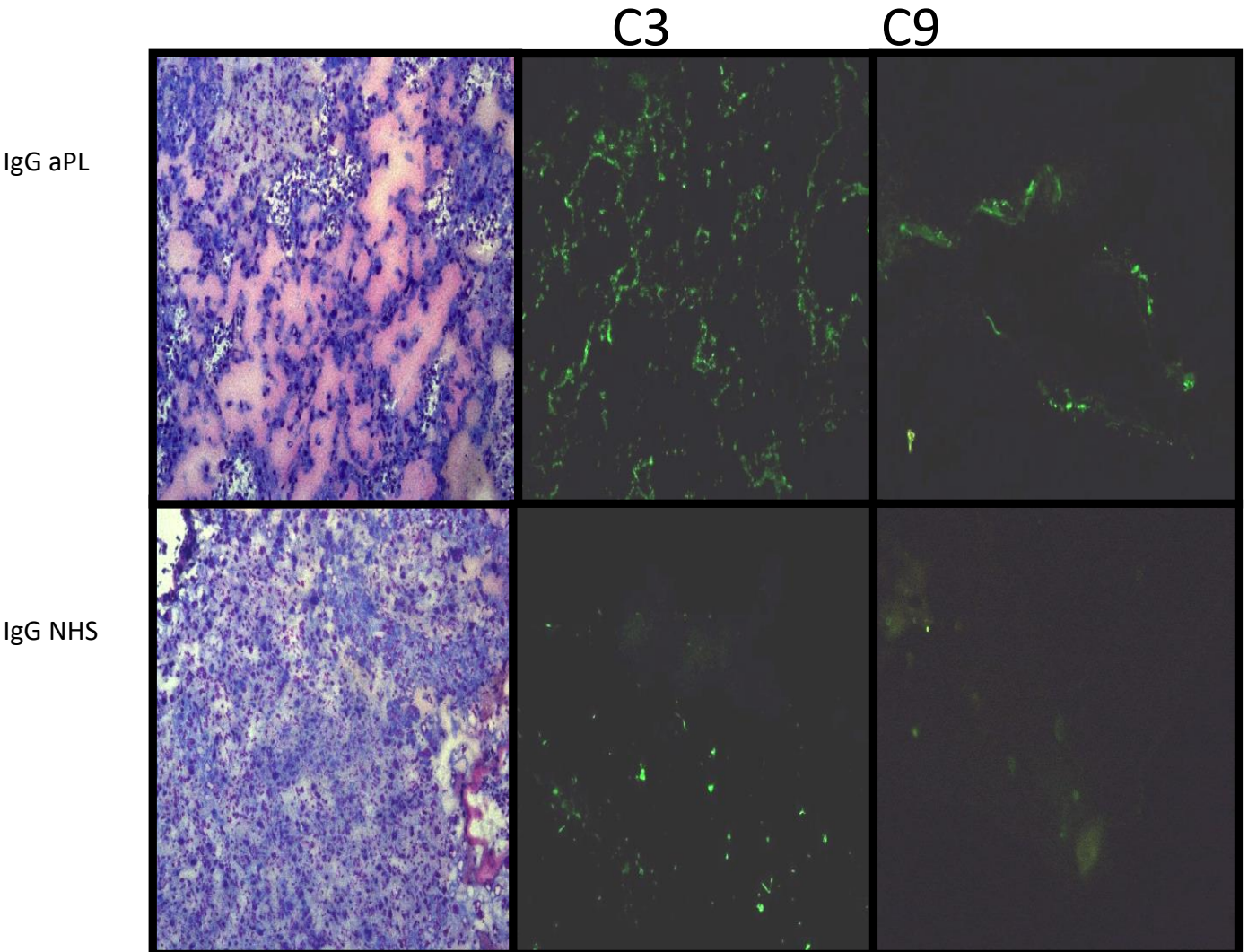

C3
C9
IgG aPL
IgG NHS

**TABLE 1** | Clinical characteristics of the PAPS patients examined for placental C deposits.

| Patients | Diagnosis | LA | aCL IgG/IgM | anti-β2GPI IgG/IgM | Outcome | Therapy |
|---|---|---|---|---|---|---|
| BAC 1 | PAPS | Pos | High/high | High/high | Fetal loss < 10 weeks | LMWH/ASA |
| BAC 2 | PAPS | Pos | High/high | High/high | Fetal loss > 10 weeks (twins) | LMWH/ASA |
| BAC 3 | PAPS | Pos | High/high | High/high | Live baby 35 weeks | LMWH/ASA/ivig/CS |
| BA | PAPS | Pos | High/high | High/high | Fetal loss > 10 weeks | LMWH/ASA |
| TD | PAPS | Neg | Med/low | Med/neg | Live baby 38 weeks | LMWH/ASA |
| SA | PAPS | Pos | nd | nd | Fetal loss > 10 weeks | ASA[a] |
| SE | PAPS | Pos | High/high | High/med | Live baby 30 weeks | LMWH/ASA |
| PA | PAPS | Pos | High/high | nd | Fetal loss > 10 weeks | None[b] |
| FO | PAPS | Neg | High/neg | High/neg | Live baby 38 weeks | LMWH/ASA |
| BO | PAPS | Pos | Neg/neg | Neg/neg | Live baby 35 weeks | LMWH/ASA |
| PU | PAPS | Pos | High/neg | High/med | Live baby 38 weeks | LMWH/ASA |
| AC | PAPS | Neg | High/neg | High/med | Live baby 33 weeks | LMWH/ASA |
| BL | PAPS | Pos | High/med | High/med | Live baby 31 weeks | LMWH/ASA |

*PAPS, primary antiphospholipid syndrome (5); C, complement; LA, lupus anticoagulant; aCL, anti-cardiolipin antibodies; anti-β2GPI, anti-beta2 glycoprotein I antibodies; LMWH, low molecular weight heparin; ASA, aspirin; ivig, intravenous immunoglobulins; CS, corticosteroids; nd, not detected.*

*[a]The patient was classified as aPL-positive asymptomatic carrier, and her first pregnancy was treated with ASA only.*

*[b]The patient was not treated with the standard therapy because the positivity for aPL was found after the abortion.*

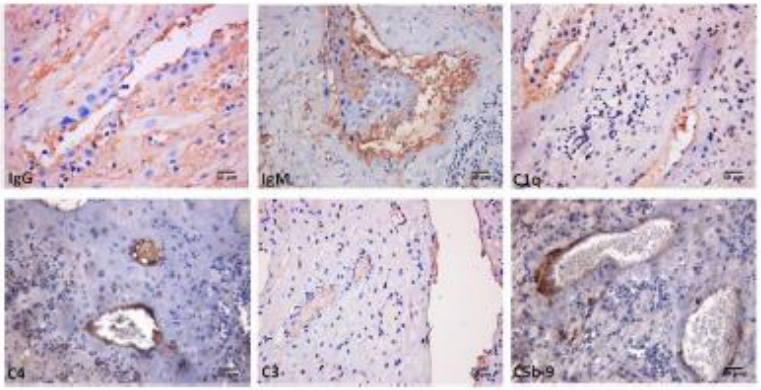



**FIGURE 1** | Immunoperoxidase staining of a representative placental decidua from a primary antiphospholipid syndrome patient showing deposition of immunoglobulin (Ig) and various C components (20x magnification).

Fredi M et al Risk Factors for Adverse Maternal and Fetal Outcomes in Women With Confirmed aPL Positivity: Results From a Multicenter Study of 283 Pregnancies. *Front Immunol*. 2018

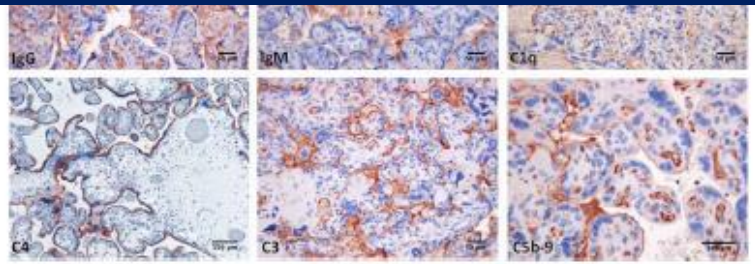



**FIGURE 2** | Immunoperoxidase staining of representative placental villi from a primary antiphospholipid syndrome patient showing deposition of immunoglobulin (Ig) and various C components (20x magnification).

Tedesco et al *Front Immunol* '18

# Acknowledgments

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UNIVERSITÀ DEGLI STUDI DI TRIESTE

UNIVERSITÀ DEGLI STUDI DI BRESCIA