ATHEROTHROMBOSIS: Basic Concepts

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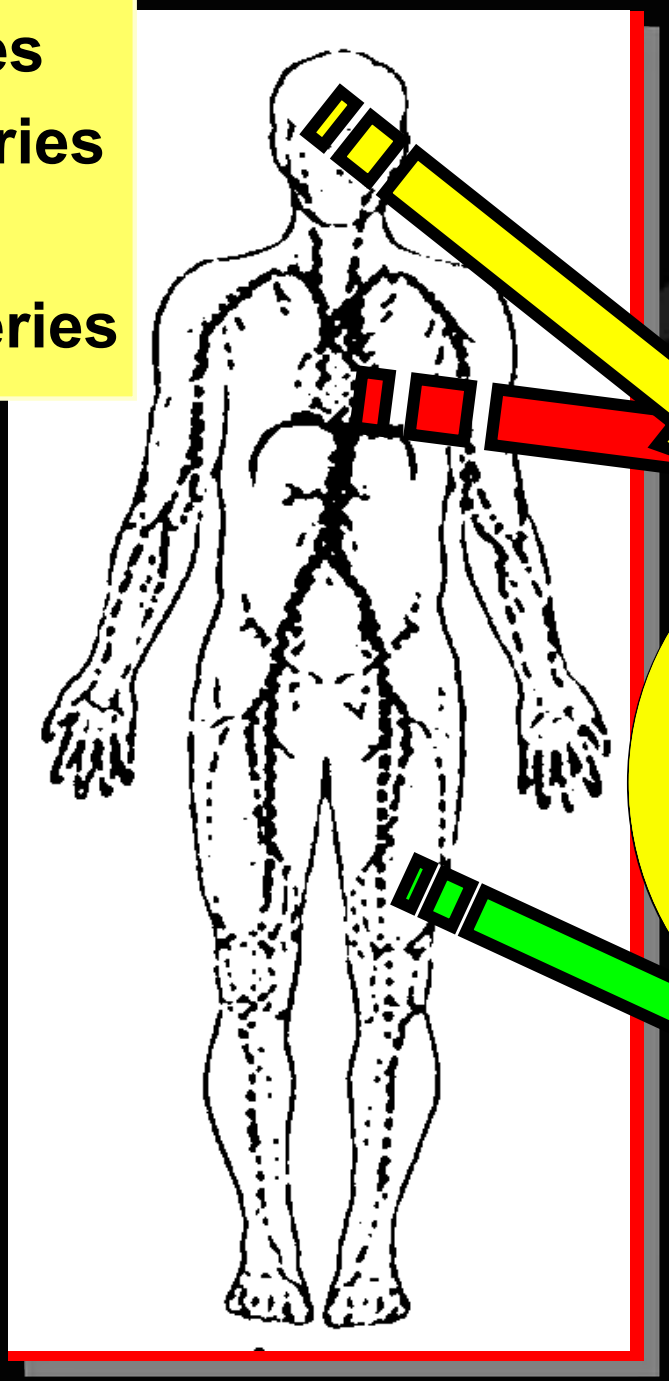
CBCS/ESC Summer School on Cardiovascular Sciences

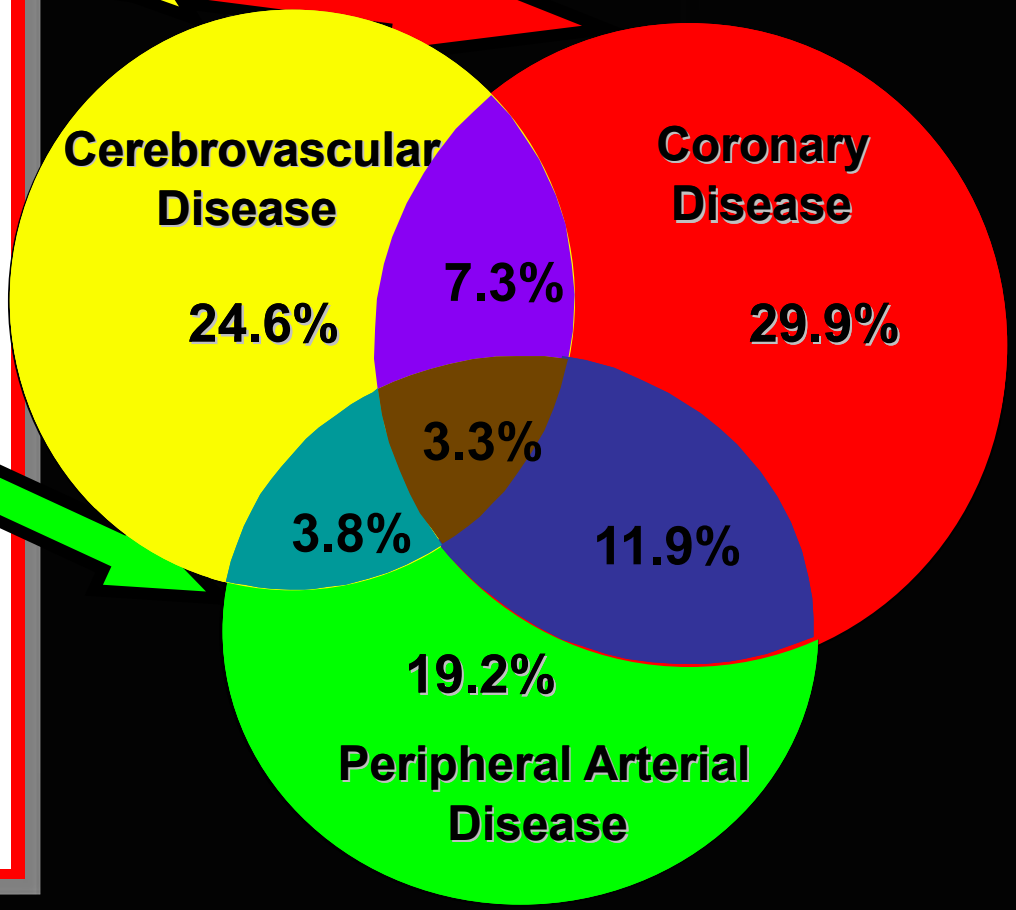
Sophia, Antipolis, June 2011

Atherosclerosis: *a systemic disease*

carotid arteries
coronary arteries
aorta
phemoral arteries



Overlap of Symptomatic Atherosclerosis in CAPRIE



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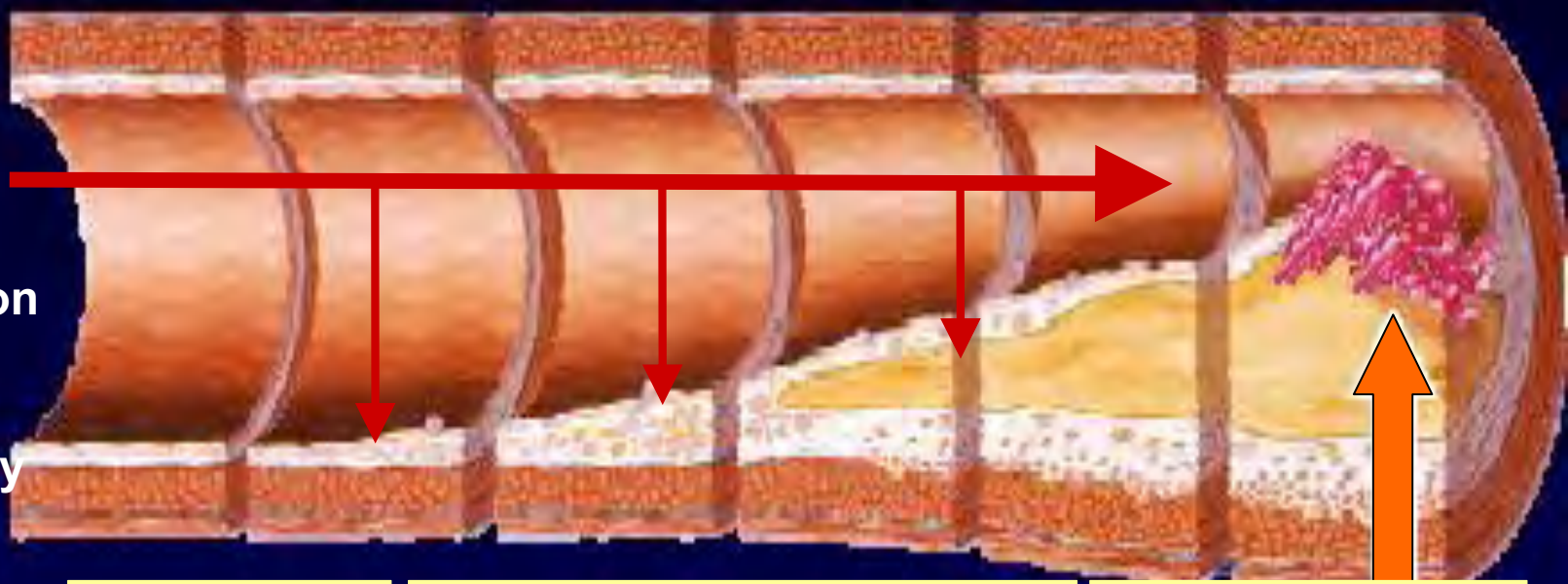
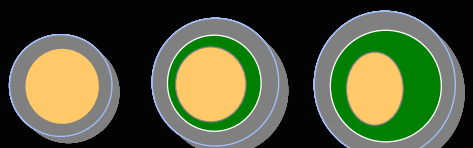
Atherosclerotic disease

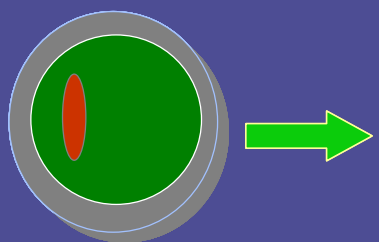
RISK FACTORS

Hyperlipemias
Diabetes
Hypertension
Chronic inflammation
Smoking
Obesity
Low physical activity
Stress
Sex
Age
Genetic factors



Normal vessel | Atherosclerosis | Atherothrombosis





Acute ischemic syndromes

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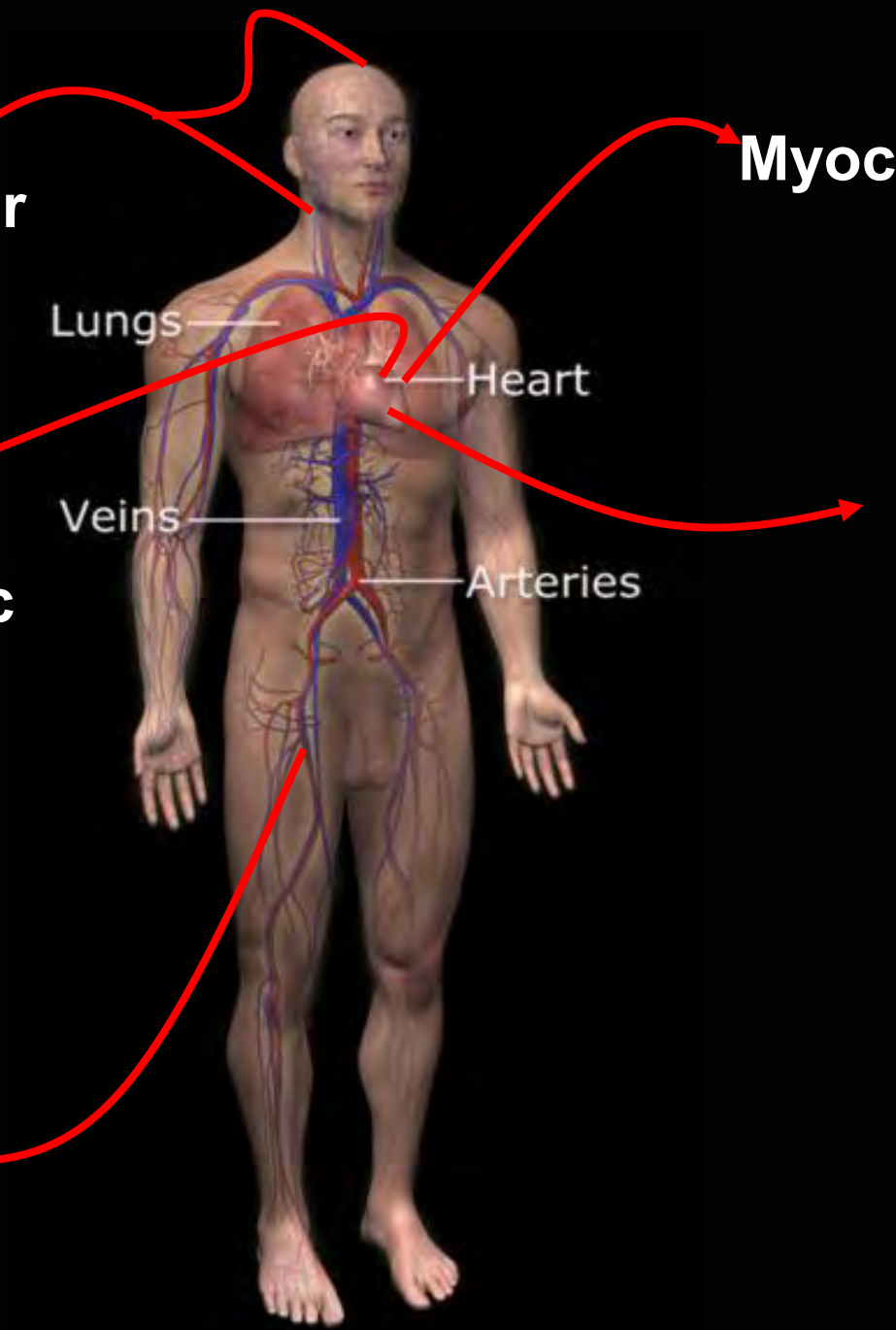
Consequences of atherothrombosis



Cerebrovascular accident

Myocardial Infarction

Sudden Cardiac Death

Chronic Ischemic Heart Disease

Gangrene of the legs

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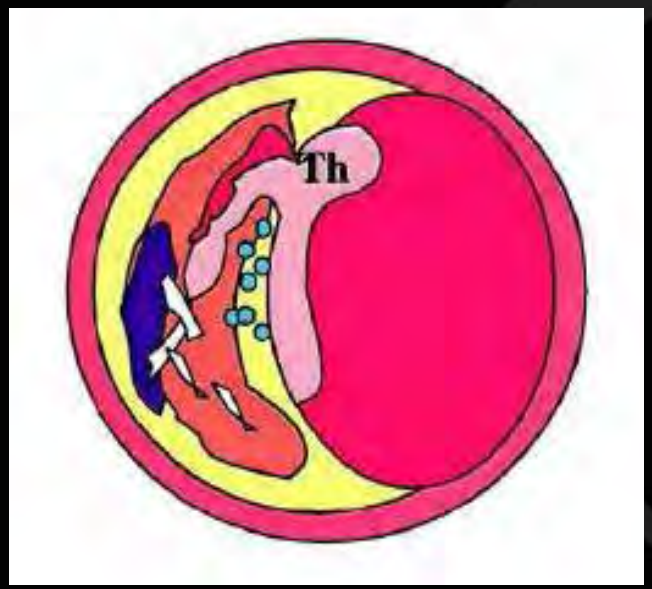
Atherothrombosis

Clinical manifestations are the result of focal atherosclerotic plaque rupture with superimposed thrombus

rupture



- ↑ necrotic core /thin fibrous plaque
- ↑ lipid core
- ↑ inflammatory cells
- ↓ smooth muscle cells
- ↓ collagen fibers

structural failure

↕

plaque DISRUPTION

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Atherothrombosis

Acute Coronay Syndromes

- Contenders:
  - Platelets
  - White cells & RBCs
  - EPCs
  - Inflammation
  - Microparticles

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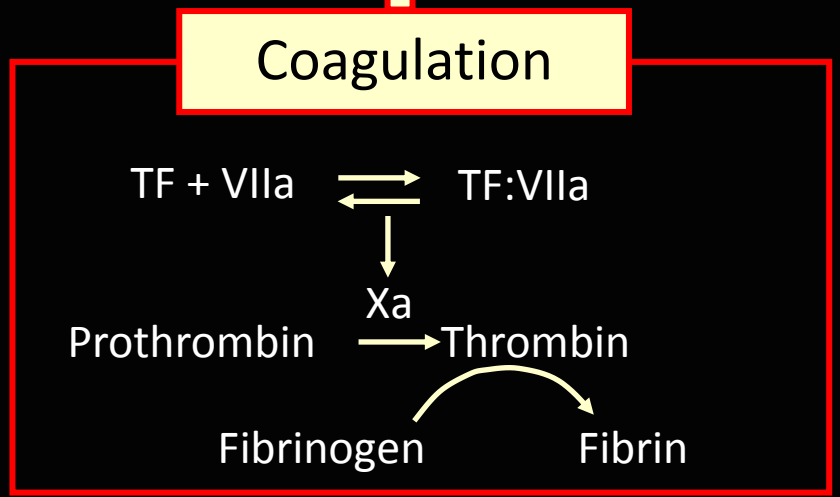
Composition of the Thrombus induced by Rupture of Atherosclerotic Plaques

Erythrocytes

Platelets

Fibrin

platelet



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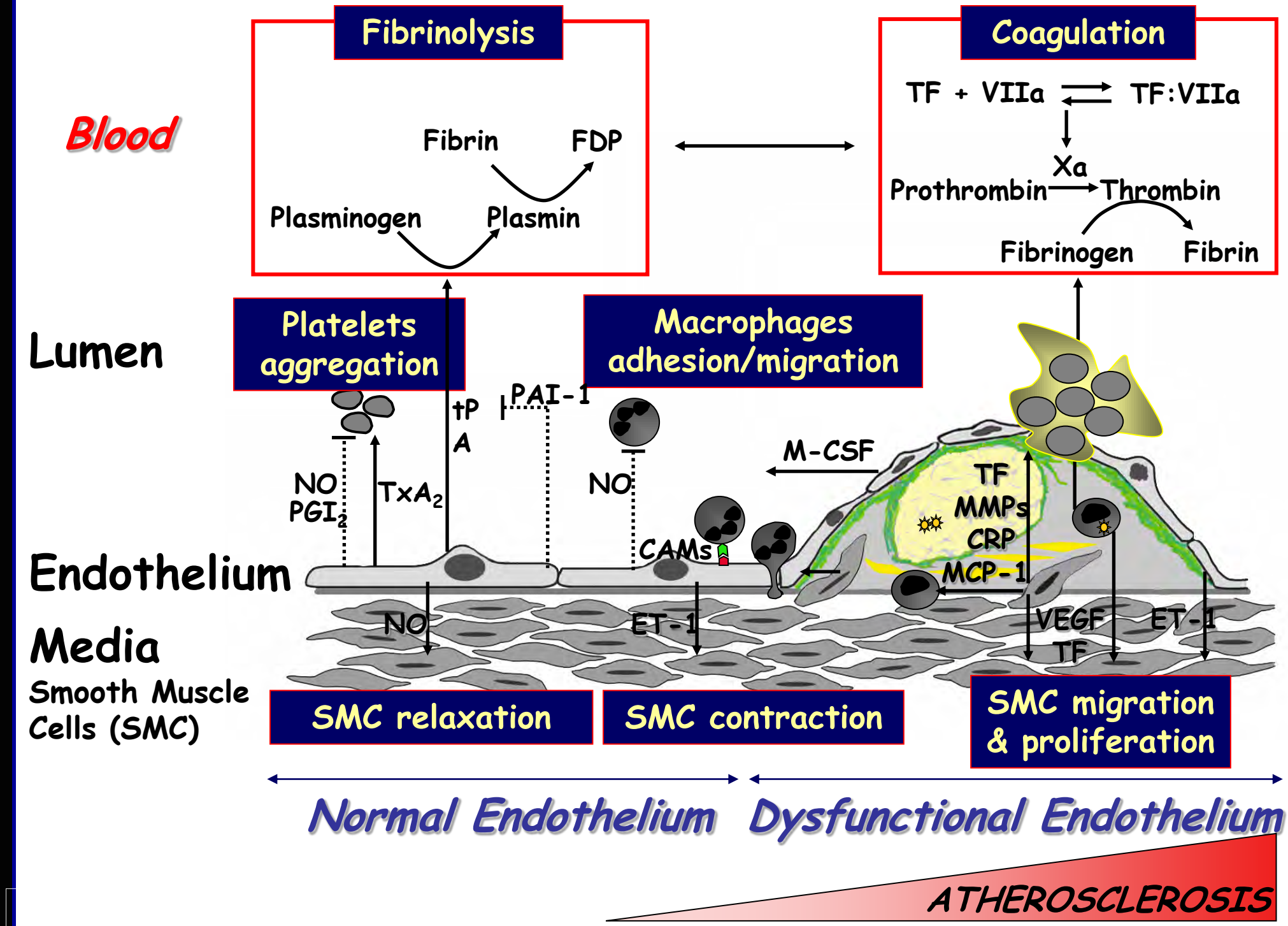
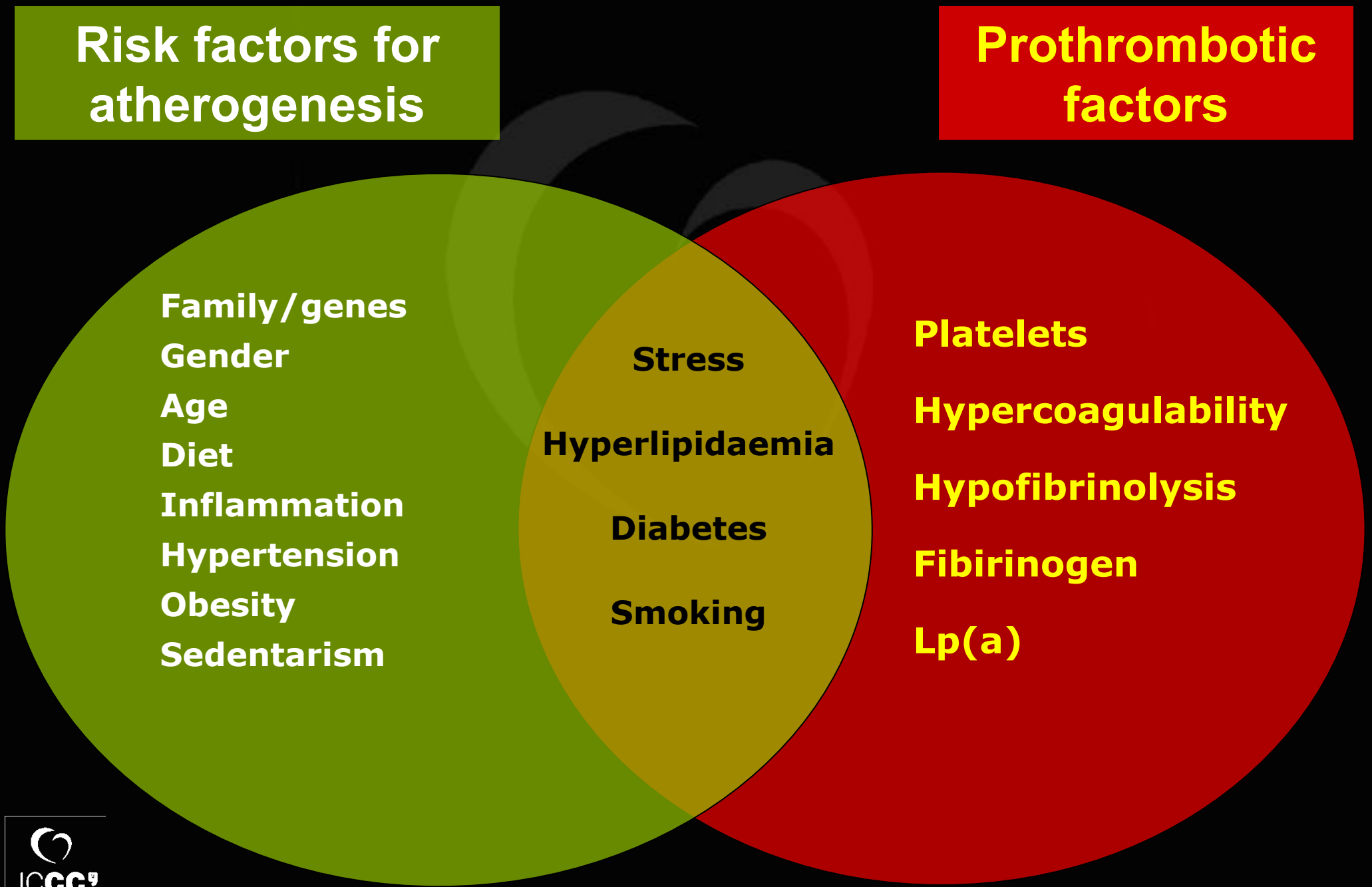
JACC 1994, Circulation 1997, Circulation 1999



Badimon L, Badimon JJ, & Vilahur G, et al *Pathophysiol Haemost Thromb, 2002*

Progress in understanding atherothrombosis



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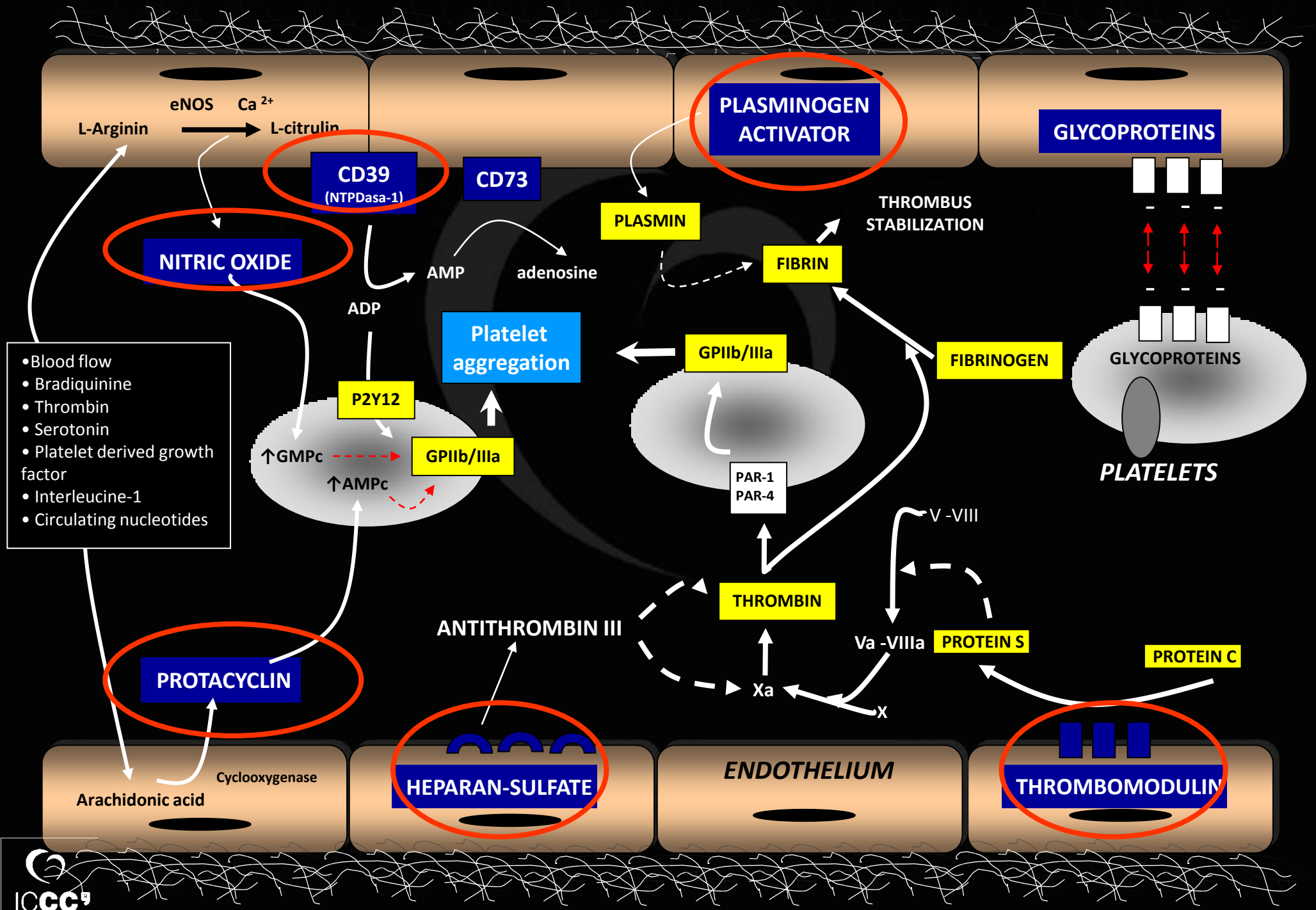
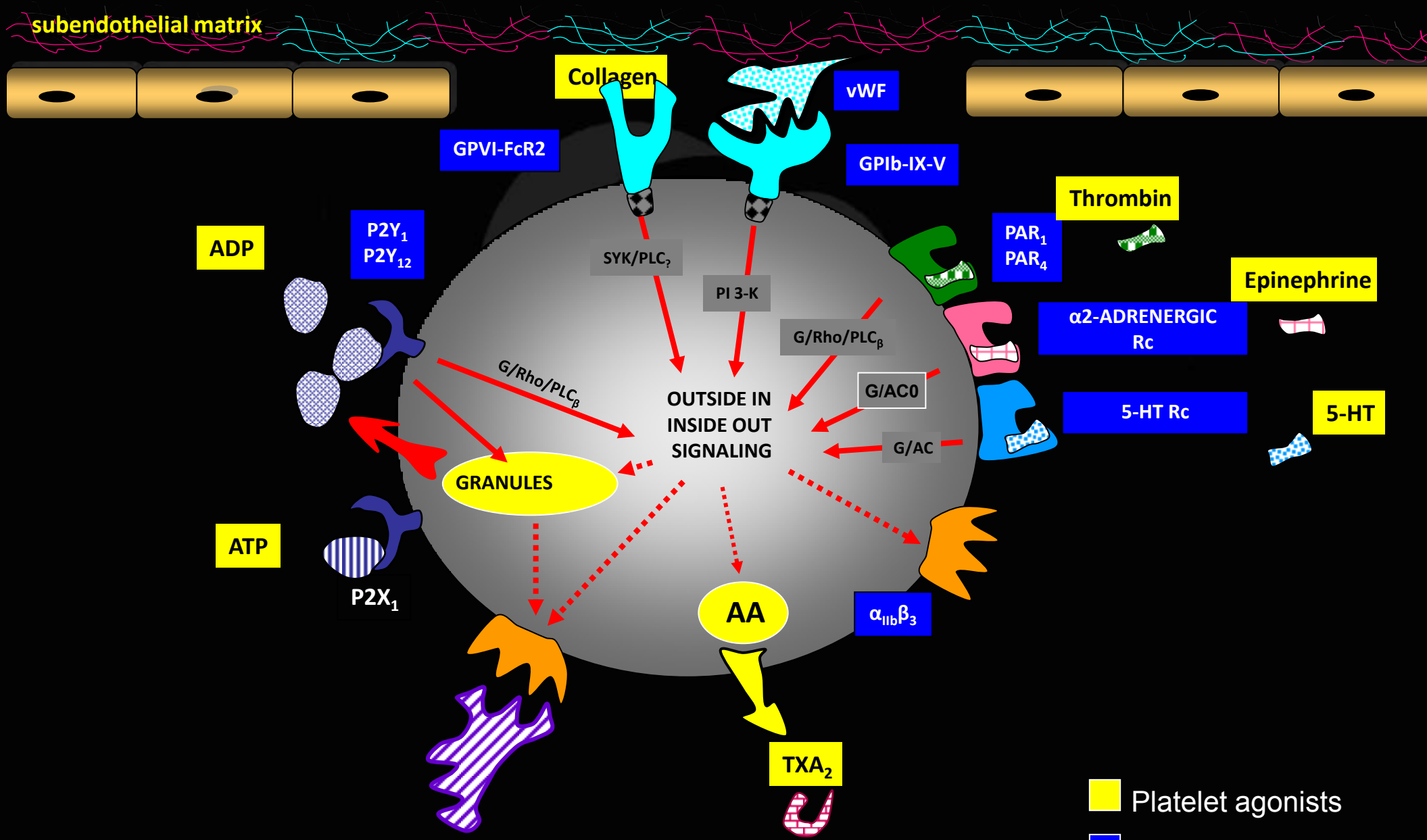
Endothelial antithrombotic properties

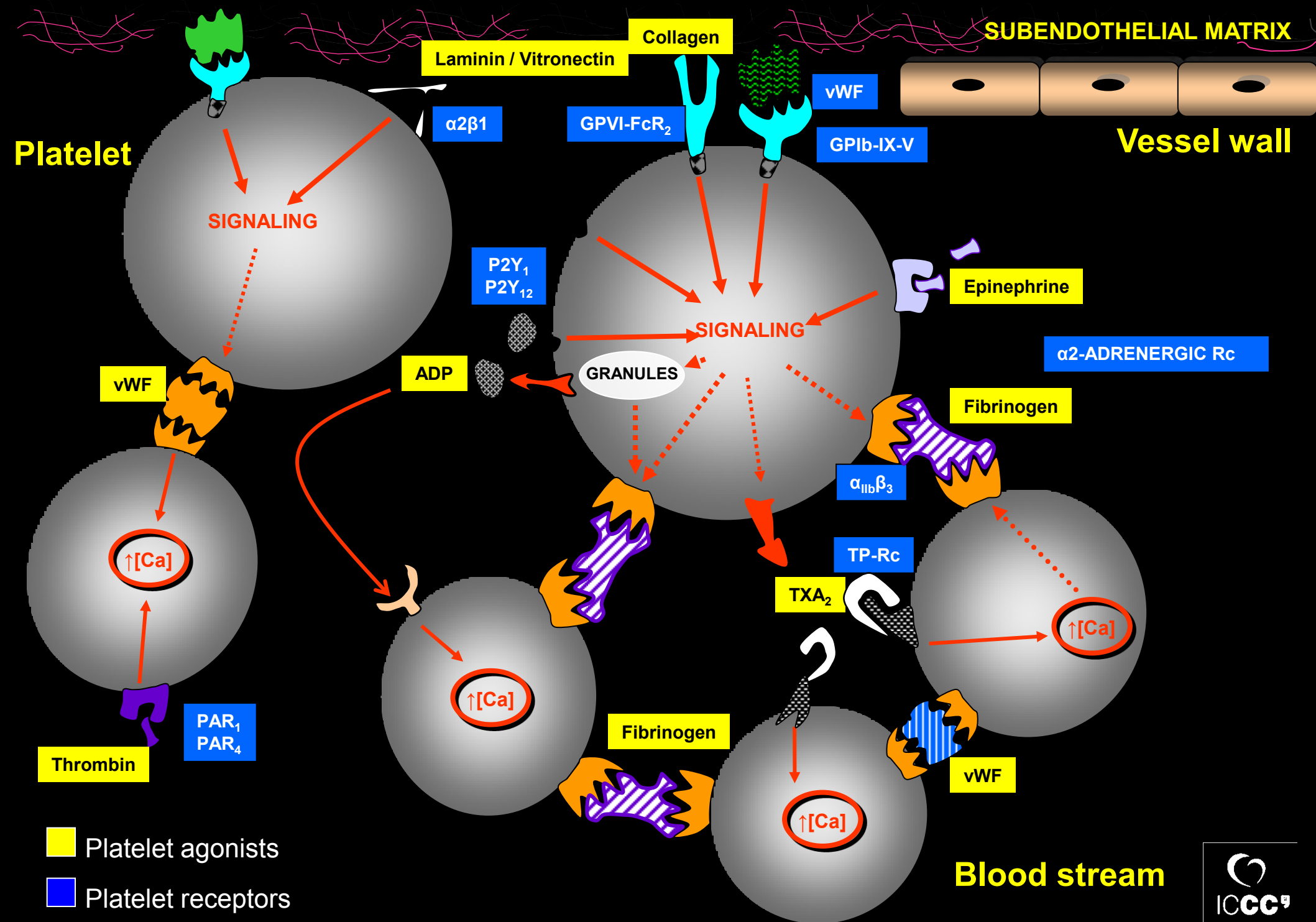


Badimon L & Vilahur G. *Rev Esp Cardiol, 2008*

Thrombus formation: Platelet adhesion



Thrombus formation: Platelet aggregation



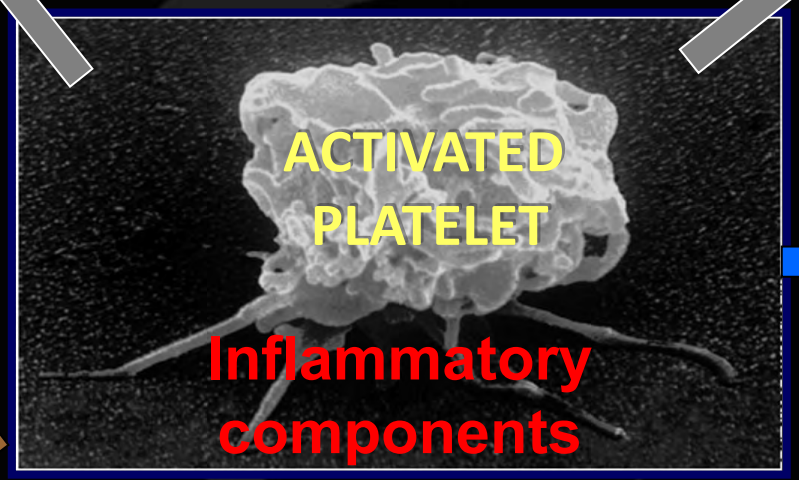
Platelet components

ADHESION PROTEINS

Fibrinogen
Fibronectin
vWF
Thrombospondin
Vitronectin
P-selectin
GPIIb/IIIa

COAGULATION FACTORS

Factor V and XI
PAI-1
Plasminogen
Protein S
GAS 6



GROWTH FACTORS

PDGF
TGF-β
EGF
bFGF

CYTOKINE-LIKE FACTORS

β-thromboglobulin
IL-1β
CD40L

CHEMOKINES

RANTES
MIP-1α
PDGF
PF4
IL-8
ENA-78

ENDOTHELIUM

Monocyte recruitment

IL- 8
MCP-1

VCAM-1
ICAM-1
E-selectin
ROS
TF
IL- 6

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Badimon L, Fernández-Ortiz A, Vilahur G. *Informa Healthcare; 2008*

Composition of the Thrombus induced by Rupture of Atherosclerotic Plaques

Erythrocytes

Platelets

Fibrin

platelet

Coagulation

TF + VIIa ⇌ TF:VIIa

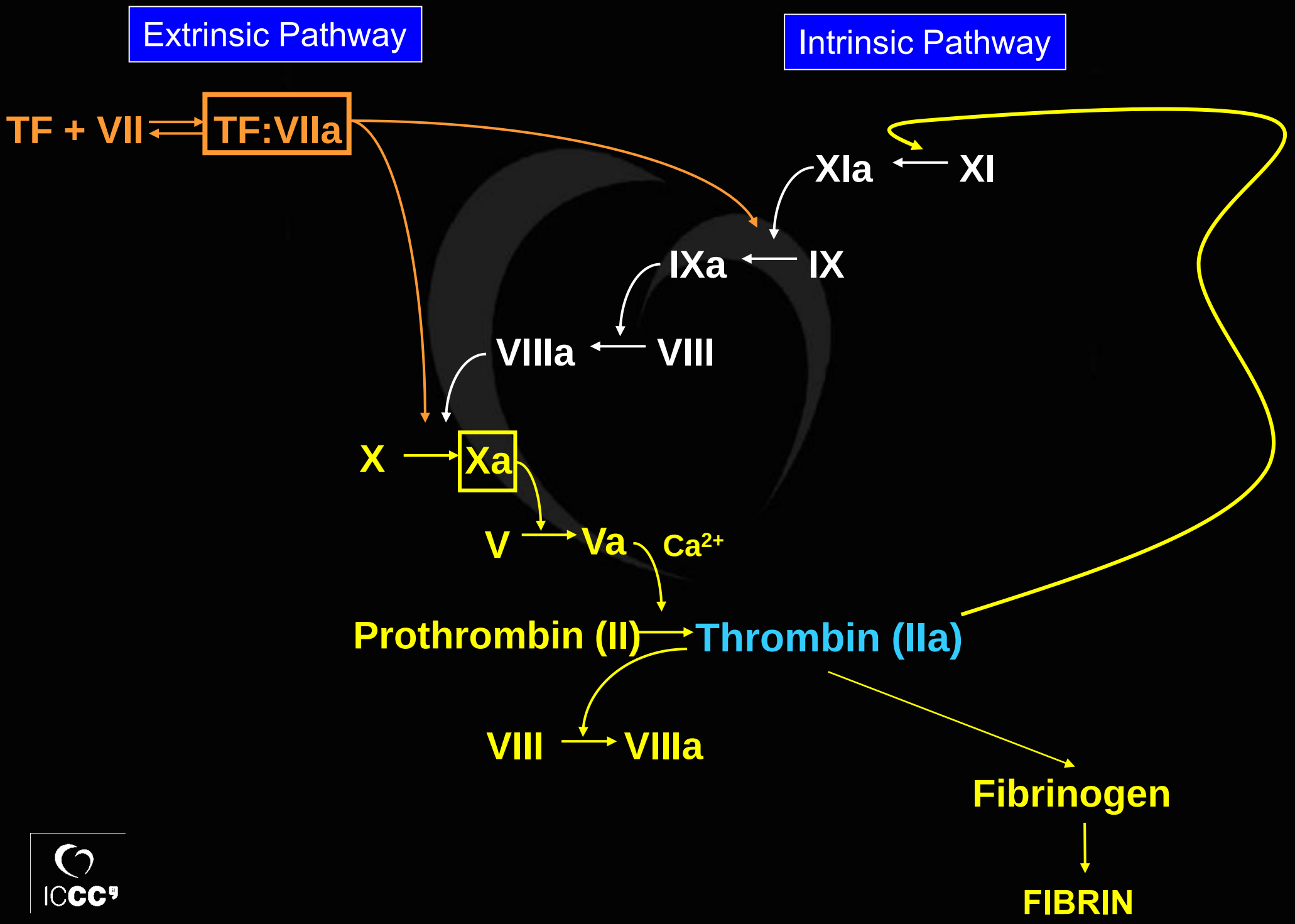
↓ Xa

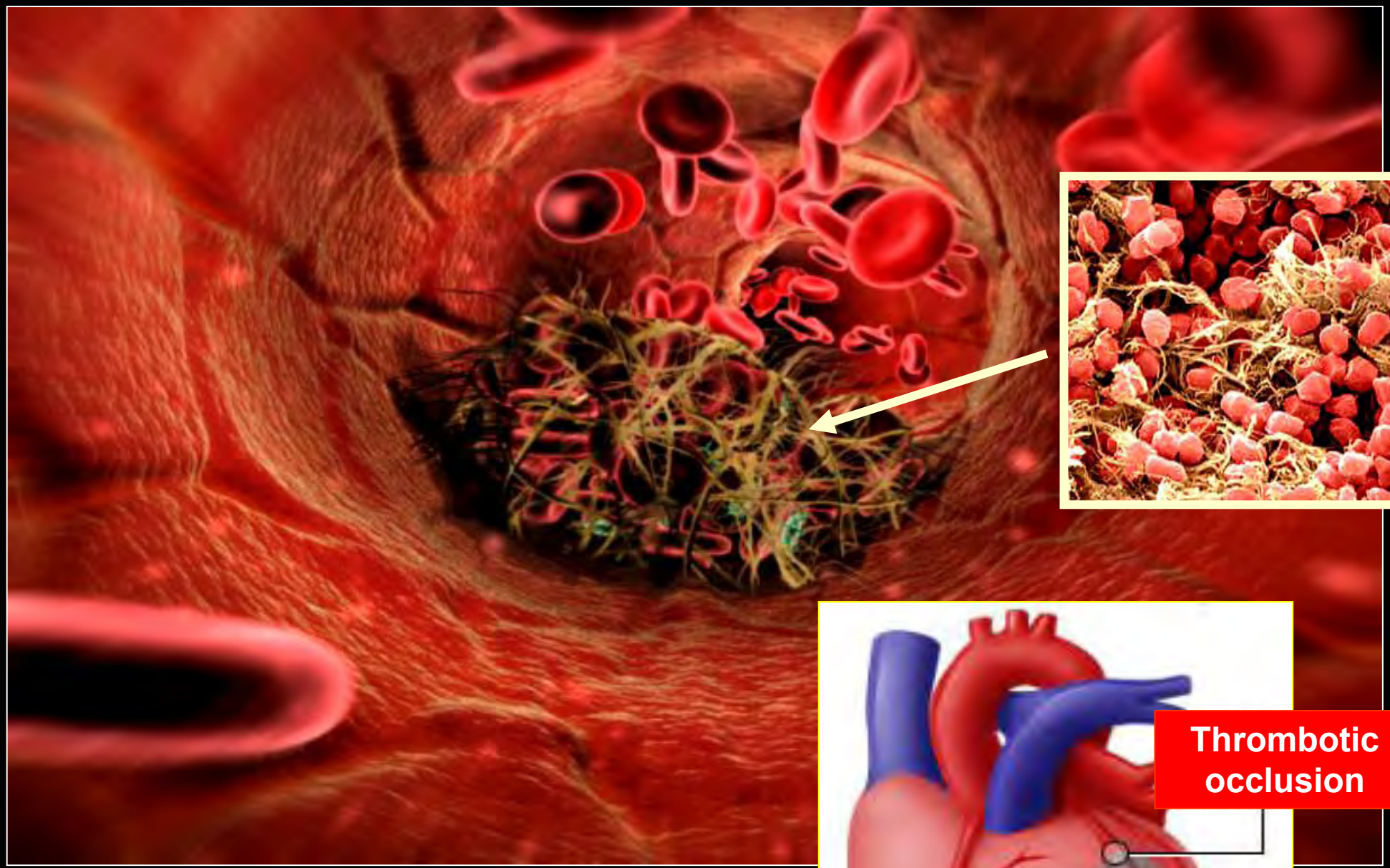
Prothrombin → Thrombin

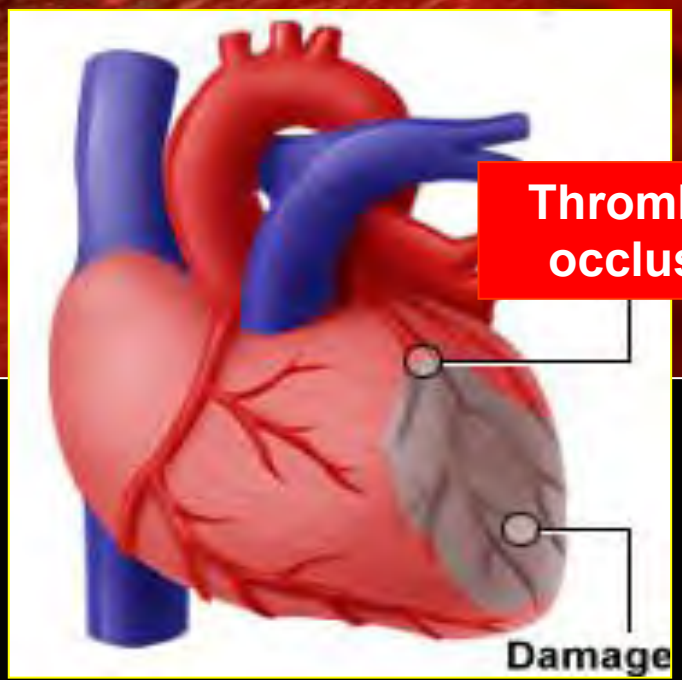
Fibrinogen → Fibrin

JACC 1994, Circulation 1997, Circulation 1999

Coagulation cascade







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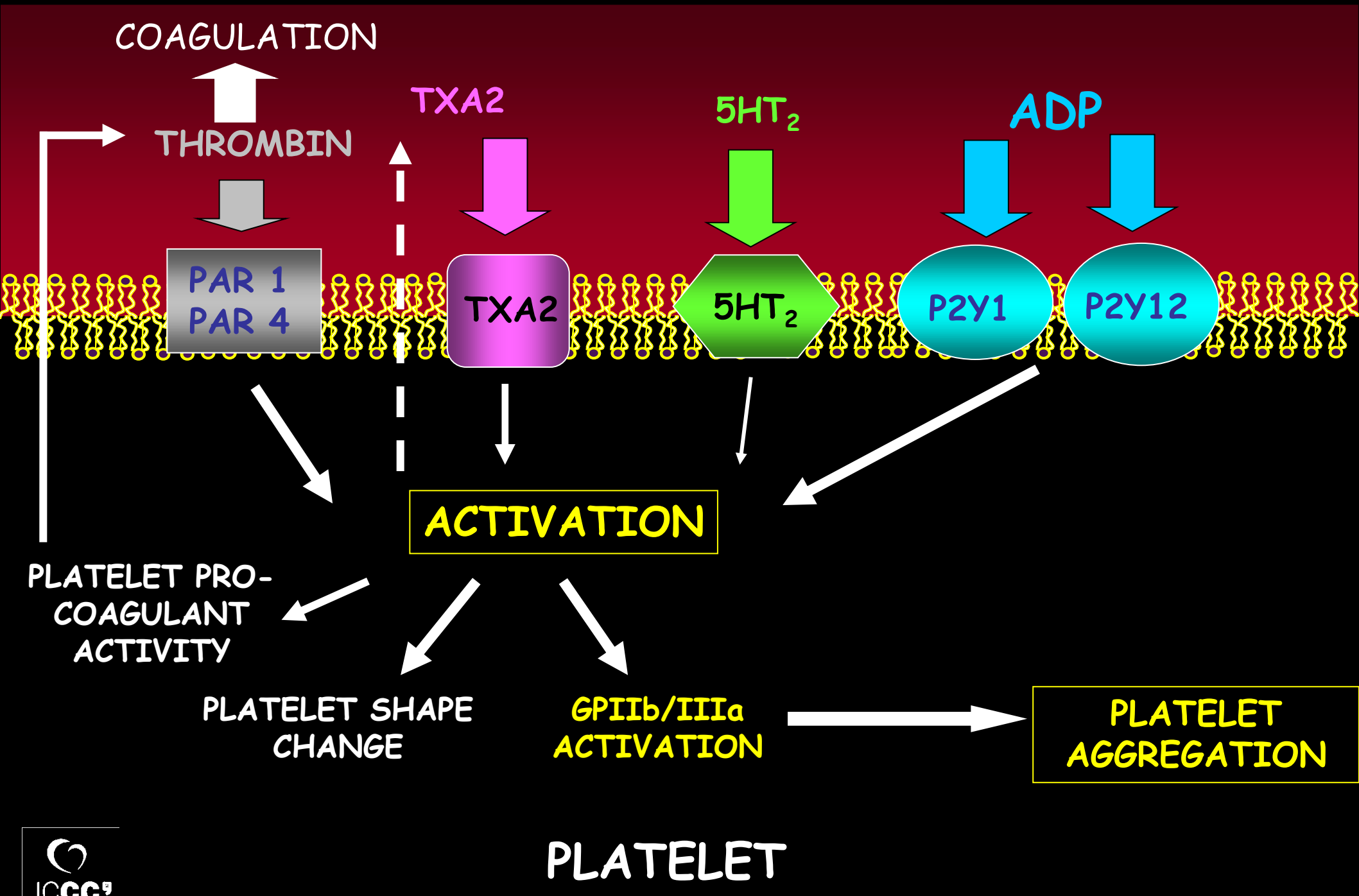
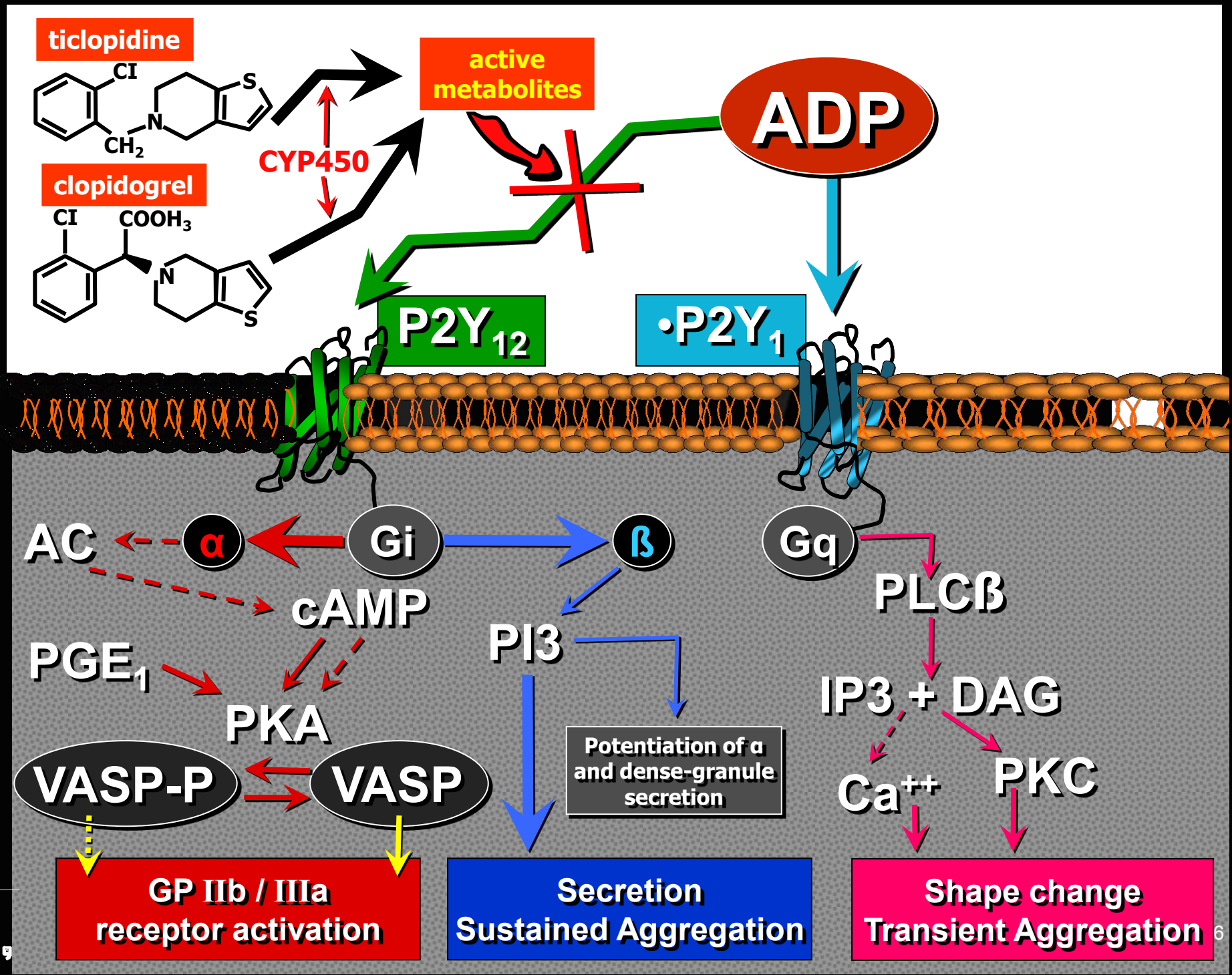
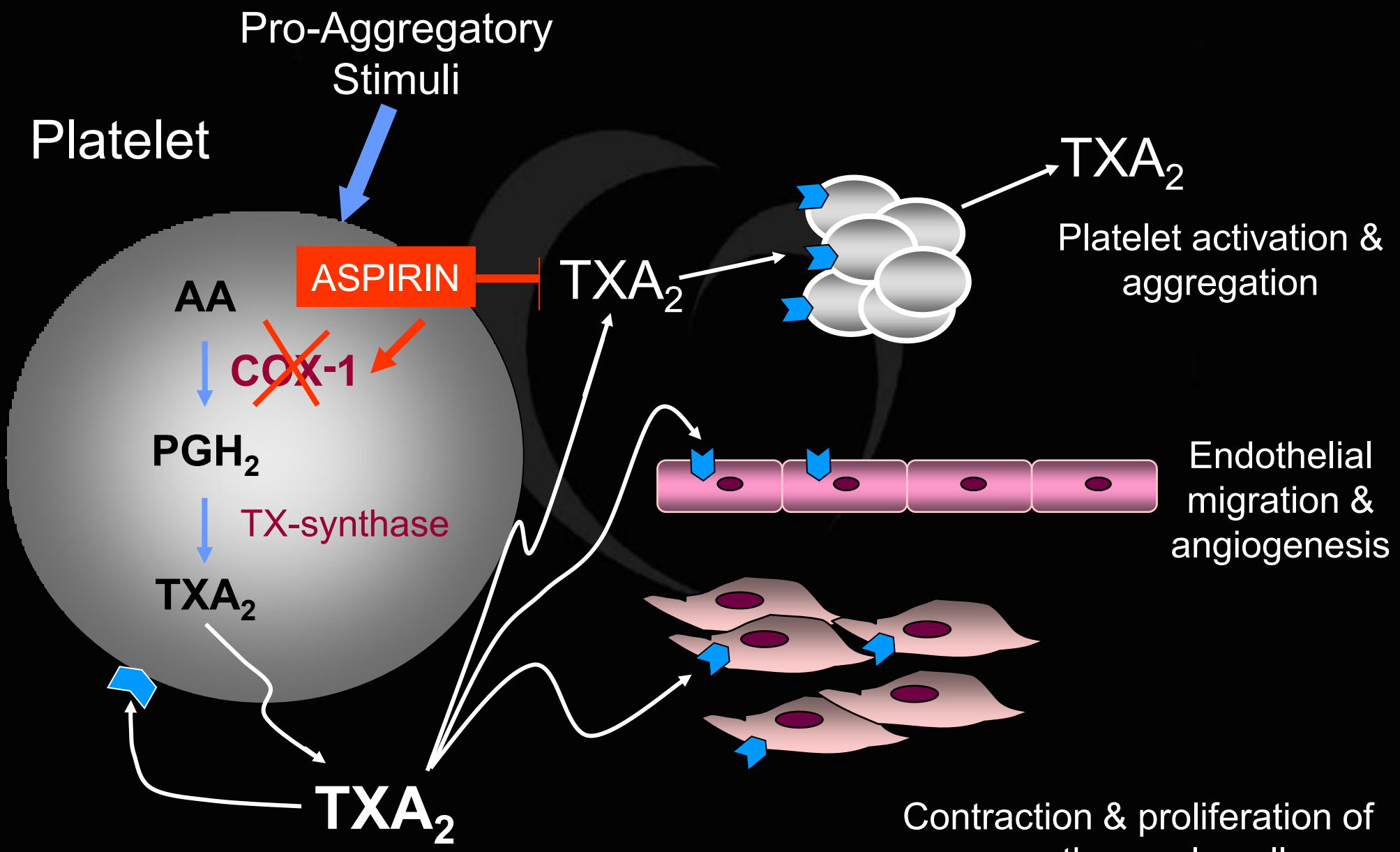
Major agonists & receptors for platelet activation



ADP & platelet P2 receptors



TXA_2 & platelet TP receptors



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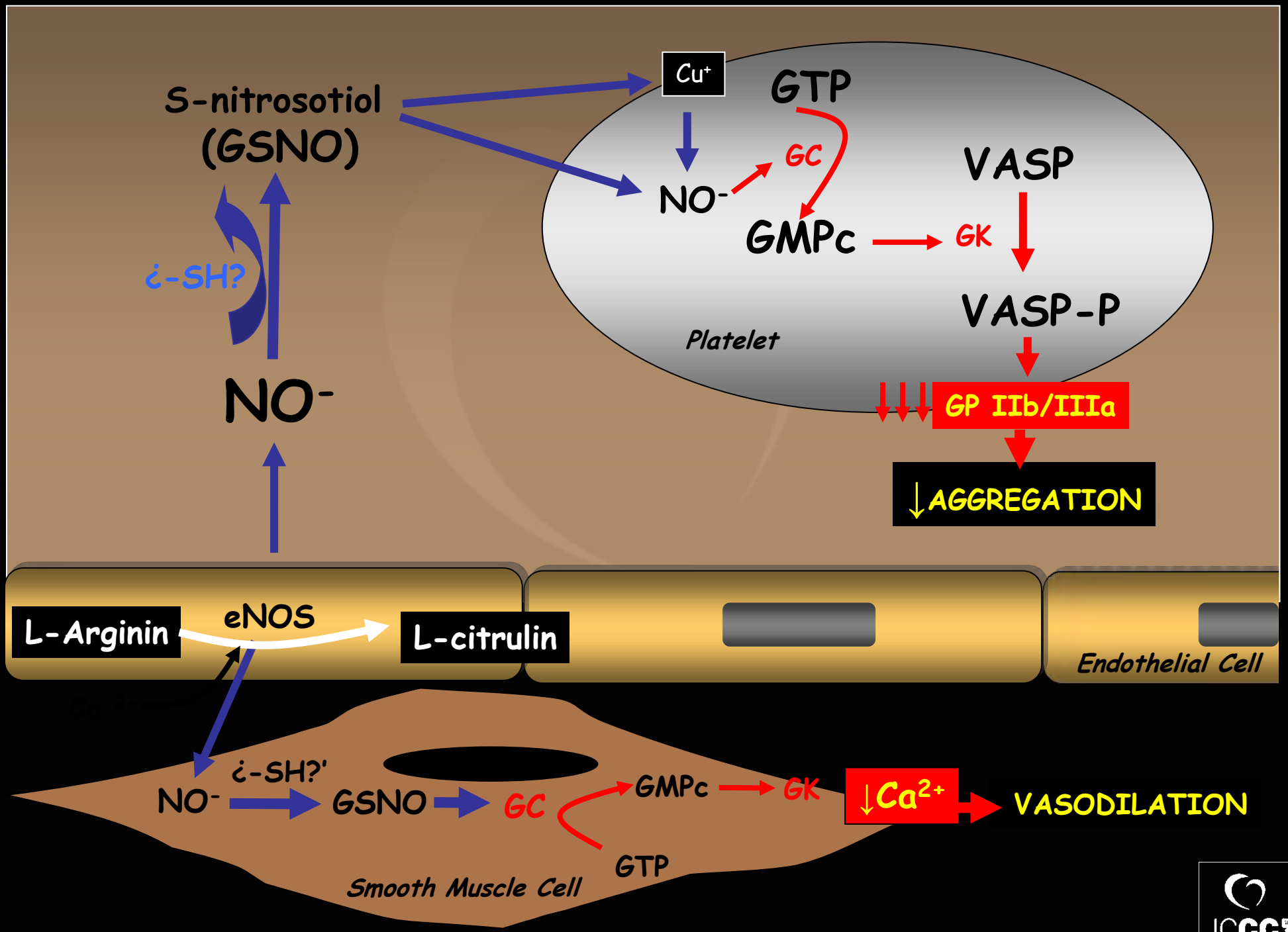
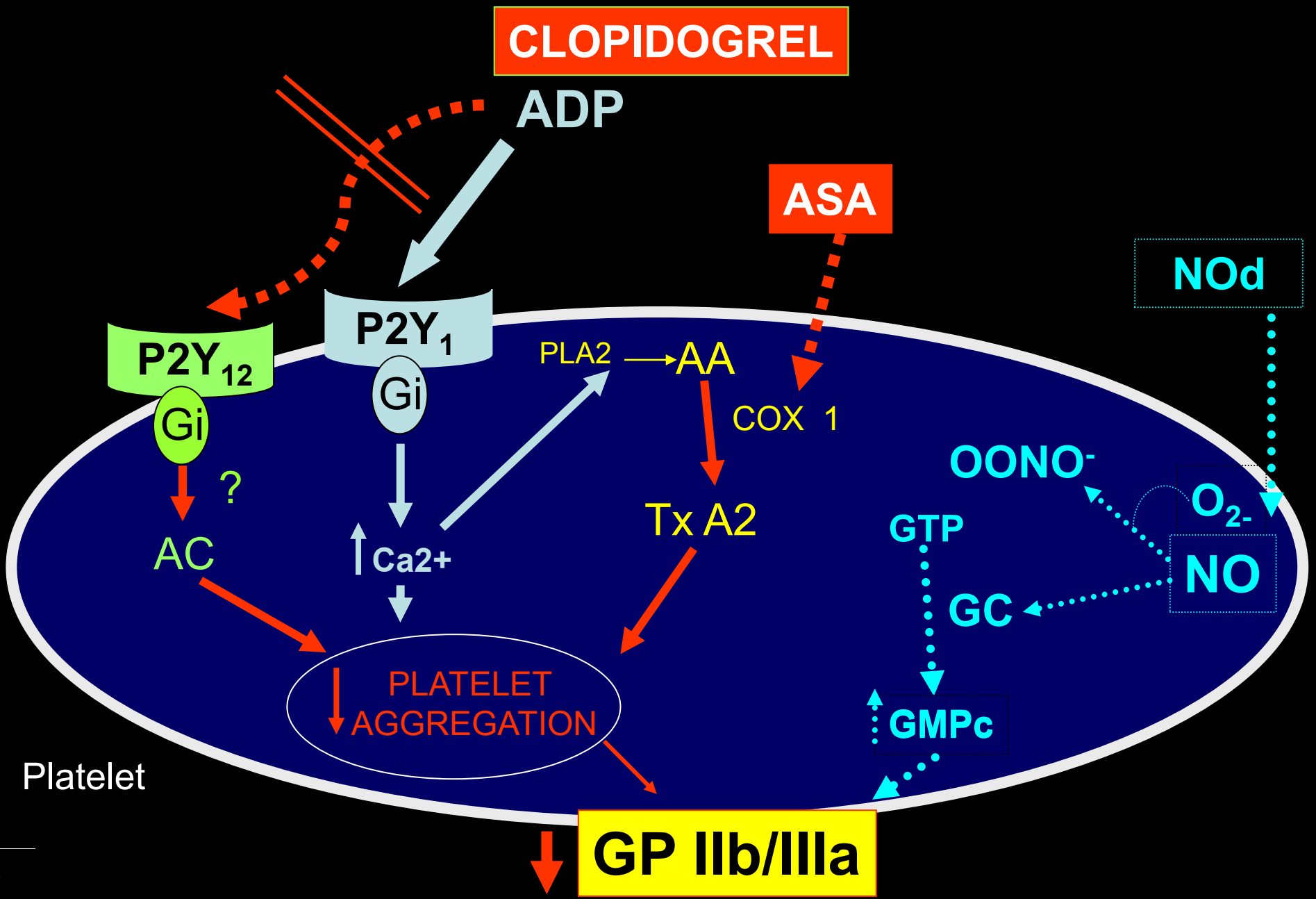
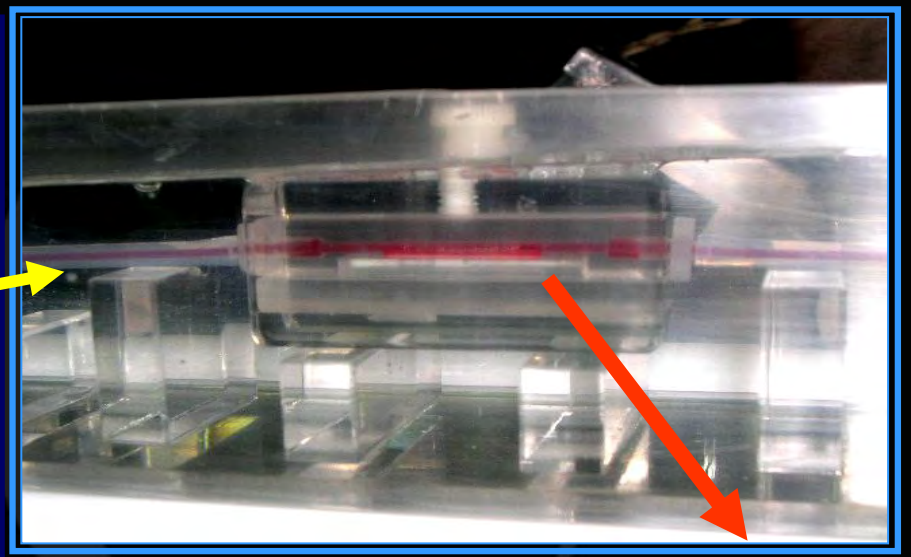
Nitric Oxide (NO)



Novel anti-platelet approaches:
Nitric Oxide (NO) donation

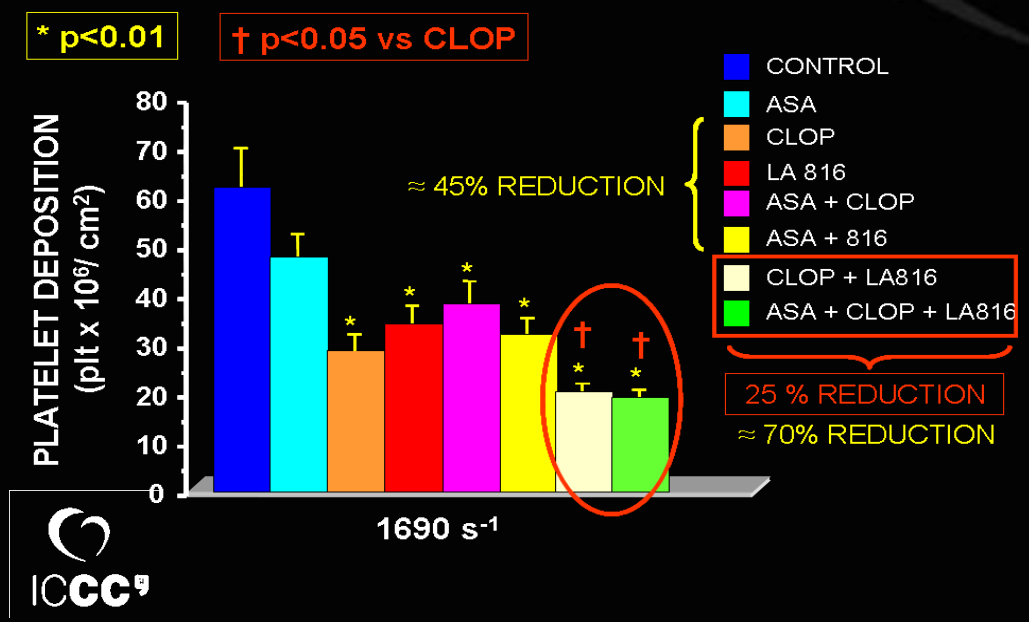


Effect of exogenous NO donation with a novel-nitrosothiol compound in thrombotic risk

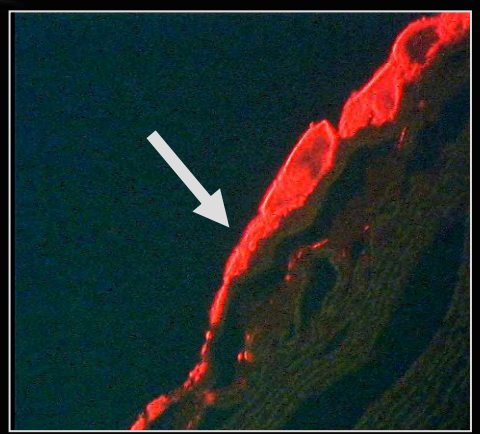


Badimon perfusion chamber

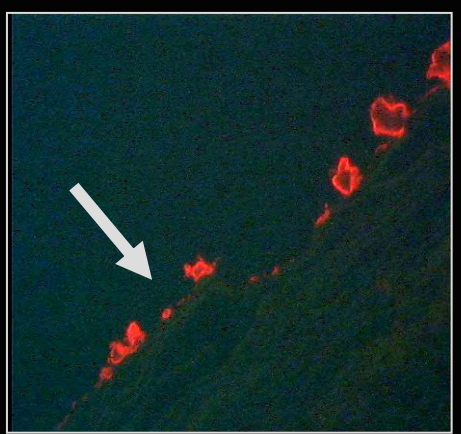
Platelet deposition



control

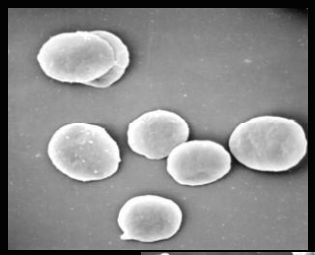


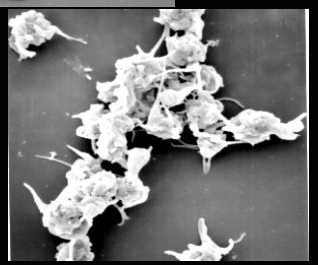
NO donor



Vilahur G et al. Circulation 2004

Proteomic Analysis





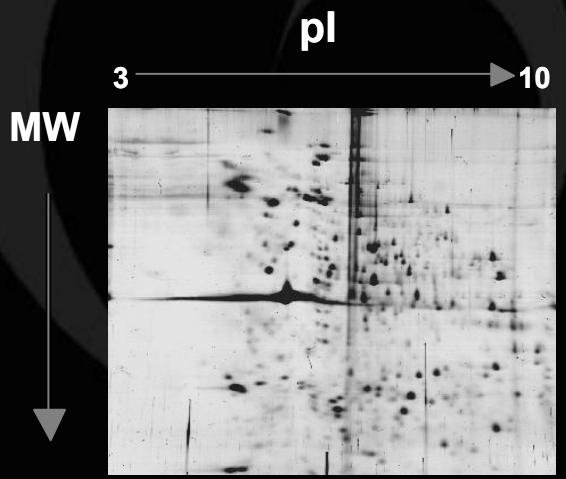
protein extracts

IEF

PAGE

2D electrophoresis

protein separation



analysis differential proteomic pattern

MALDI-ToF-ToF

protein identification / characterization

fingerprinting

LC/MS/MS

Linear ion Trap

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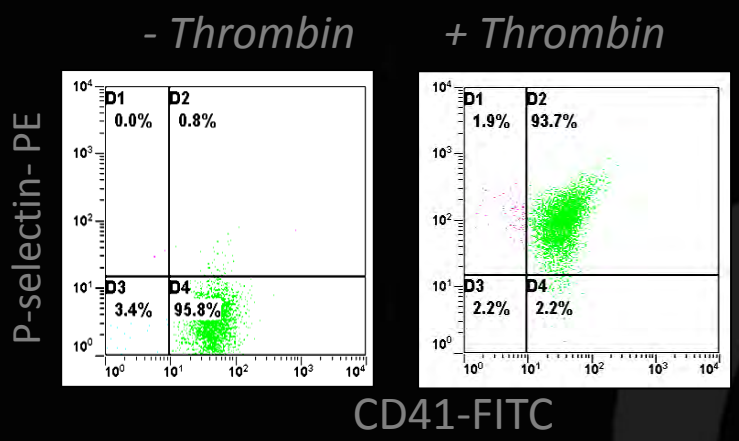
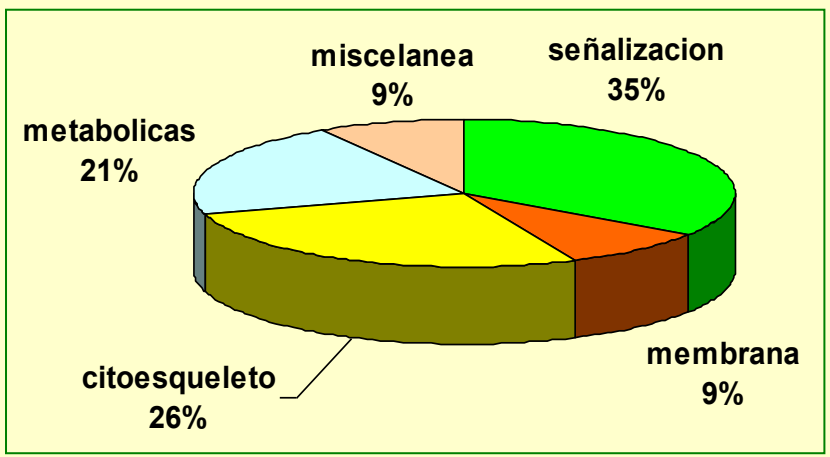
Differential Protein Pattern in Thrombin stimulated Platelets

Effect of chronic treatment with NOd



NOd reverts the proteomic profile of 24 different proteins in Thrombin activated Platelets



NUMBER	NAME	ACCESS NUMBER	MW THEORIC	COVERAGE	FUNCTION
5701	Gelsolin	P20305	76	12.4 %	CYTOSKELETON
4471	ARP3	Q9GL80	47	19.6 %	
5163	Tropomyosin	Q8MKF3	34	31.6 %	
1114	MLC	P12829	22	19.8 %	
6506	Coronin	Q9ULV4	55	10.1 %	
6102	Tubulin	P02554	50	19.0 %	
1656	Filamin A	Q96C61	89	24.0 %	
3714	Advillin	O75366	92	14.3 %	
5513	Aldehyde dehydrogenase	P05091	55	13.6 %	METABOLISM
6643	G3DP	Q9TUY4	69	14.3 %	
7276	Esterase D	Q9GJT2	32	22.3 %	
1767	Valosin containing protein	P03974	89	18.1 %	
6113	Ubiquitation factor E4A	Q14139	33	12.5 %	
3312	G protein beta 1	O45040	39	21.2 %	SIGNALING
	Casein kinase II	Q61177	48	12.2 %	
	Voltage dependent Ca channel	O00555	47	19.8 %	
	Voltage dependent annion channel	Q29380	36	20.4 %	
	Grb2	P62993	25	30.9%	
	Rho GDI	P52565	23	26.0 %	
	PI3K	P42339	93	14.1 %	
	4-3-3 zeta	P63104	30	26.5 %	
	PDI A3	P30101	57	30.3 %	
	SNAP	Q9LXZ5	43	24.7 %	
	HSP70	Q922P8	71	38.4 %	

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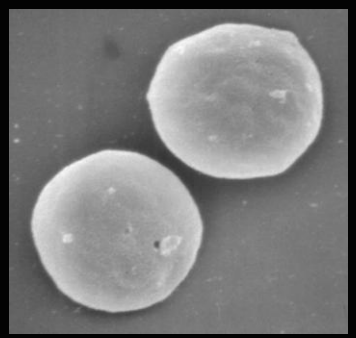
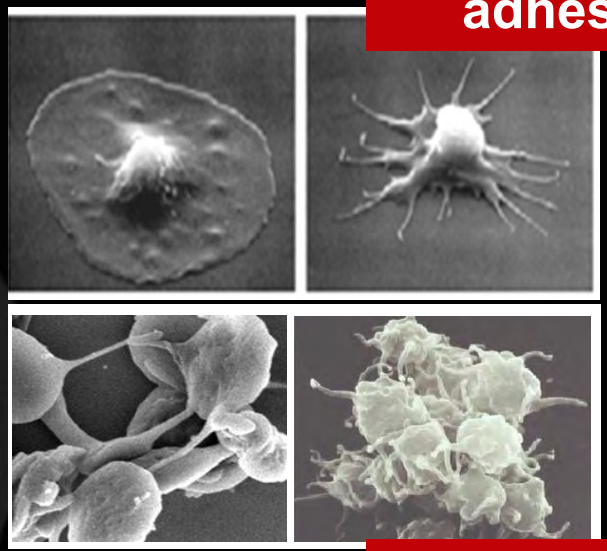
Padro T, Peña E, Molins B, Vilahur G, Badimon L . Eur Heart J -A- 2006

NO-donors affect proteins that modulate platelet cytoskeleton



Cytoskeleton modifications



adhesion

aggregation

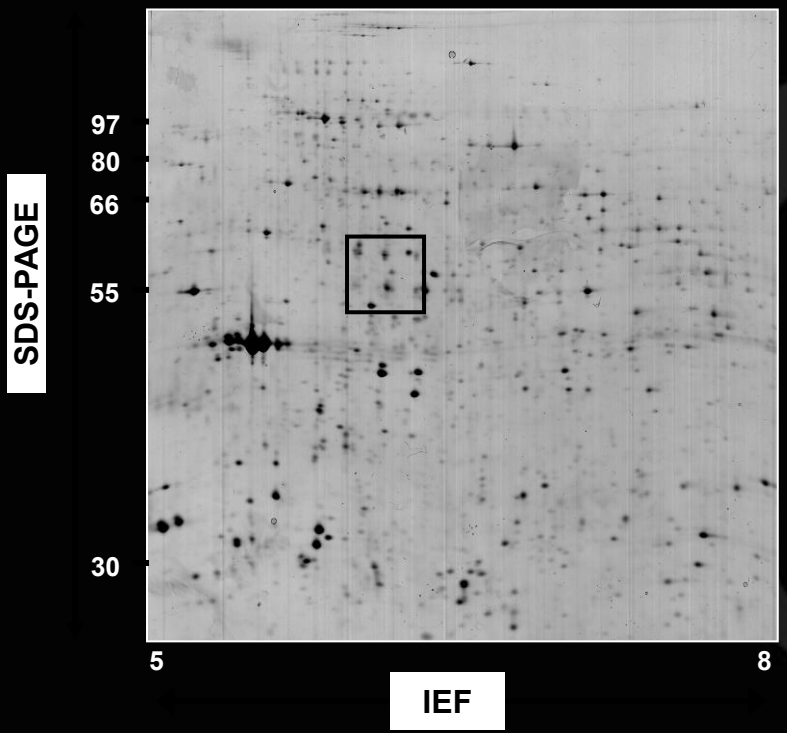
proteína	número Swiss-Prot	MWt	% cobertura
Advillin	O75366	92	14.3 %
Coronin	Q9ULV4	55	10.1 %
Gelsolina	P20305	76	12.4 %
Myosin Light Cha	P12829	22	19.8 %
Tropomyosin	Q8MKF3	34	31.6 %
ARP2/3	Q9GL80	54	19.6 %
Filamin A	Q96C61	71	24.0 %
b-Tubulin	P02554	33	19.0 %

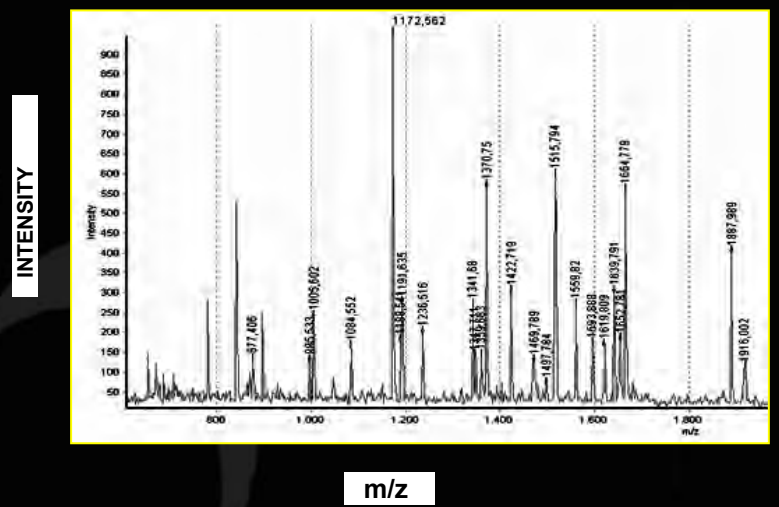
→ Actin fibers formation

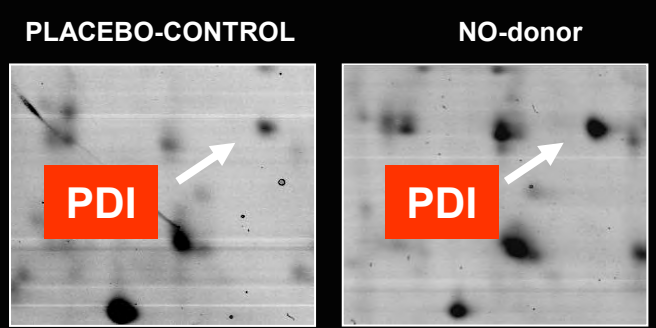
→ Actin fibers stabilization

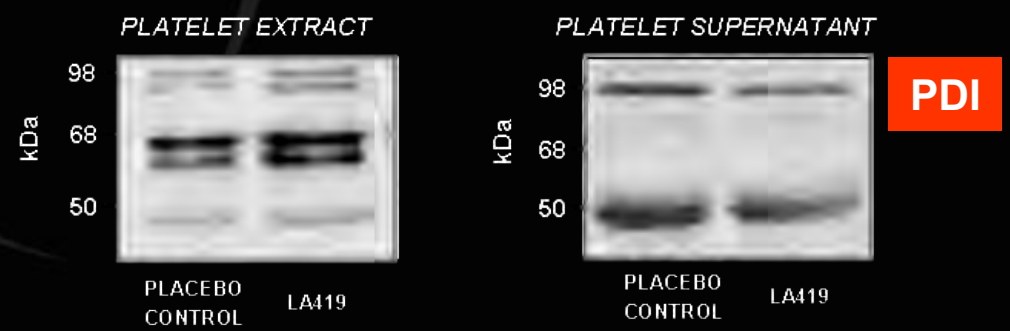
→ microtubuls

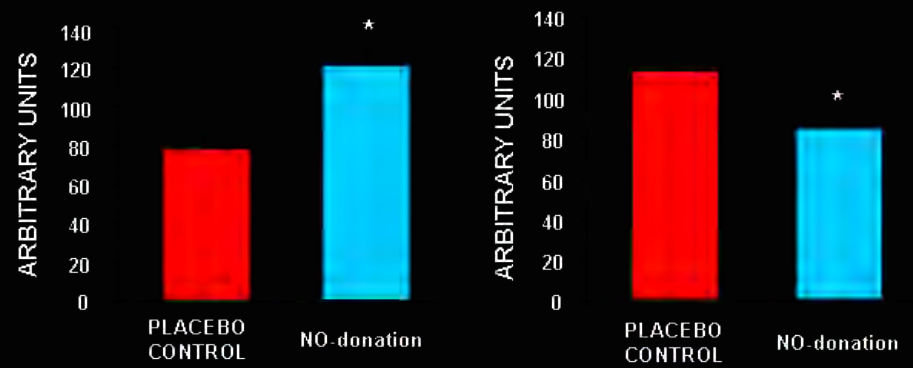
NO-donor effects on Protein-Disulphide Isomerase (PDI)





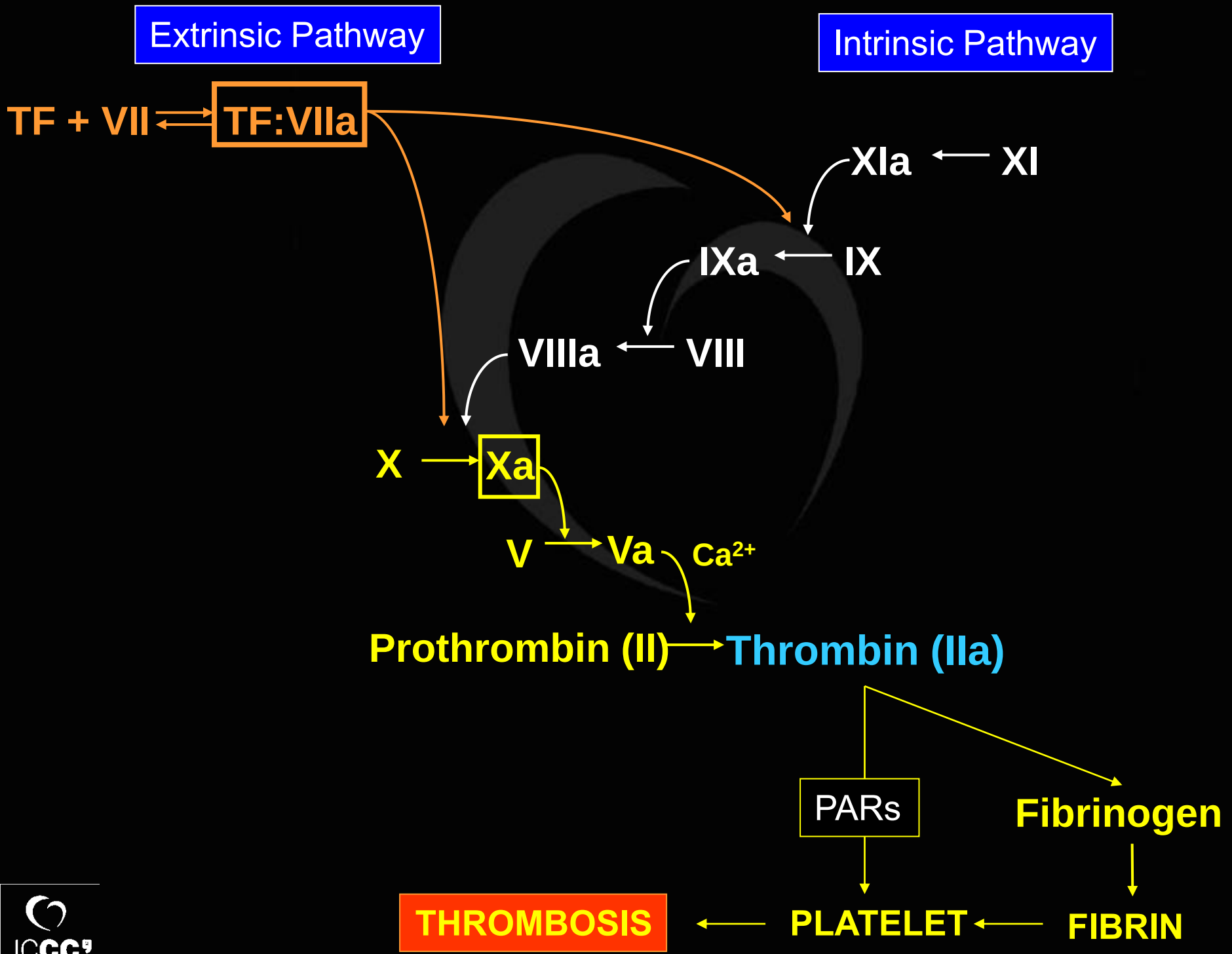




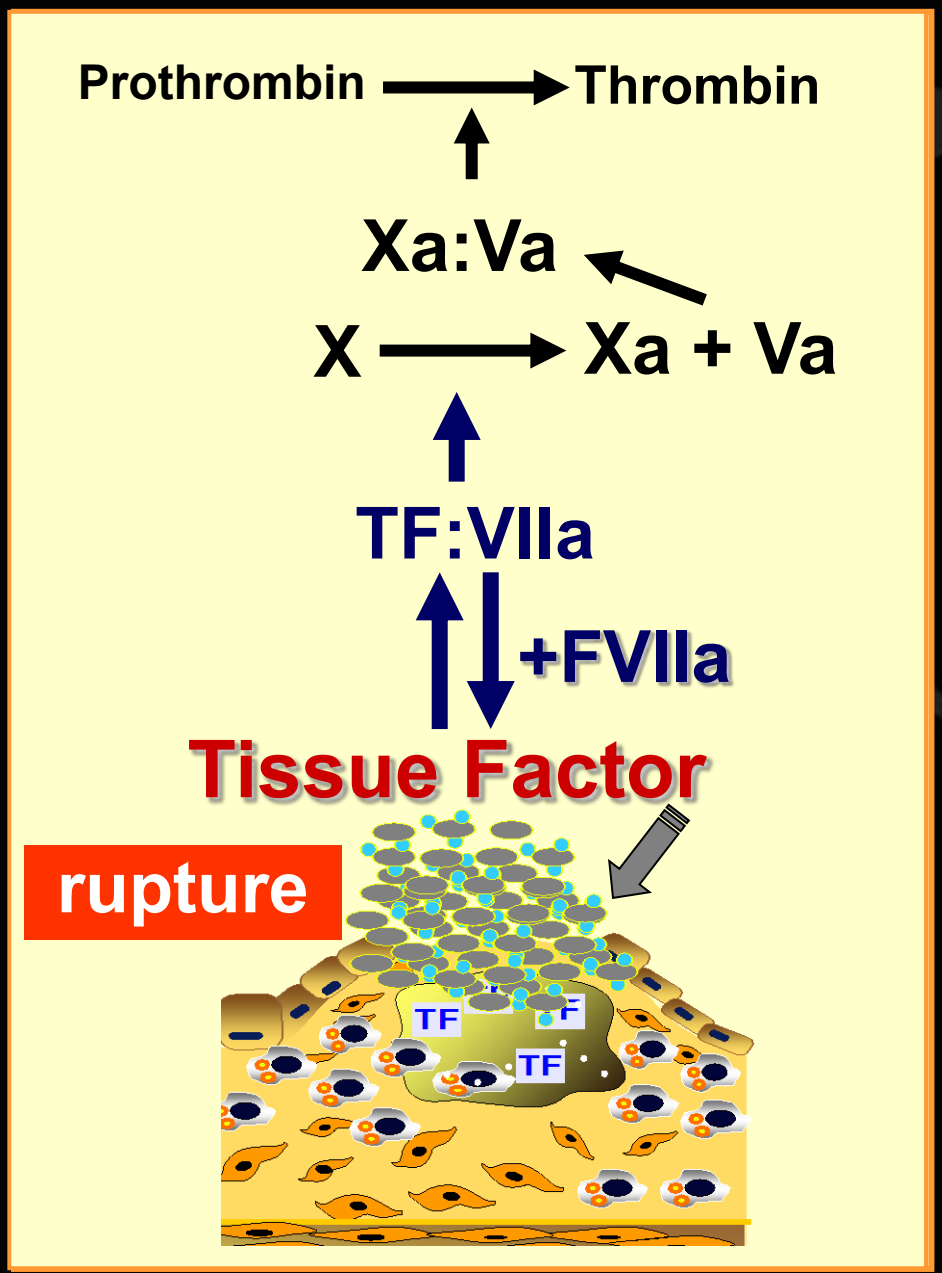


Vilahur G, Pena E, Padró, Badimon L. *Thromb Haemost 2007*

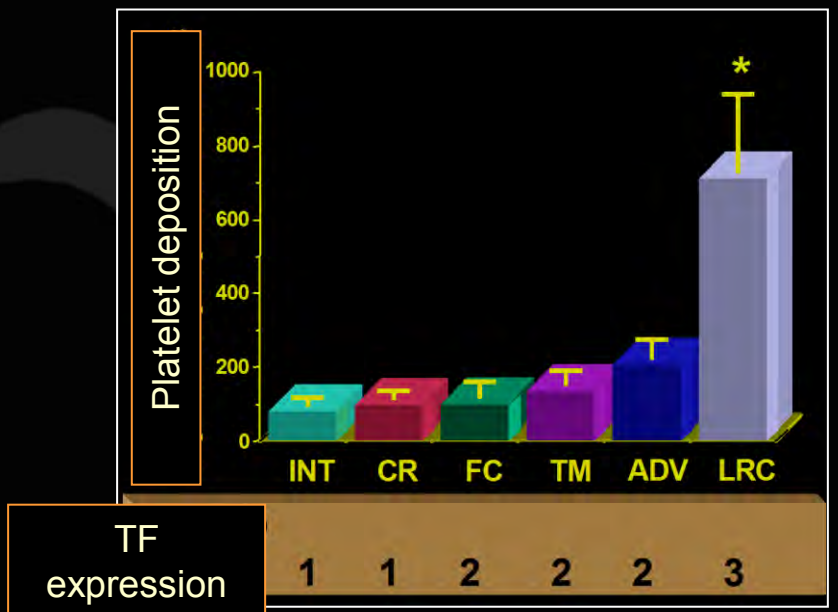
THROMBIN: Coagulation cascade



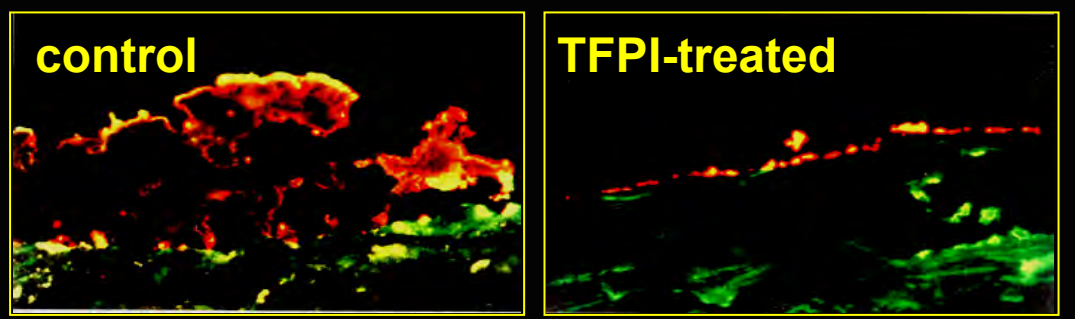
TF: thrombosis on atherosclerotic plaques



TF AND THROMBOGENICITY



Fernández-Ortiz, *Circulation 1994*
Toschi et al. *Circulation 1997*



Badimon et al. *Circulation 1999*

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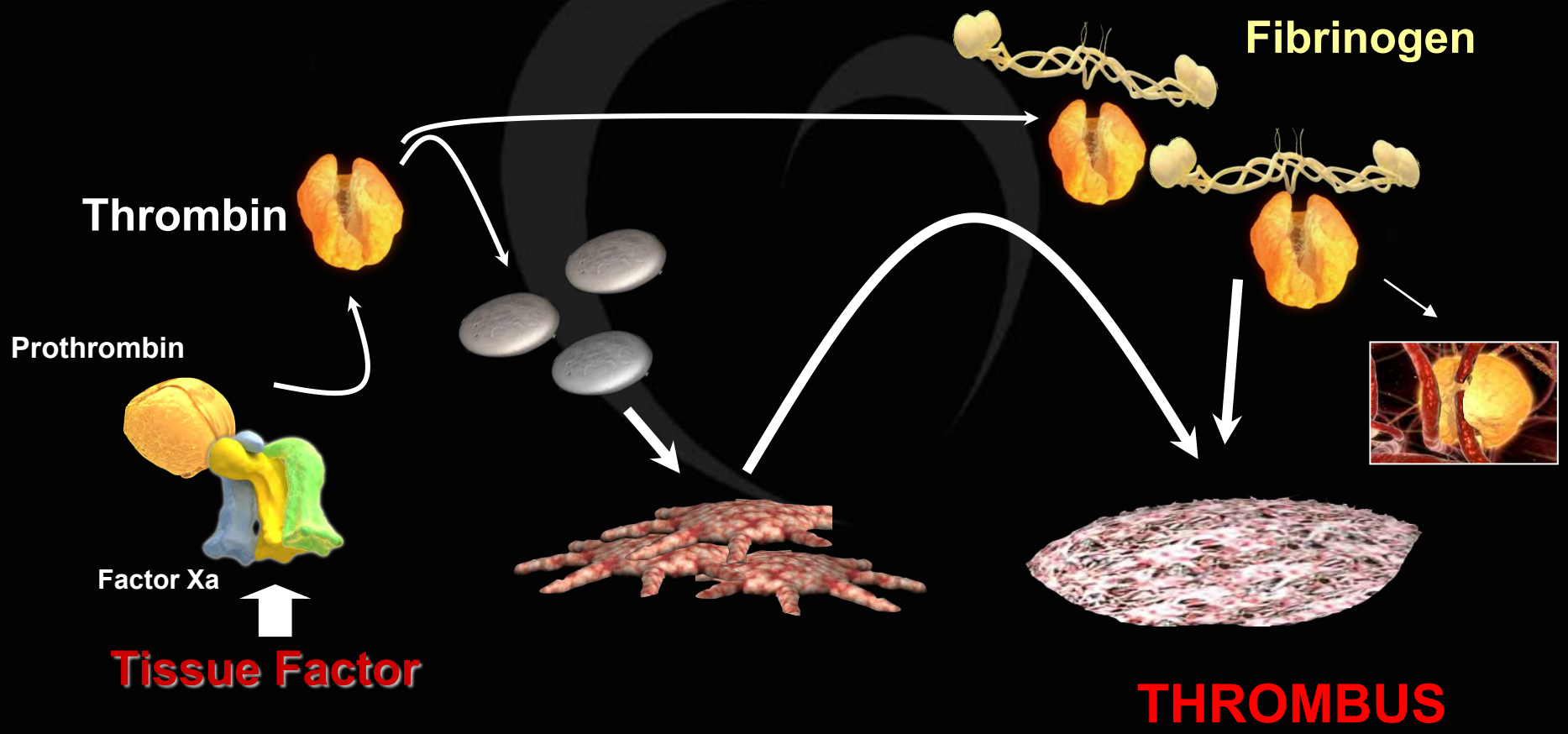
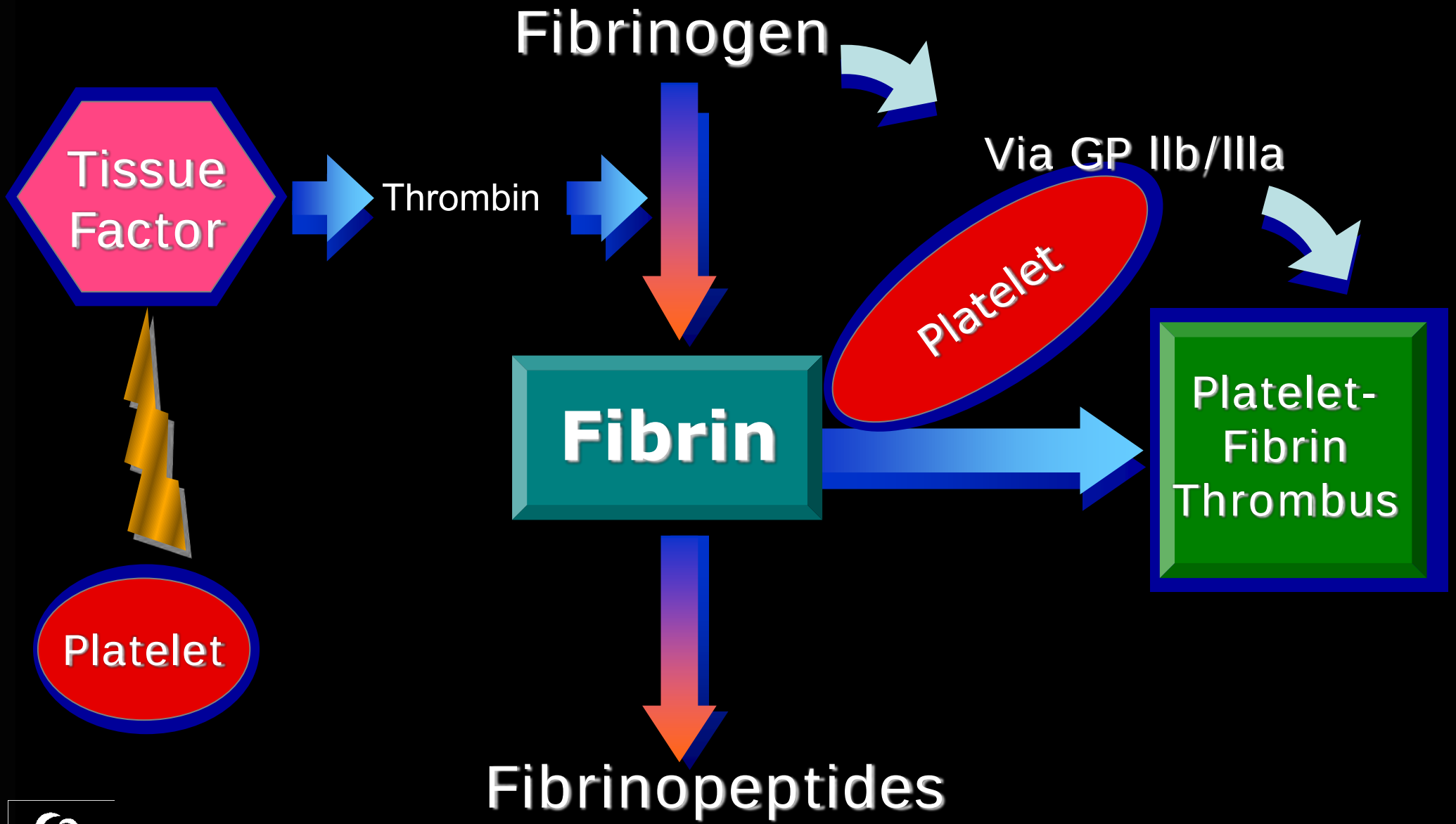
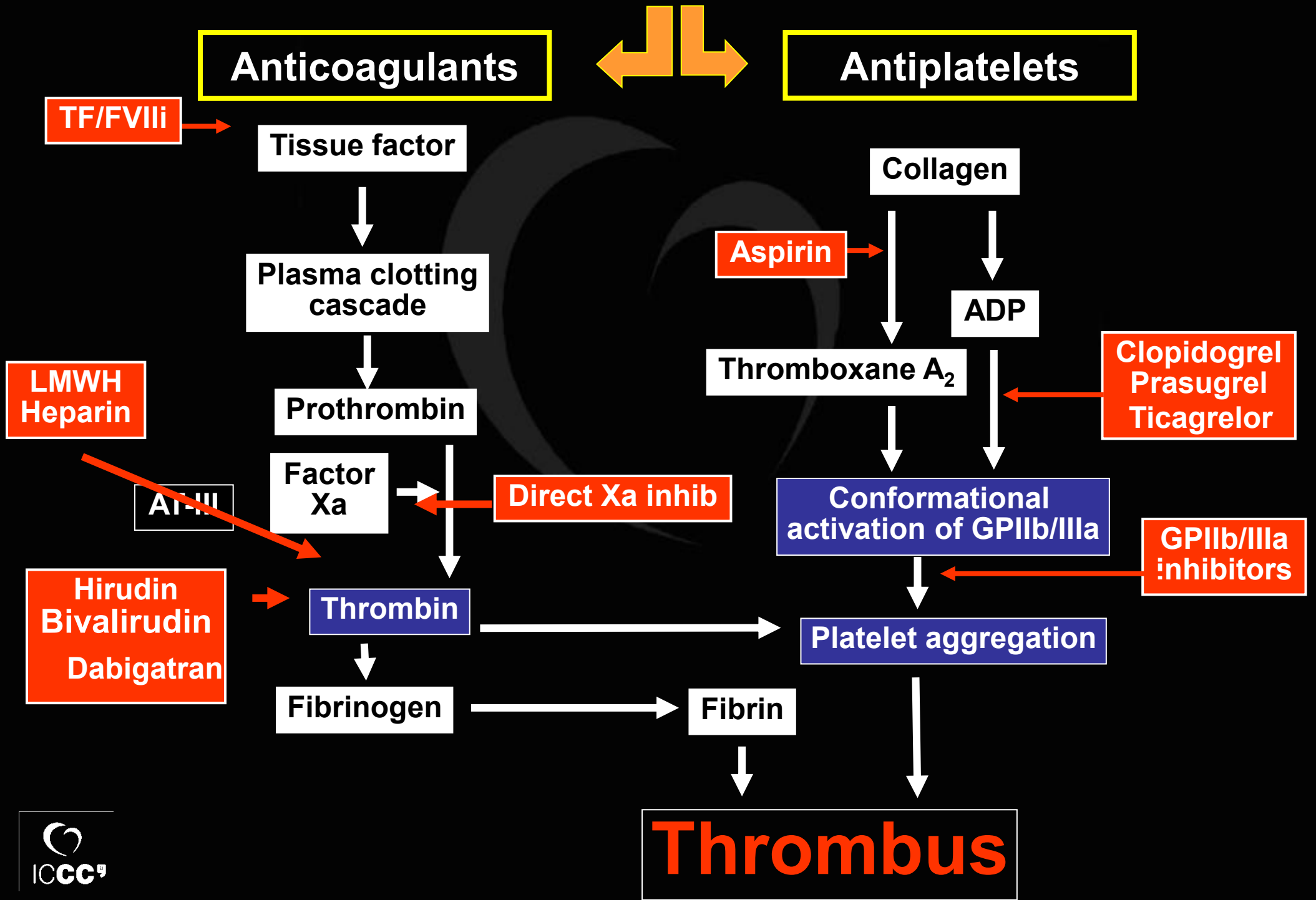
Platelets and coagulation interaction:
Tissue factor

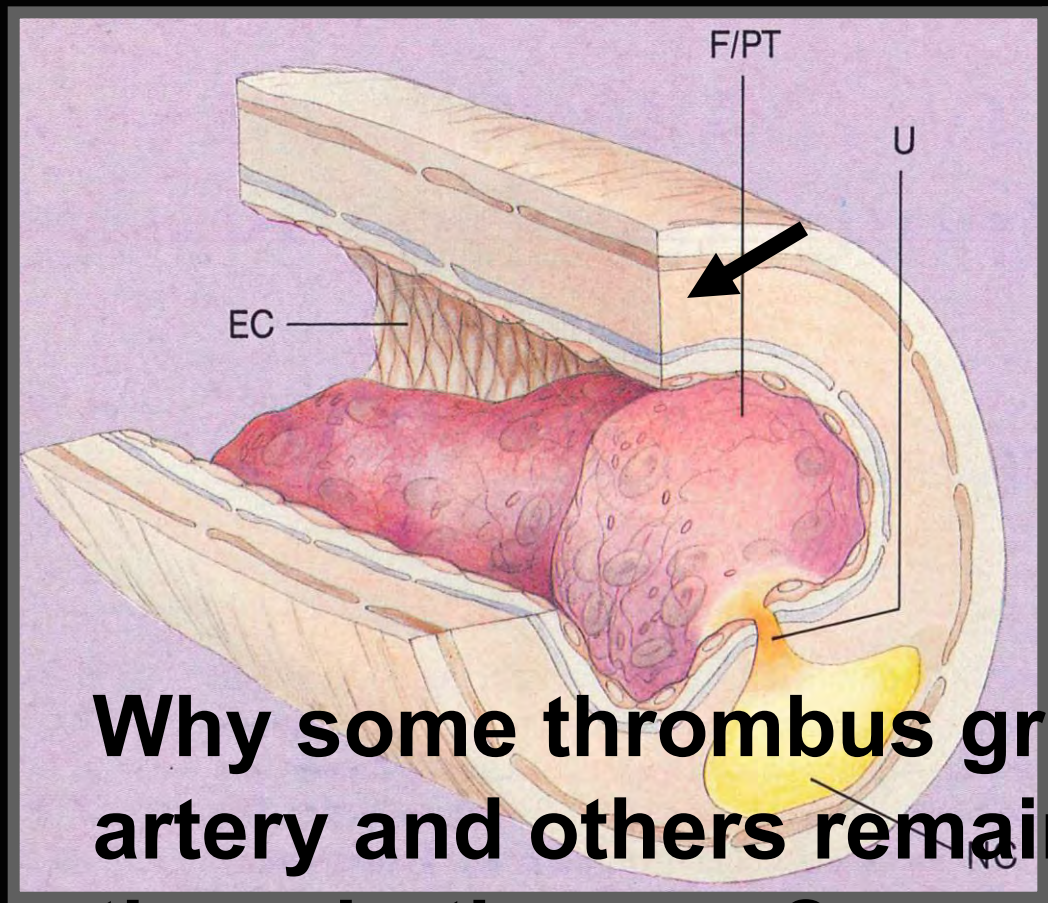


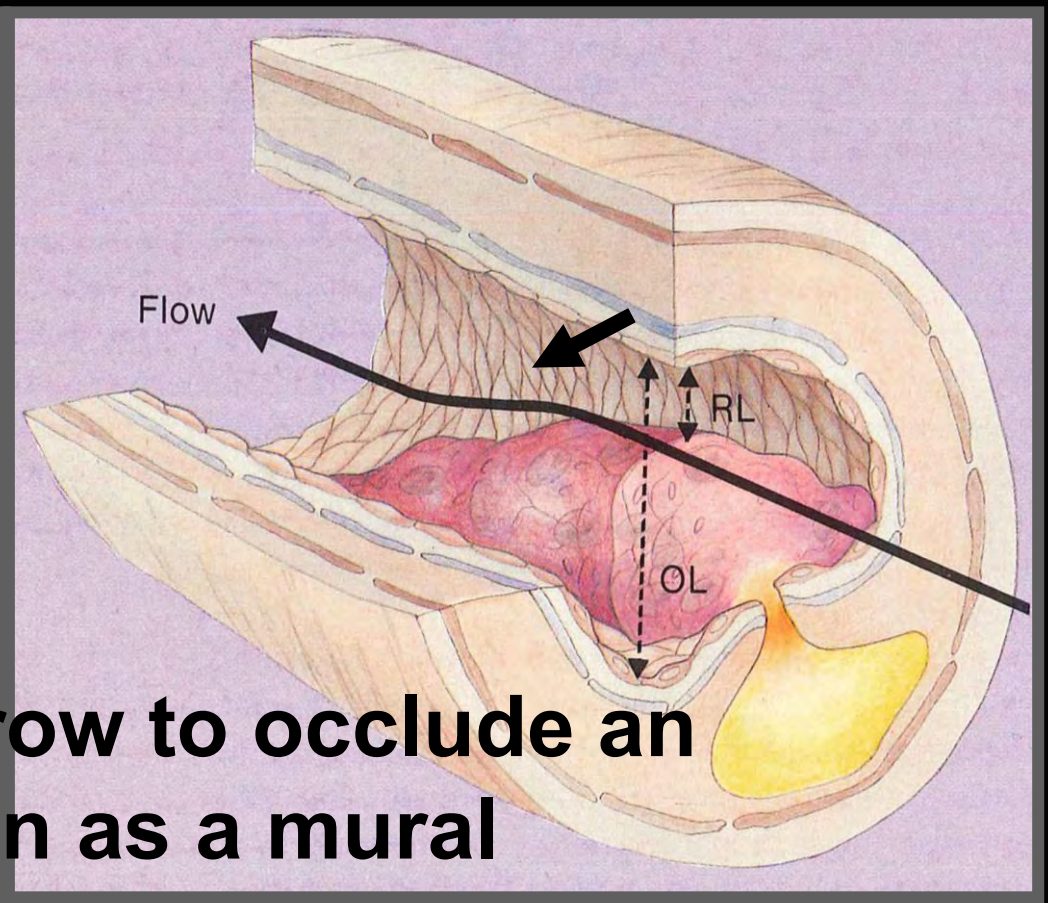
Platelets and coagulation interaction:
Tissue factor



Targets for ANTITHROMBOTICS





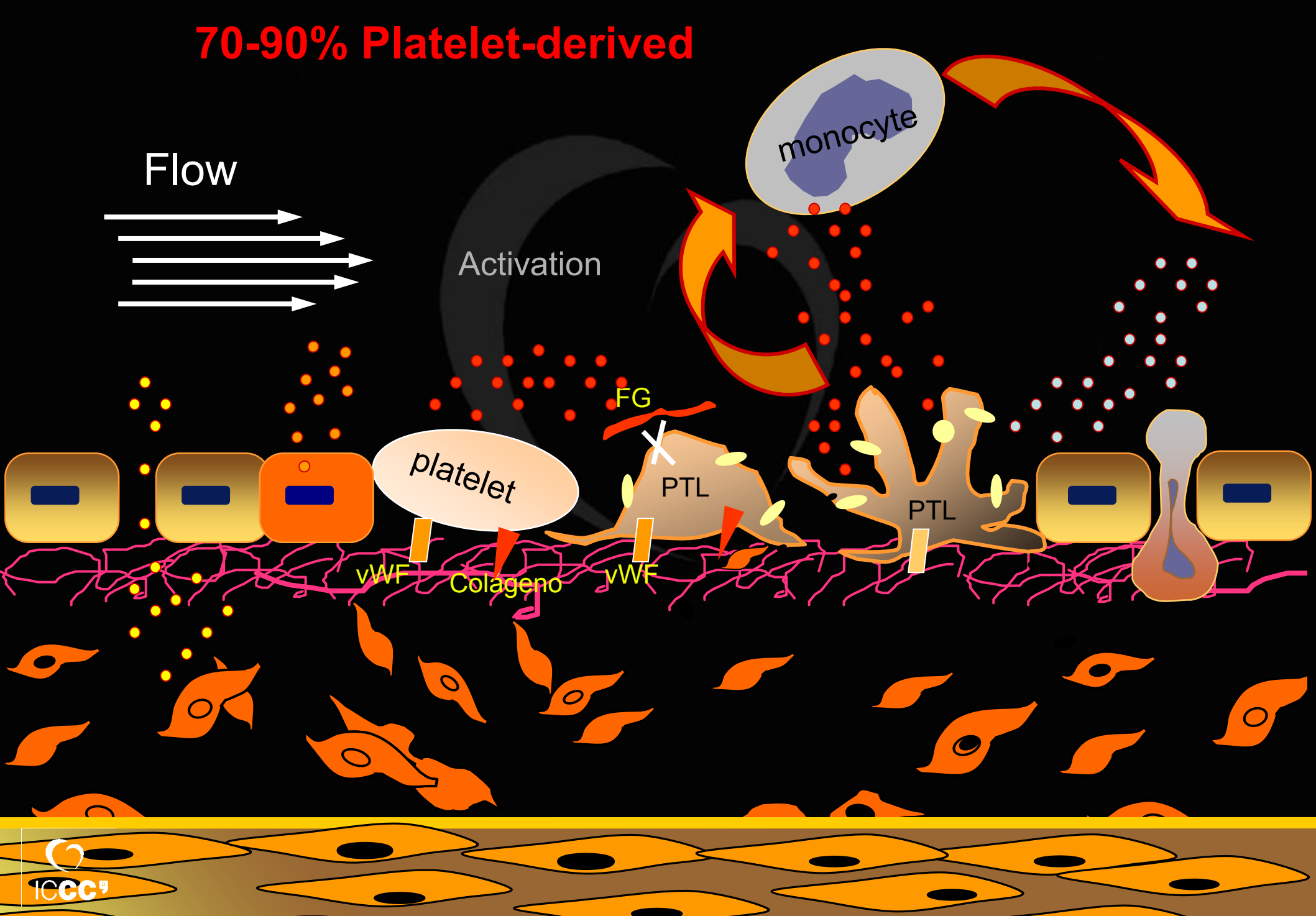


Why some thrombus grow to occlude an artery and others remain as a mural thrombotic mass?

Clinical implications are significantly different.

However, the mechanisms to explain both situations are not yet known!

Blood Microparticles



Proteomic Profile of the Platelet-MPs

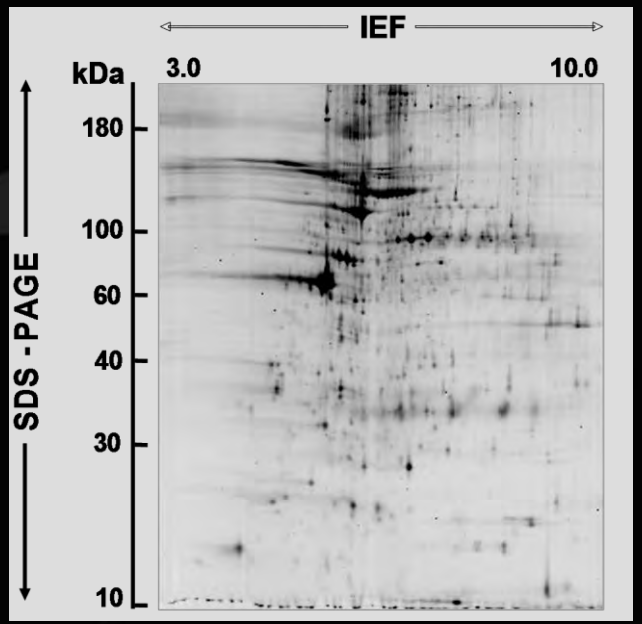
2D-electrophoresis & Maldi-ToF/ToF

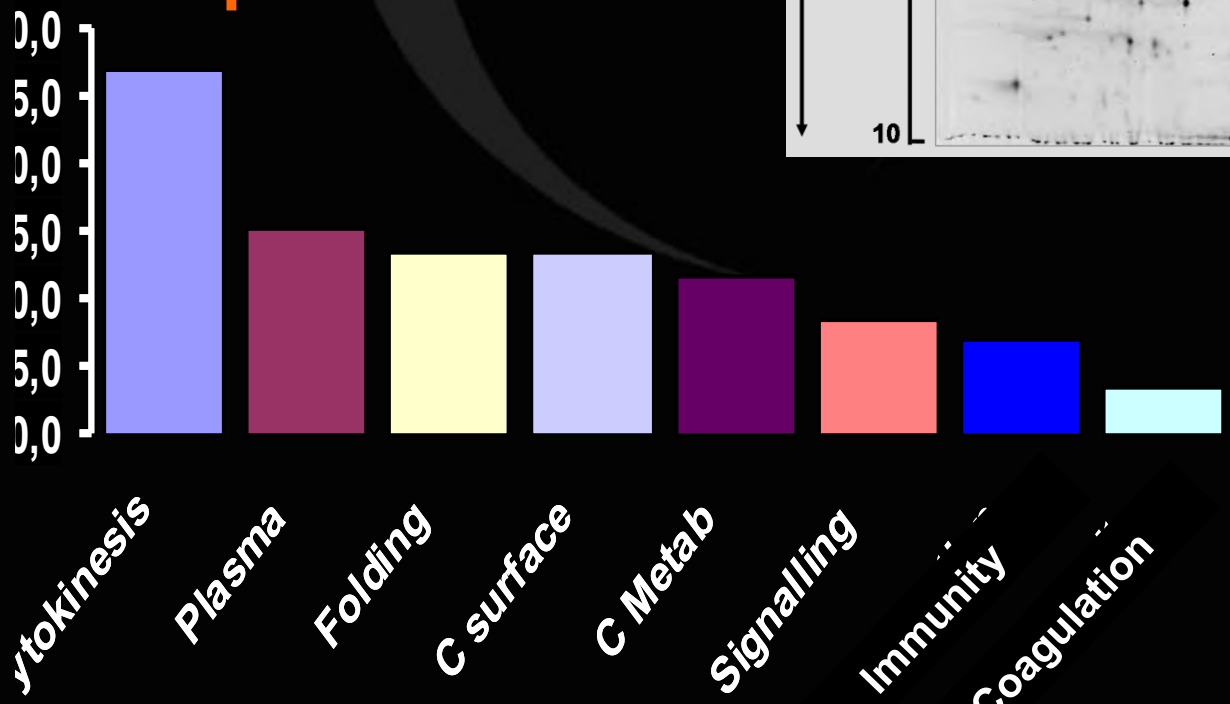
MPs- fraction (385 spots)

156 spots

MS

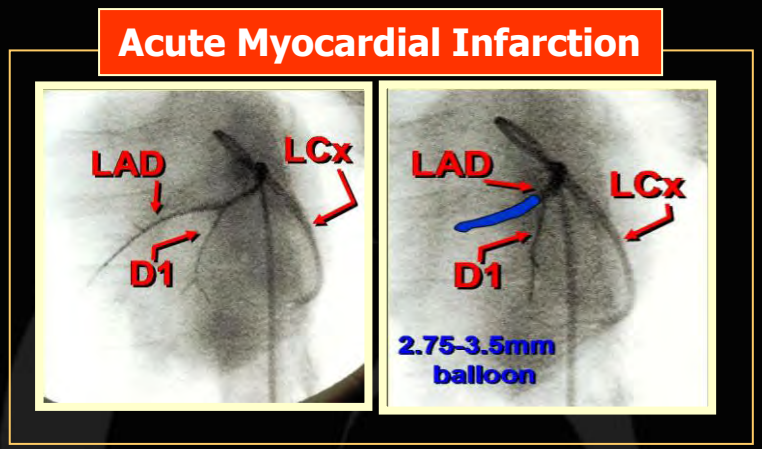
56 proteins

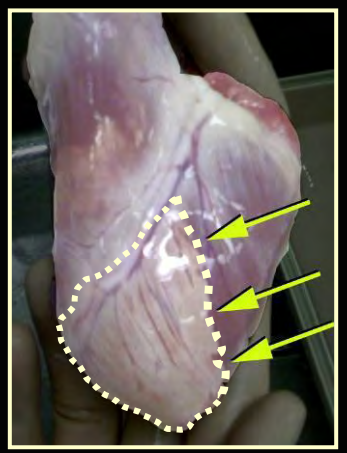




Padró T, Suades R, Segales N, Badimon L, *Cong Soc Esp Arterioscler, 2009*

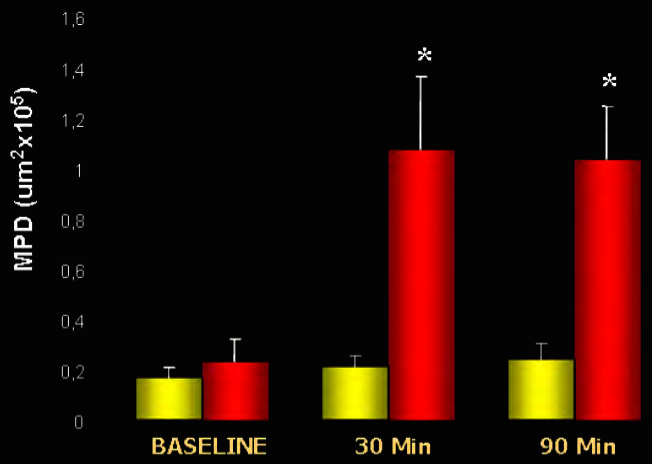
Potential effect of ischemia on thrombotic risk

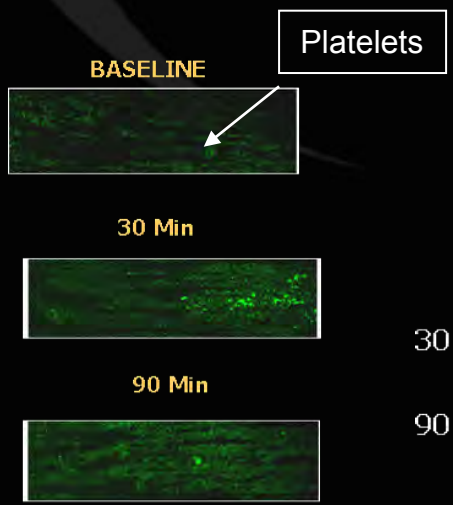




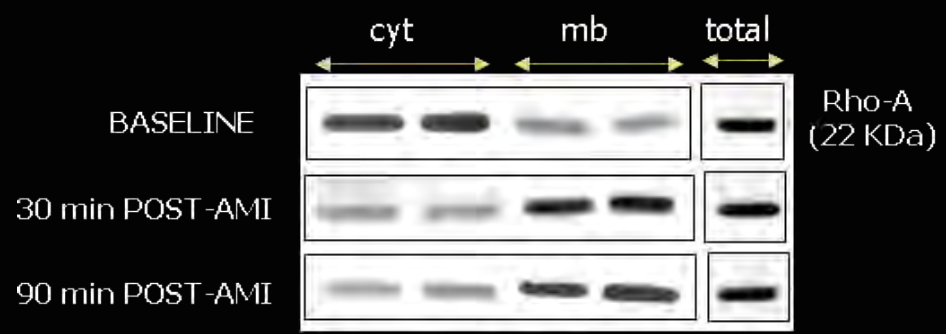
TIME OF CARDIAC ISCHEMIA

PLATELET DEPOSITION





Rho-A PLATELET EXPRESSION



SHAM-OPERATED ANIMALS ANIMALS SUBJECTED TO ISCHEMIA

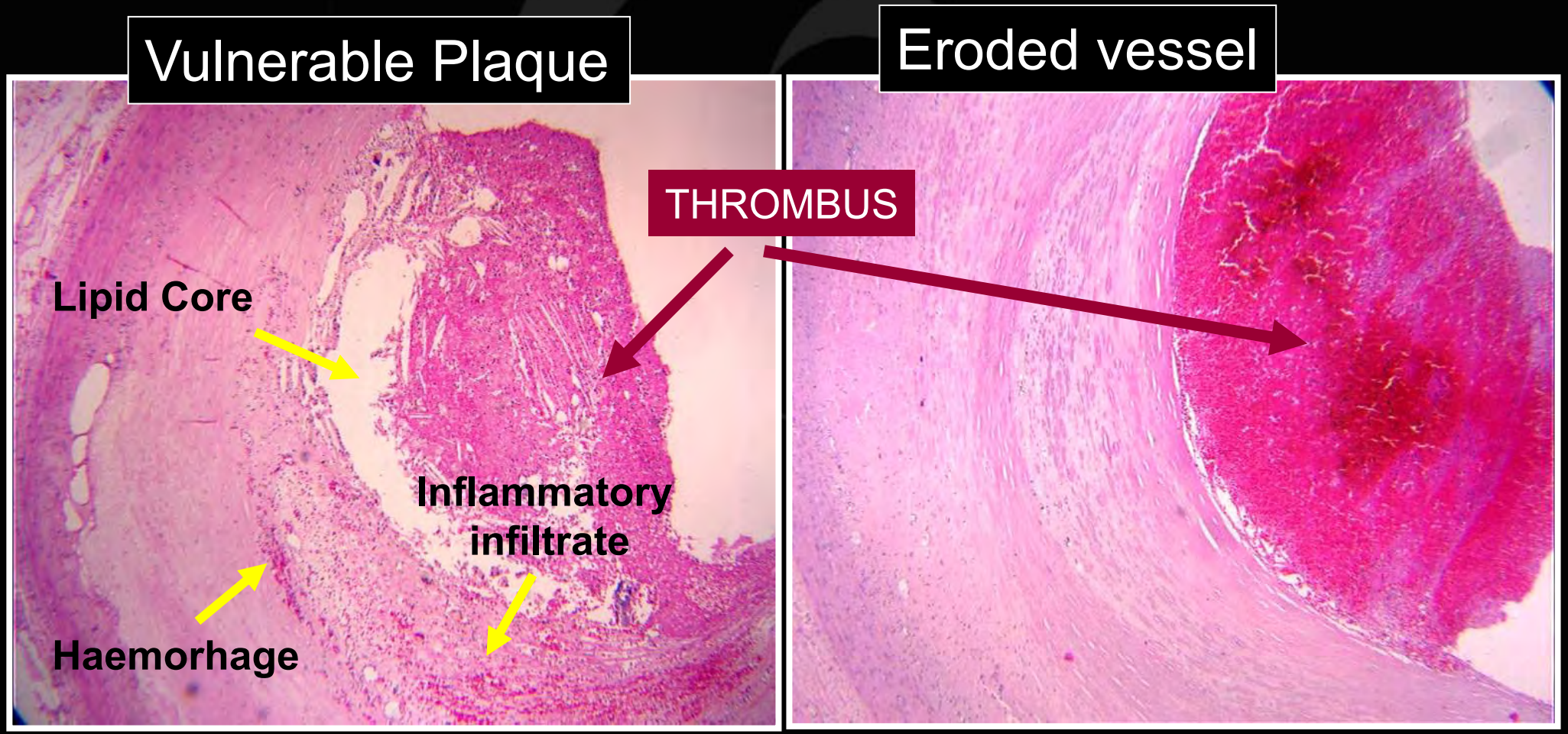
ICCC'9

Vilahur G, Hernández-Vera R, Badimon L, et al, *J Thromb Haemost 2009*

Atherothrombosis = Disrupted Vulnerable Plaque ?

EULALIA STUDY

(Cases of Sudden Death)



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