Platelet dysfunction in hypertension: implications for anti-platelet treatment

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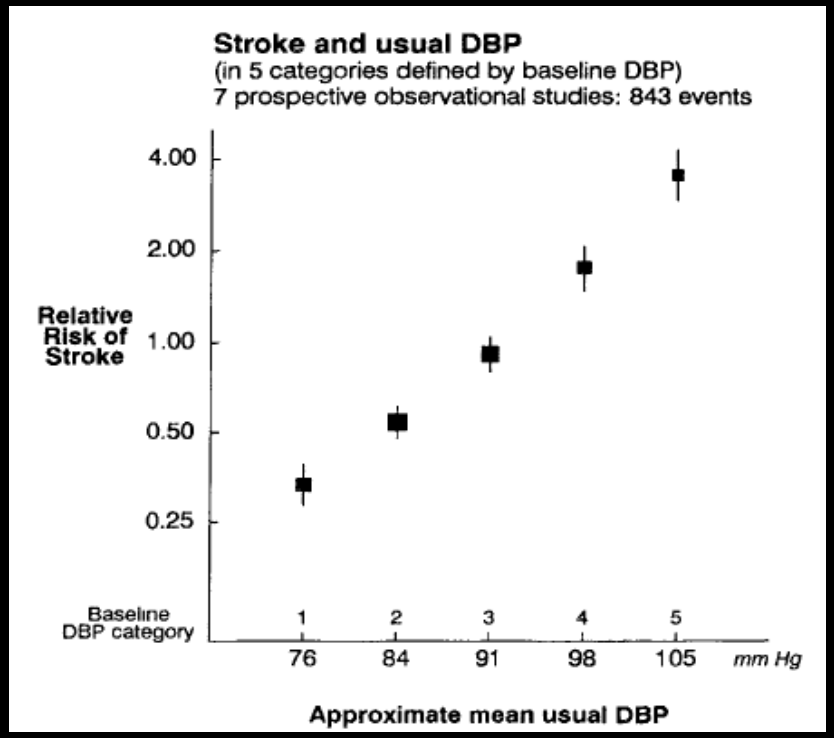
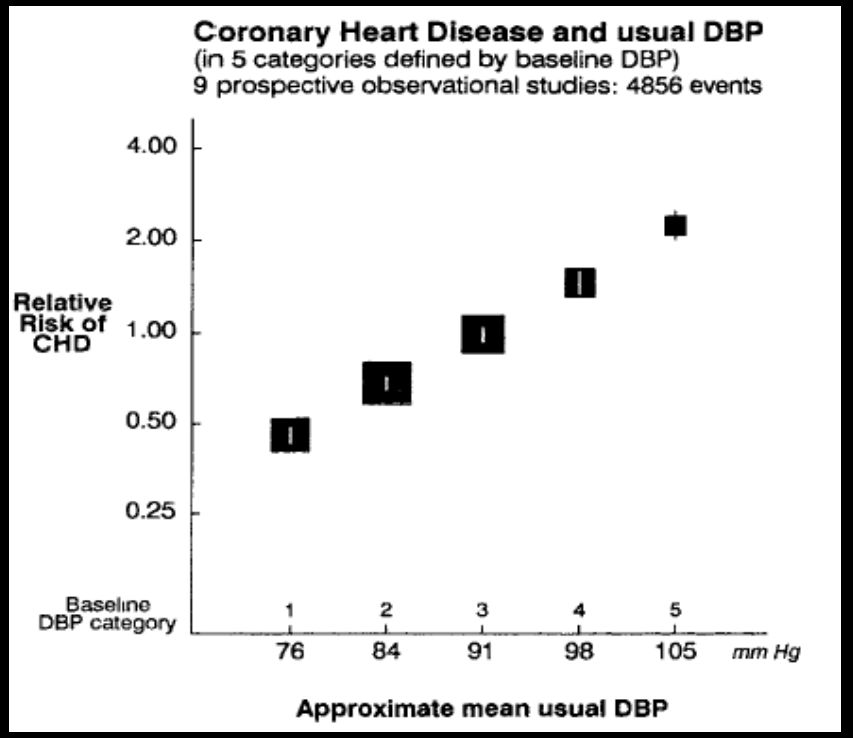
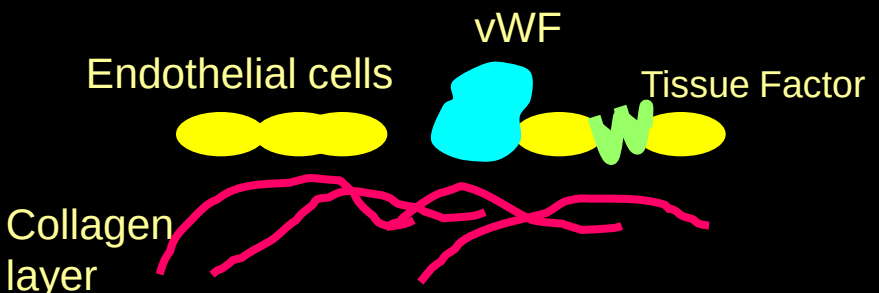
Thrombotic events: the link to blood pressure





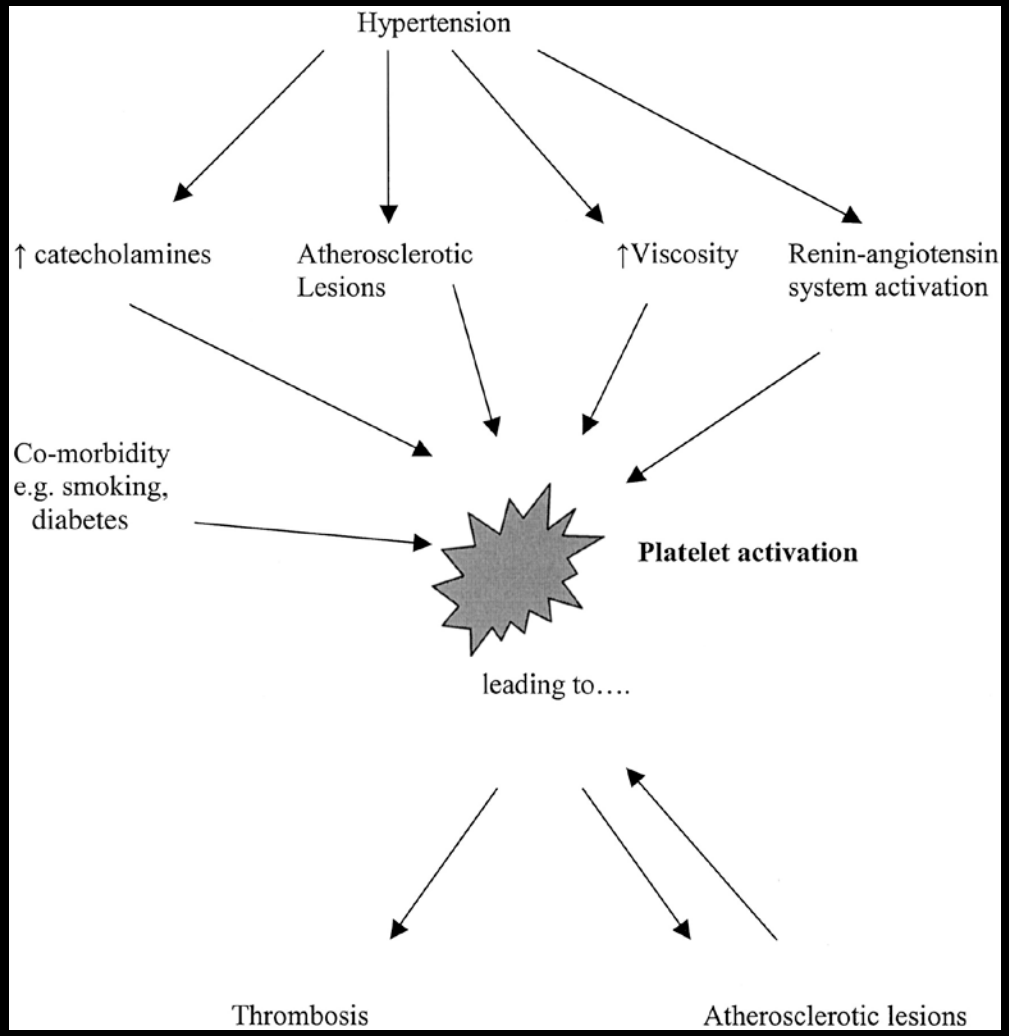
MacMahon *et al Lancet* 1990;335:765-774

Essential hypertension and arterial thrombosis: putative pathogenetic mechanisms



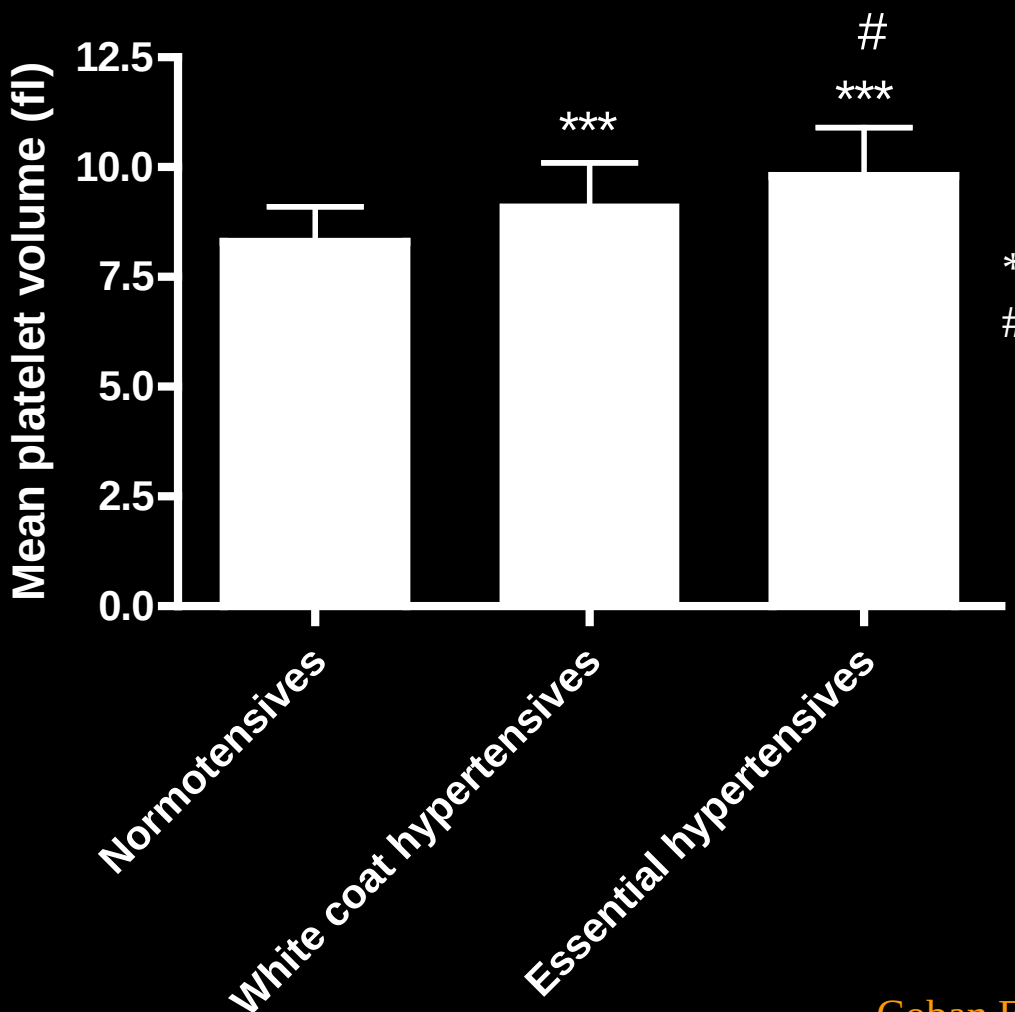
- Endothelial damage
  - ↓ Nitric oxide
- Disturbances in blood flow – secondary to:
  - Atherosclerosis
  - Elevated blood viscosity
- Platelet activation

Possible routes leading to and from platelet activation in hypertension



Blann AD *et al. Hypertension* 2003;42:1-7

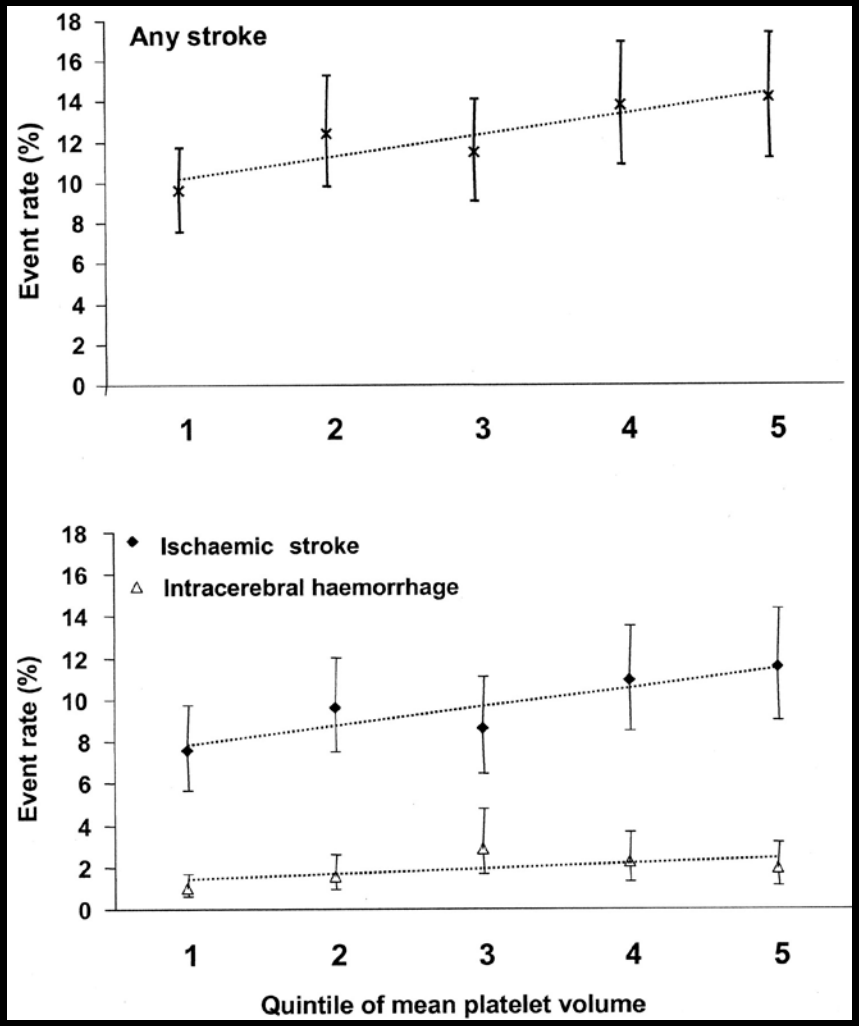
Platelets are enlarged in hypertension



*** P < 0.001 vs normotensives
# P < 0.05 vs w-c hypertensives

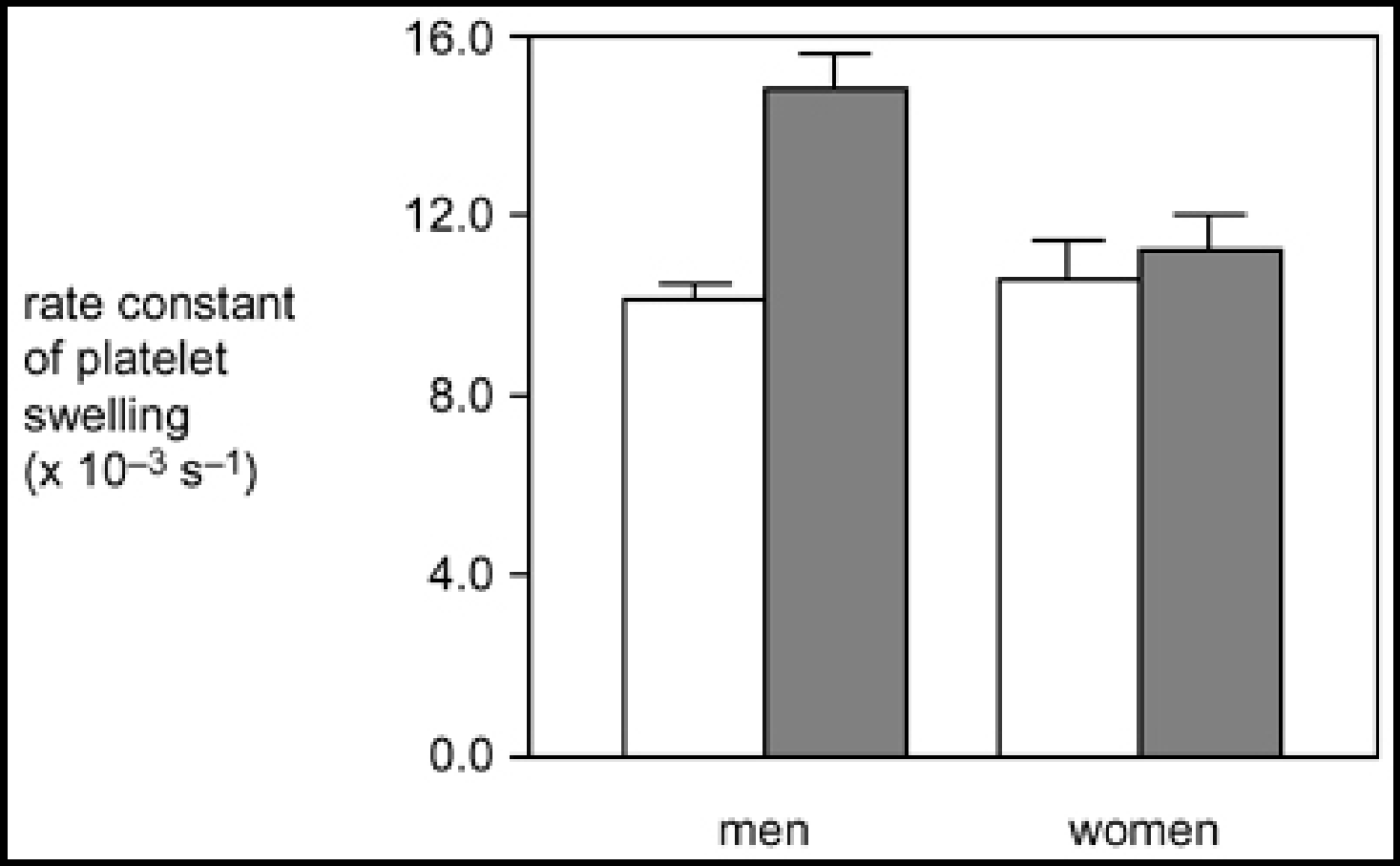
Coban E. *et al. Platelets* 2005;16:435-8

MPV is correlated with incidence of CVA



Bath P *et al. Stroke* 2004;35:622-6

Platelets from hypertensive men swell faster, through Na^+/H^+ exchange



Stratton PD & Ritter JM. *J Hypertens* 1997;15:1403-6

Markers of activation are increased in platelets from hypertensives

Table 1. Baseline Characteristics of Patients and Controls

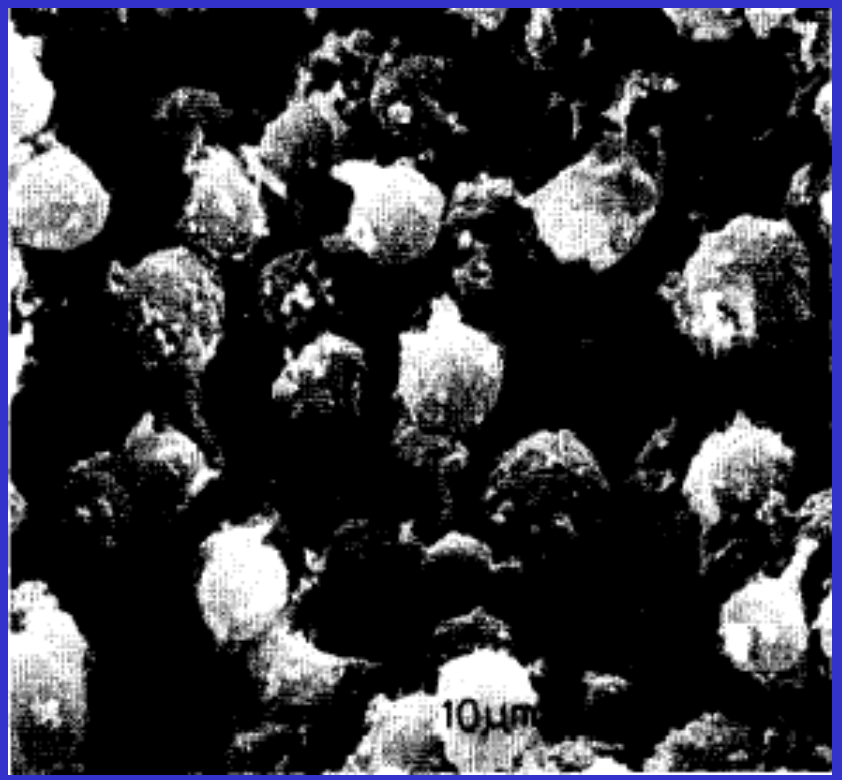
	Healthy Controls (n = 59)	All Hypertensives (n = 199)	p Value*
Age (yrs)	68 ± 11	68 ± 10	0.7
Males	41 (69%)	151 (76%)	0.3
SBP (mm Hg)	130 ± 10	147 ± 22	0.001
DBP (mm Hg)	80.5 ± 9	80.1 ± 11	0.9
Smokers (% admitting)	0	12.5	<0.001
Platelet indexes			
Platelet count × 10^6/ml	227 ± 49	239 ± 65	0.2
MPV (fl)	6.36 ± 0.9	7.8 ± 3.03	0.005
MPM (pg)	1.76 ± 0.17	1.9 ± 0.21	<0.001
MPG (g/dl)	29 ± 2.3	27.1 ± 2.5	<0.001
Beta-TG (IU/ml)	100 (57–282)	307 (210–354)	<0.001
Soluble P-selectin (ng/ml)	77 (38–110)	150 (106.8–213)	<0.001
Platelet P-selectin ($\times 10^{-6}$ ng/cell)	78.3 (47–120)	99.6 (73.2–138.5)	0.004

*Obtained from the unpaired *t* test (for age, blood pressure, platelet count, MPV, MPM, MPG), the Mann-Whitney *U* test (beta-TG, soluble and platelet P-selectin), or the chi-square test (males, smokers). Data are presented as the mean value ± SD or median value (interquartile range).

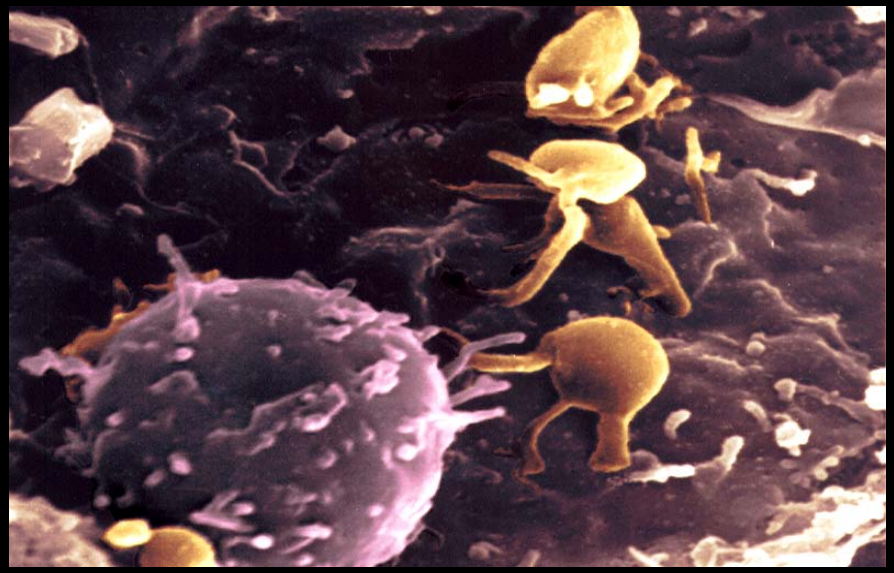
Beta-TG = beta-thromboglobulin; DBP = diastolic blood pressure; MPG = mean platelet granularity; MPM = mean platelet mass; MPV = mean platelet volume; SBP = systolic blood pressure.

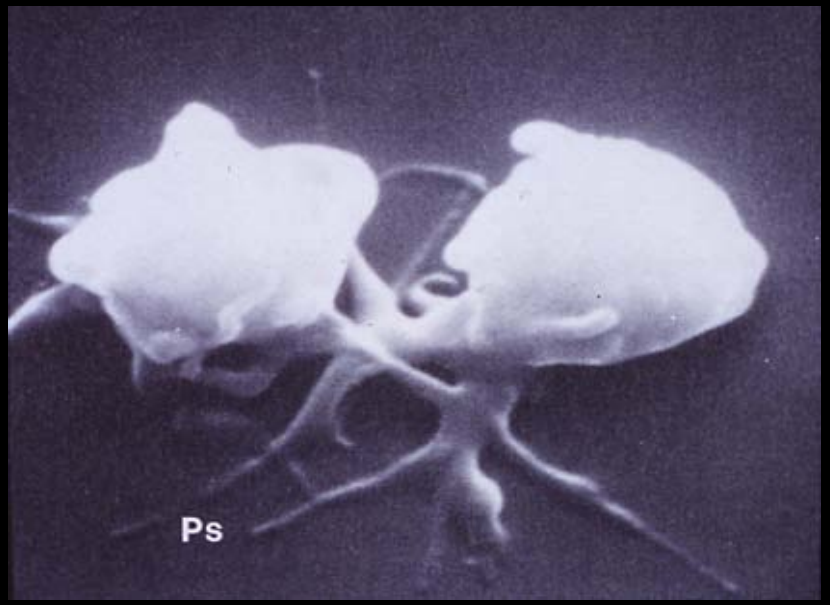
Nadar SK *et al. J Am Coll Cardiol* 2004;44:415-22

Platelets from hypertensive patients show structural changes: pseudopod formation



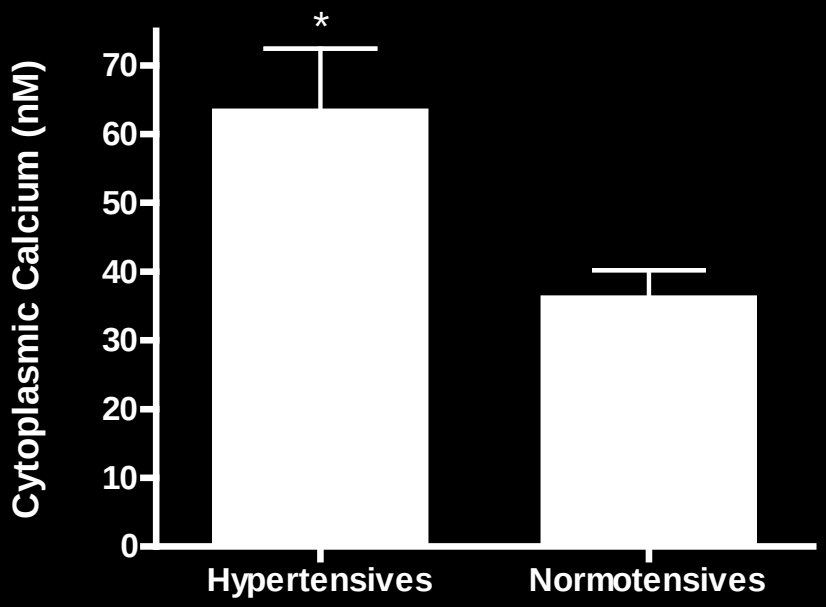
Nityanand S *et al. Thromb Res* 1993;72:447-54



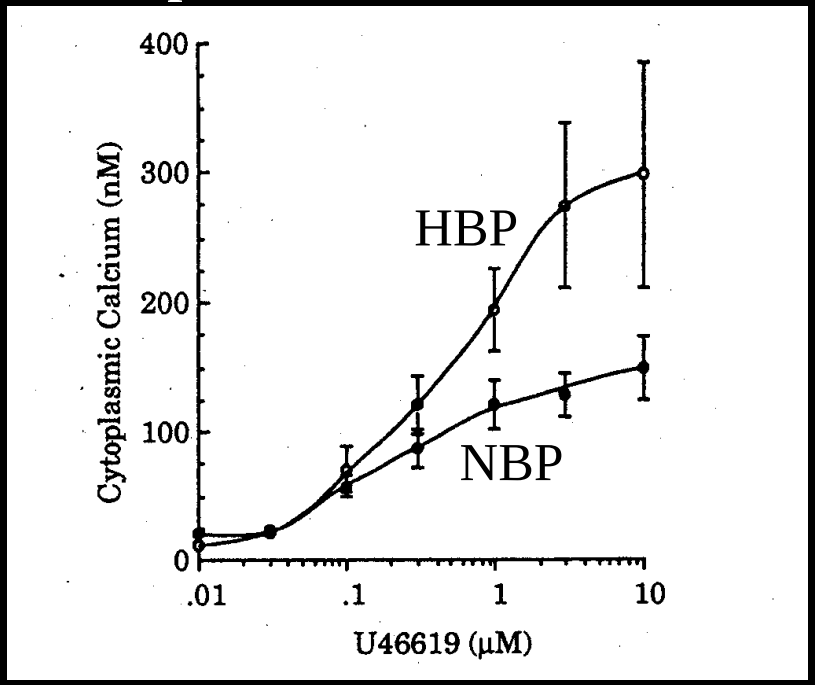

Ps

Cytosolic Ca^{2+} in platelets from hypertensive patients

Basal



Response to U46619



Woods JD *et al. J Hypertens* 1993;11:1369-73

Platelet abnormalities in hypertension

Morphological

Shape changes

↑Platelet volume & mass

Biochemical/functional

↑ Cytosolic calcium

↑ Release of β-TG

↑ P-selectin (soluble and on platelets)

↑ Aggregability to agonists

Biological effects of NO

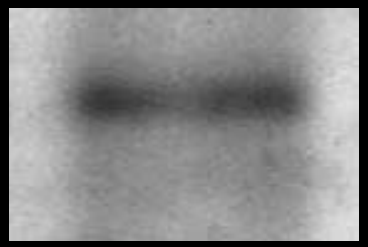
Mostly mediated through cGMP

- Relaxes vascular (and other) smooth muscle
- **Inhibits platelet activation, adhesion and aggregation**
- Neurotransmitter
- Tumoricidal and bactericidal
- Anti-atherogenic

Platelets express NOS-3 >> NOS-2

NOS-2

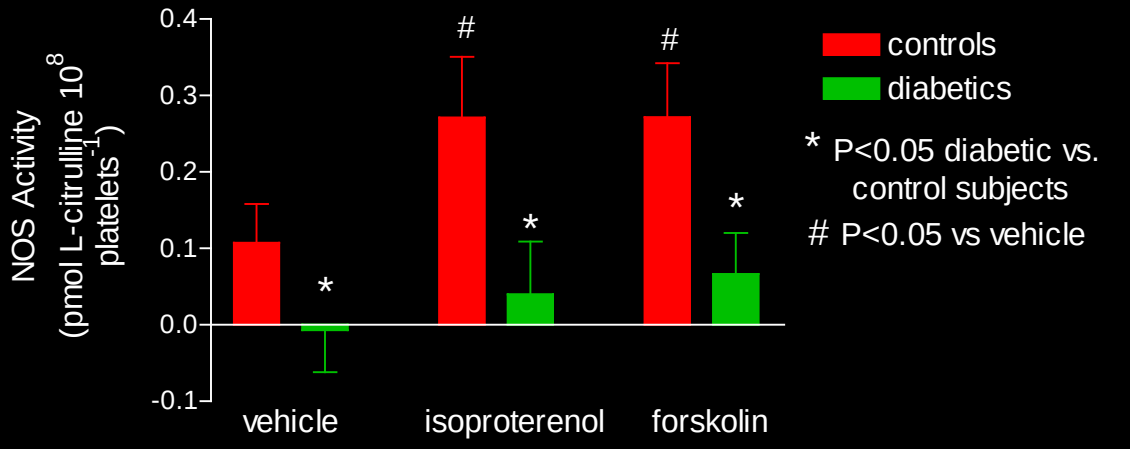
α-Tubulin

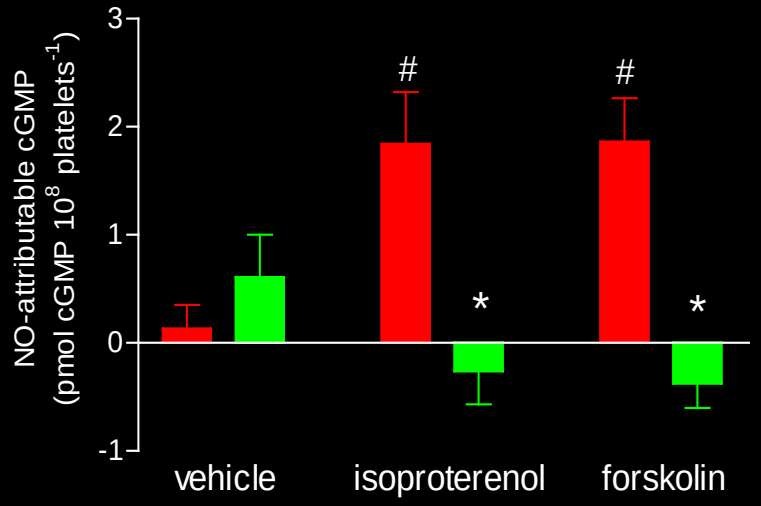


NOS-3

α-Tubulin

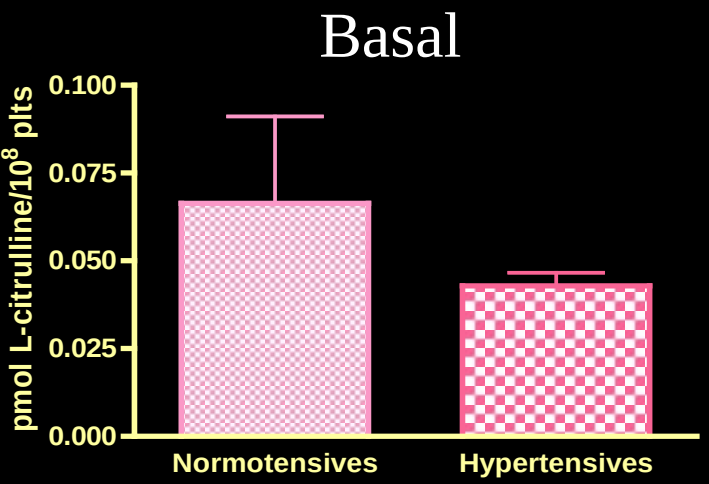
Platelets from type 2 diabetic patients exhibit decreased basal and stimulated NOS activity

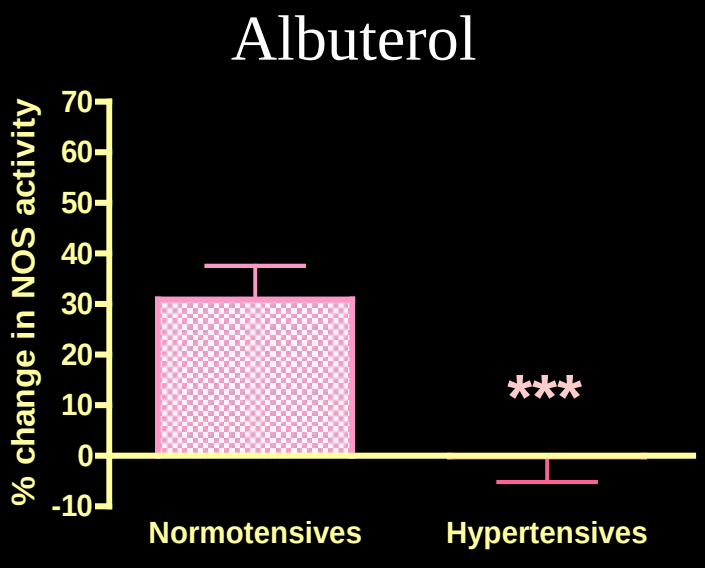


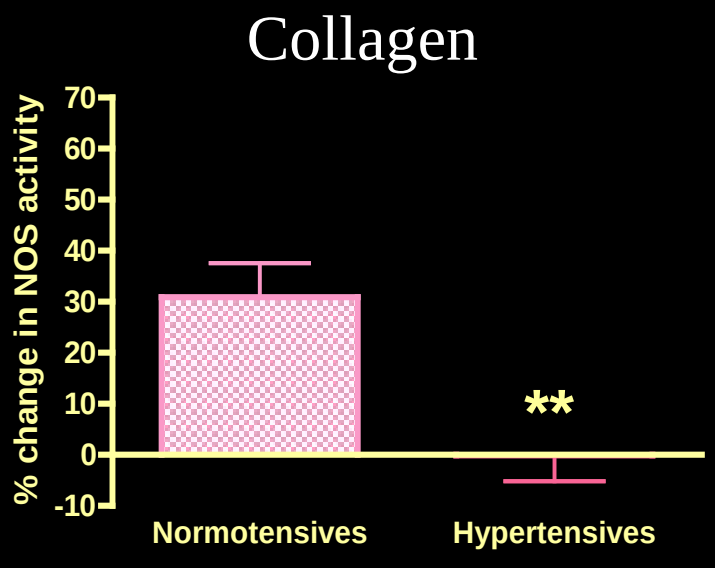


Queen LR *et al. Diabetologia* 2003;46:1474-82

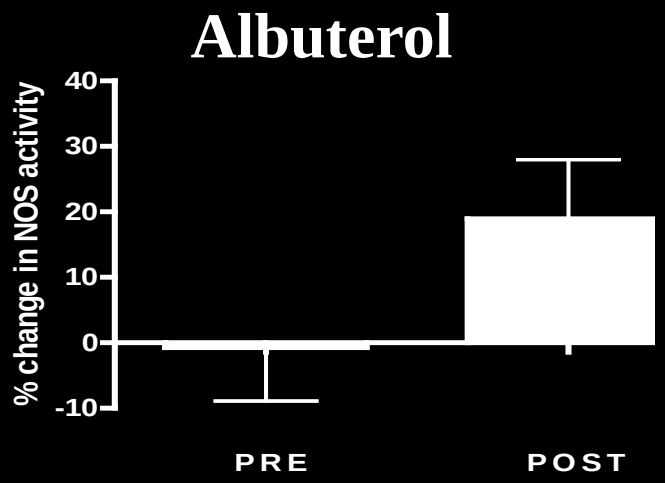
Stimulated, but not basal, NOS activity, is impaired in hypertensive subjects

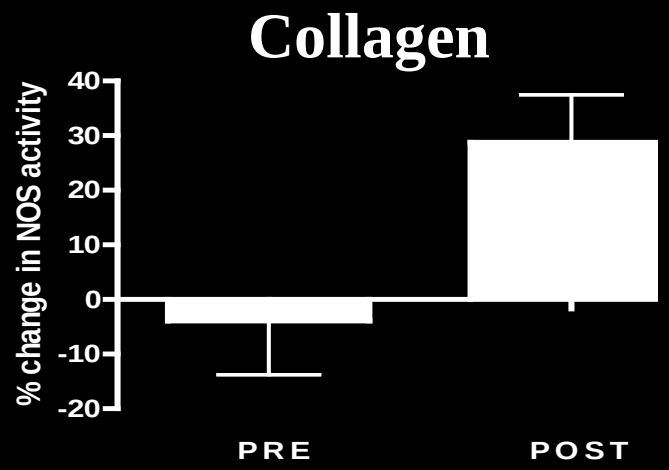


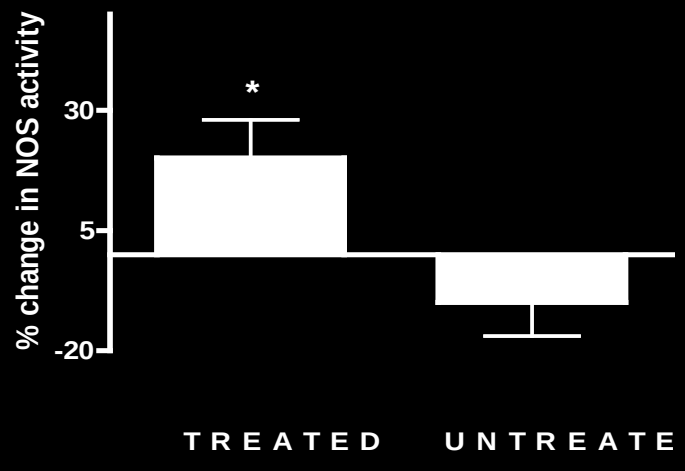


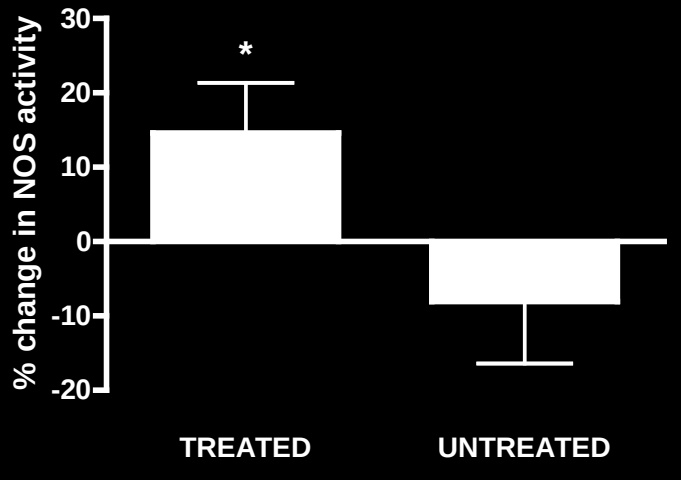


Treatment of hypertension may increase stimulated platelet NOS activity

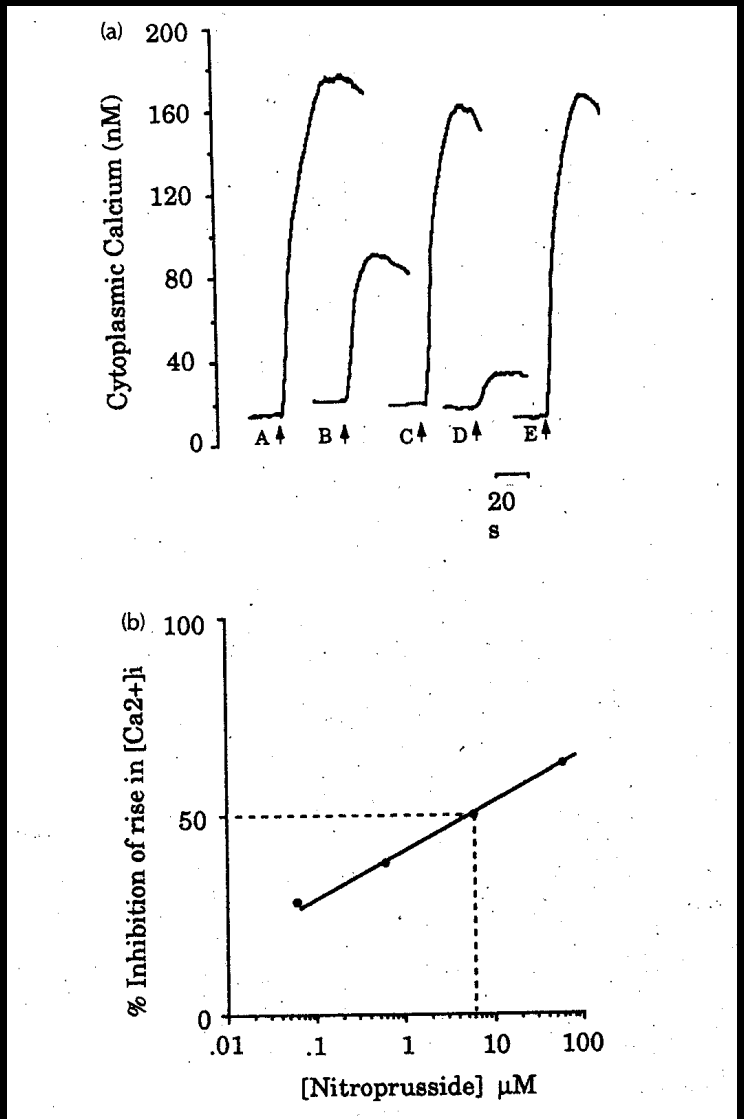


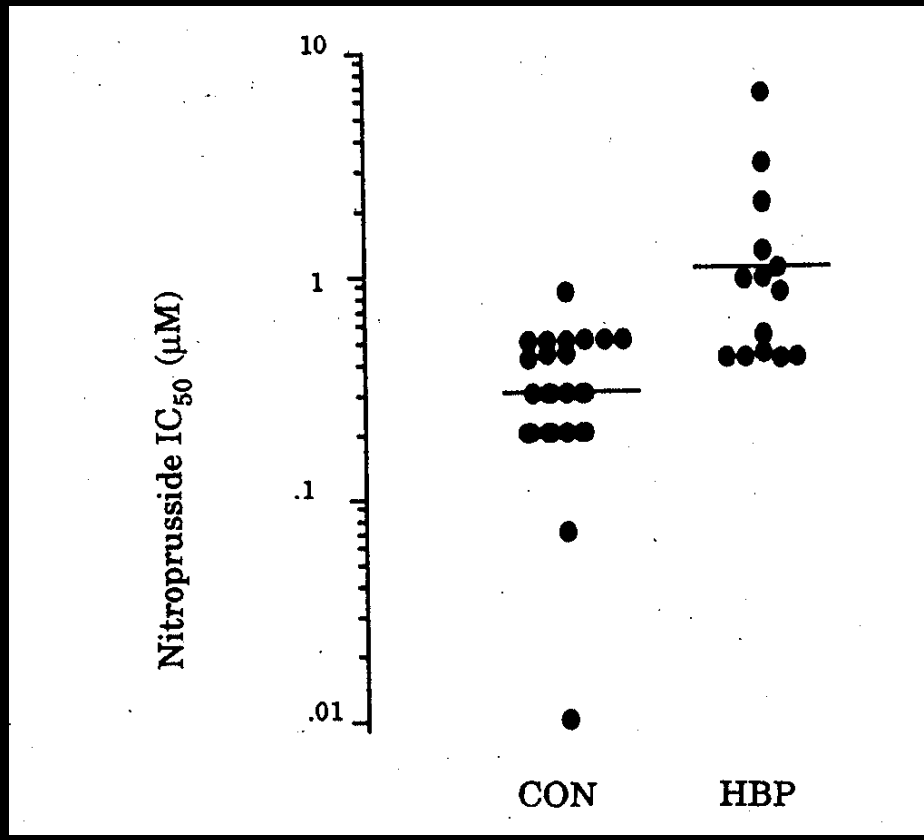






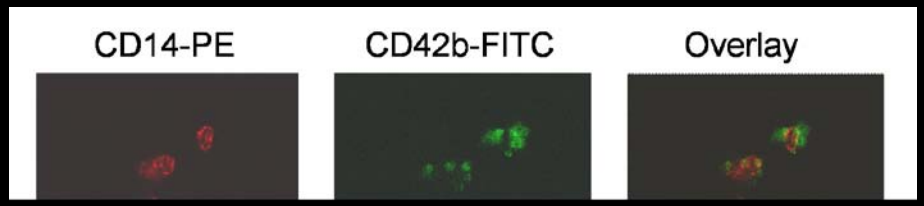
Reduced sensitivity of platelets from hypertensive patients to exogenous NO

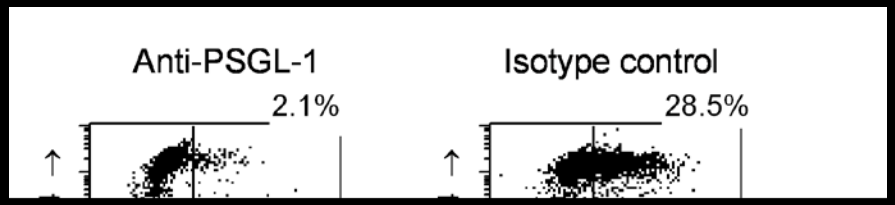




Woods JD *et al. J Hypertens* 1993;11:1369-73

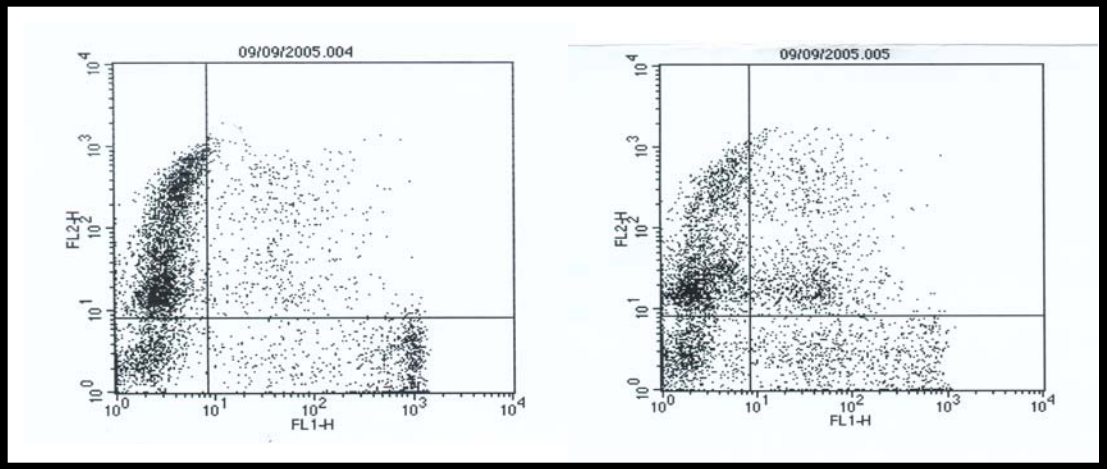
Monocyte-platelet aggregates: a sensitive marker of platelet activation





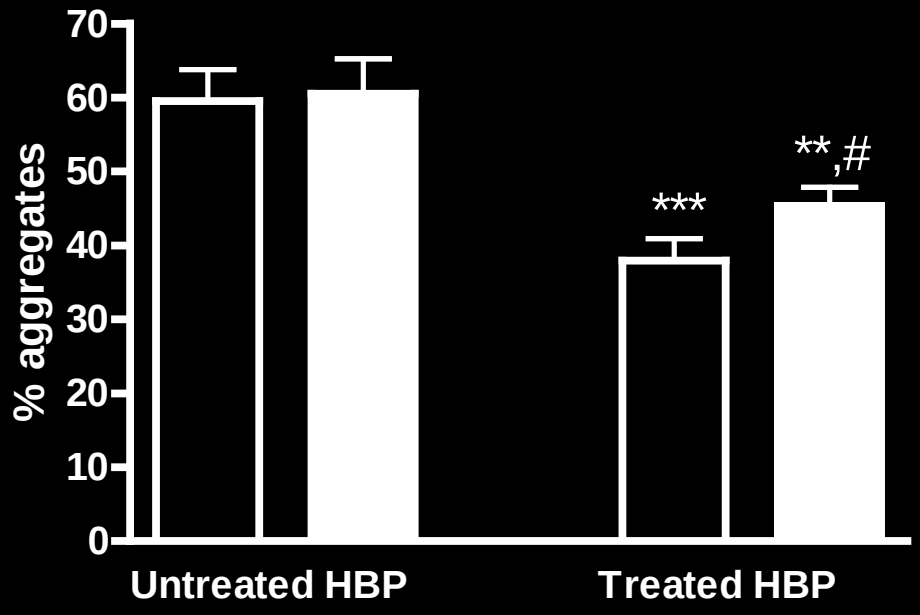
-L-NMMA

+ L-NMMA



Circulating MPA increase in relation to blood pressure

Basal NO production inhibits MPA formation in treated but not untreated hypertensives

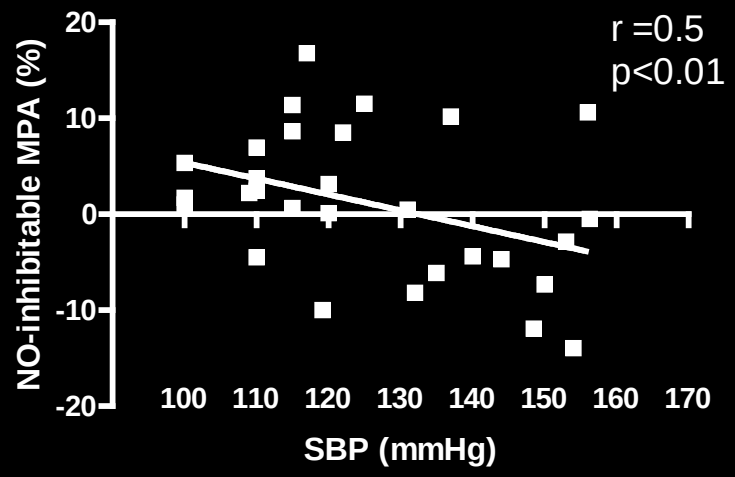


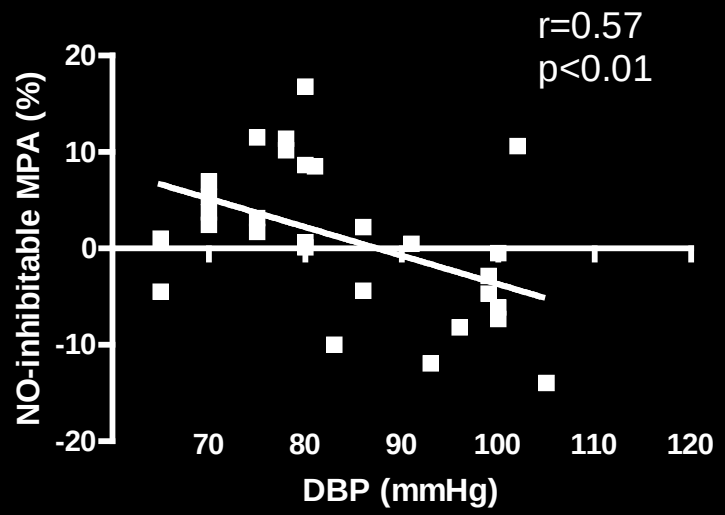
**, *** P < 0.01 and < 0.001 vs untreated

# P< 0.05 vs absence of L-NMMA

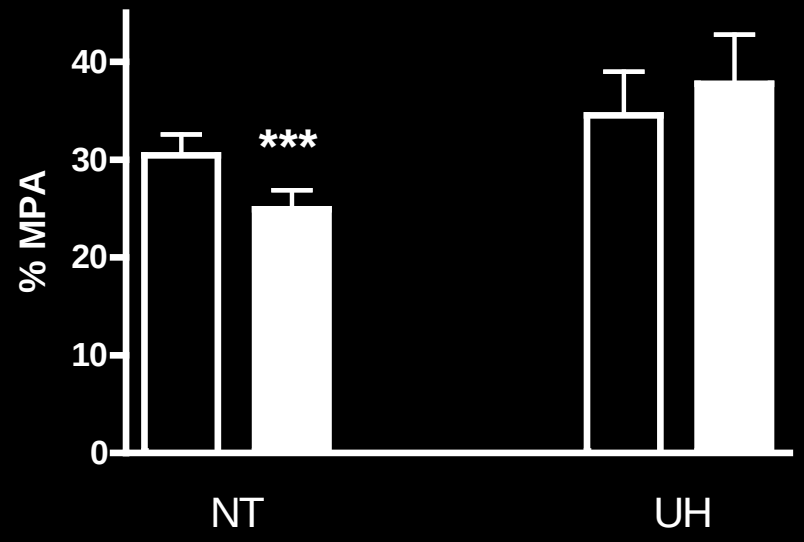
Clear bars: basal, filled bars: following L-NMMA treatment

Correlation between “NO-inhibitable” MPA and BP





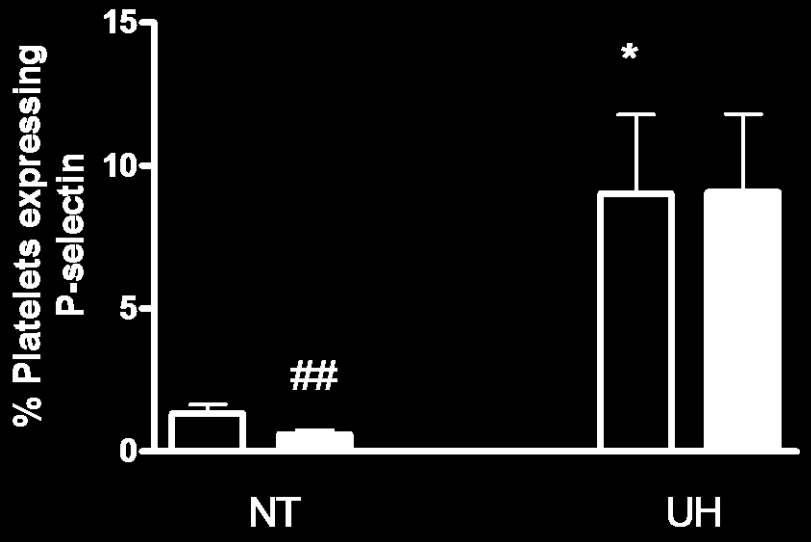
Effect of an NO donor (spermine NONOate) on MPA in normotensives and untreated hypertensives



*** P < 0.001 vs basal

Clear bars: basal, filled bars: following SNO treatment

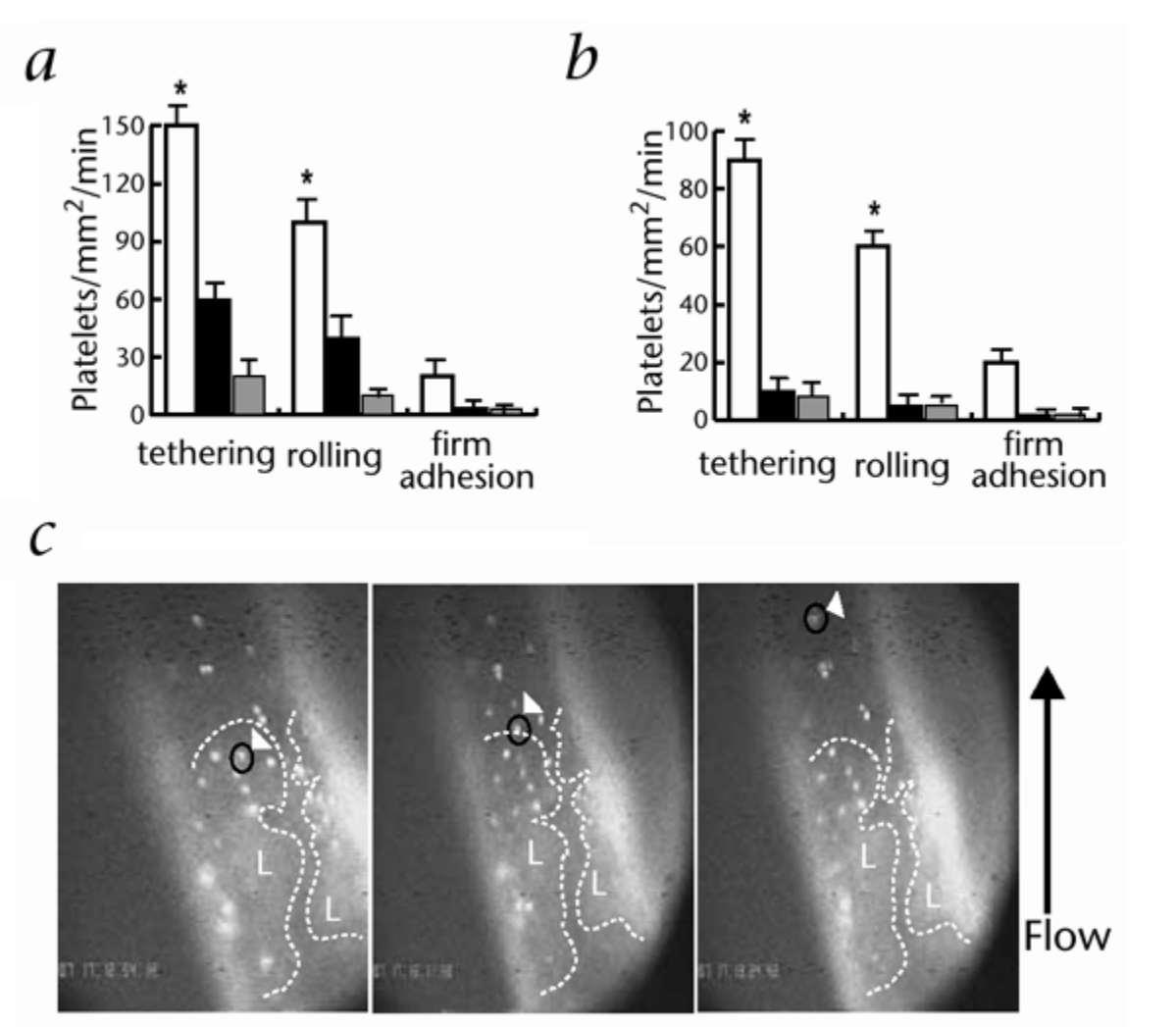
% of platelets expressing P-selectin, and the effect of an NO donor (spermine NONOate) on this



* P < 0.05 vs normotensives
P< 0.01 vs basal

Clear bars: basal, filled bars: following SNO treatment

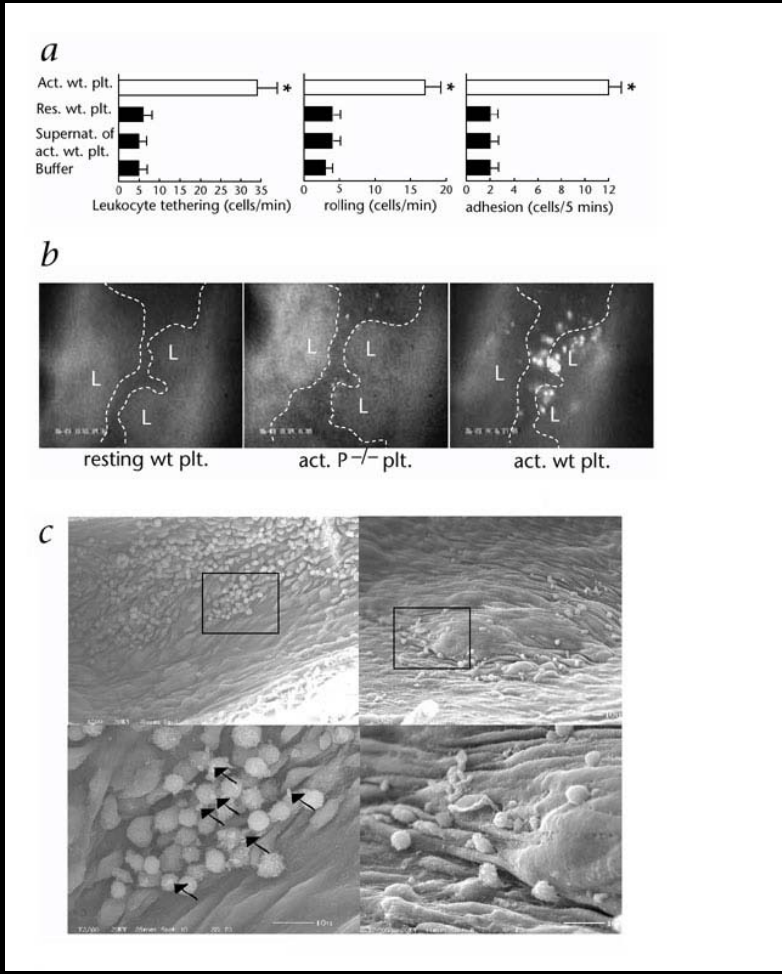
Interactions of activated platelets with atherosclerotic arteries

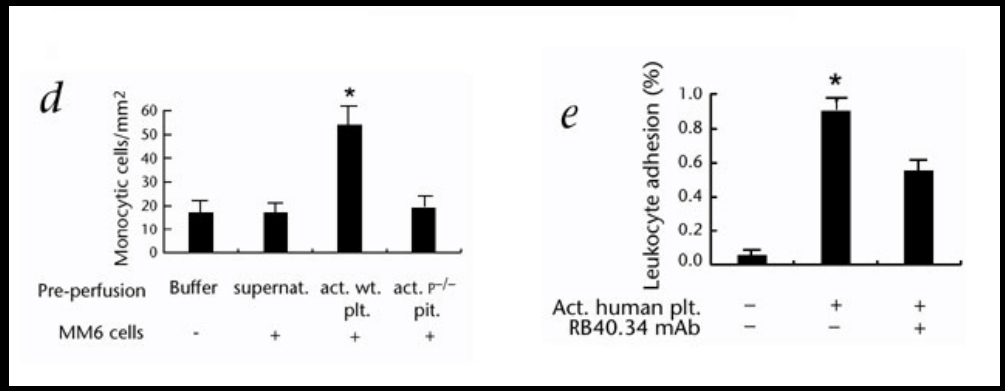


Interaction of activated wild-type (white bars), *Selp* -/- (P-/-) platelets (black bars) or resting platelets (grey bars) with aortic endothelial cells (A) or atherosclerotic carotid arteries (B)

Huo Y. *et al. Nature Med* 2003;9:61-7

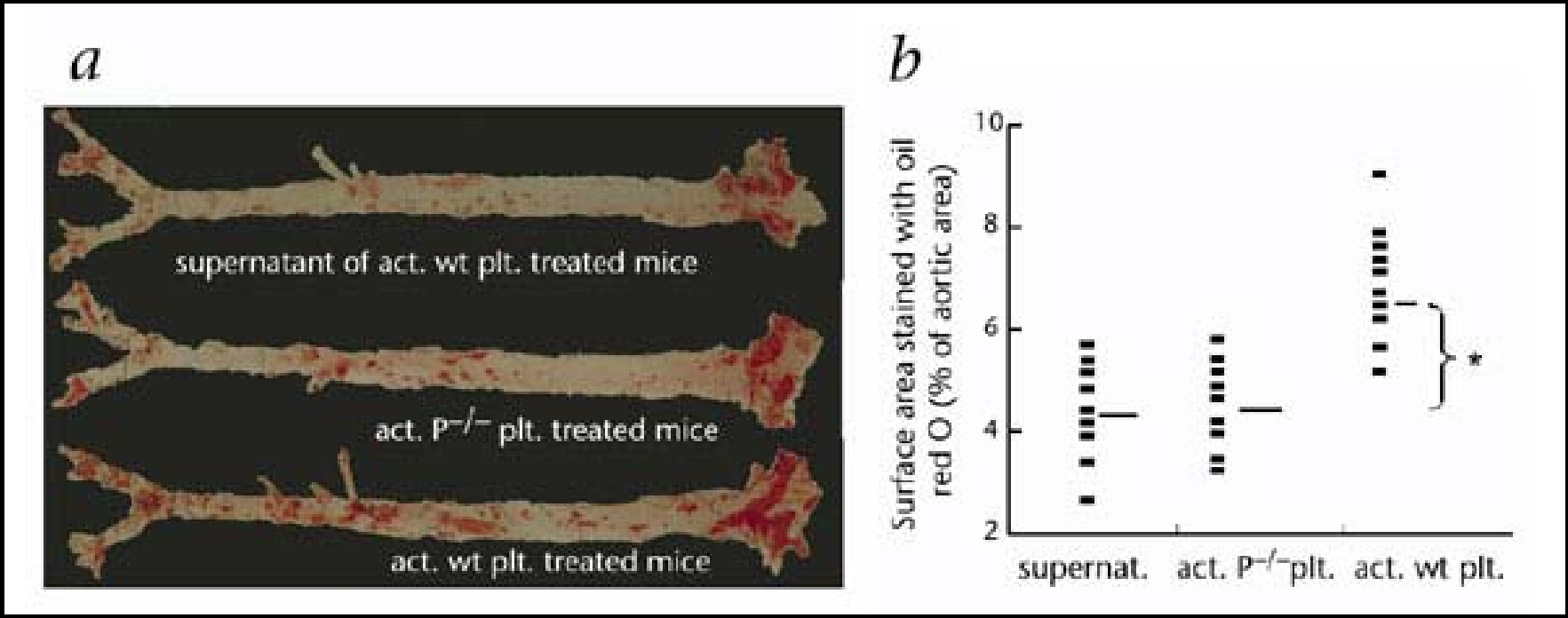
Monocyte–endothelial interactions: effect of activated platelets





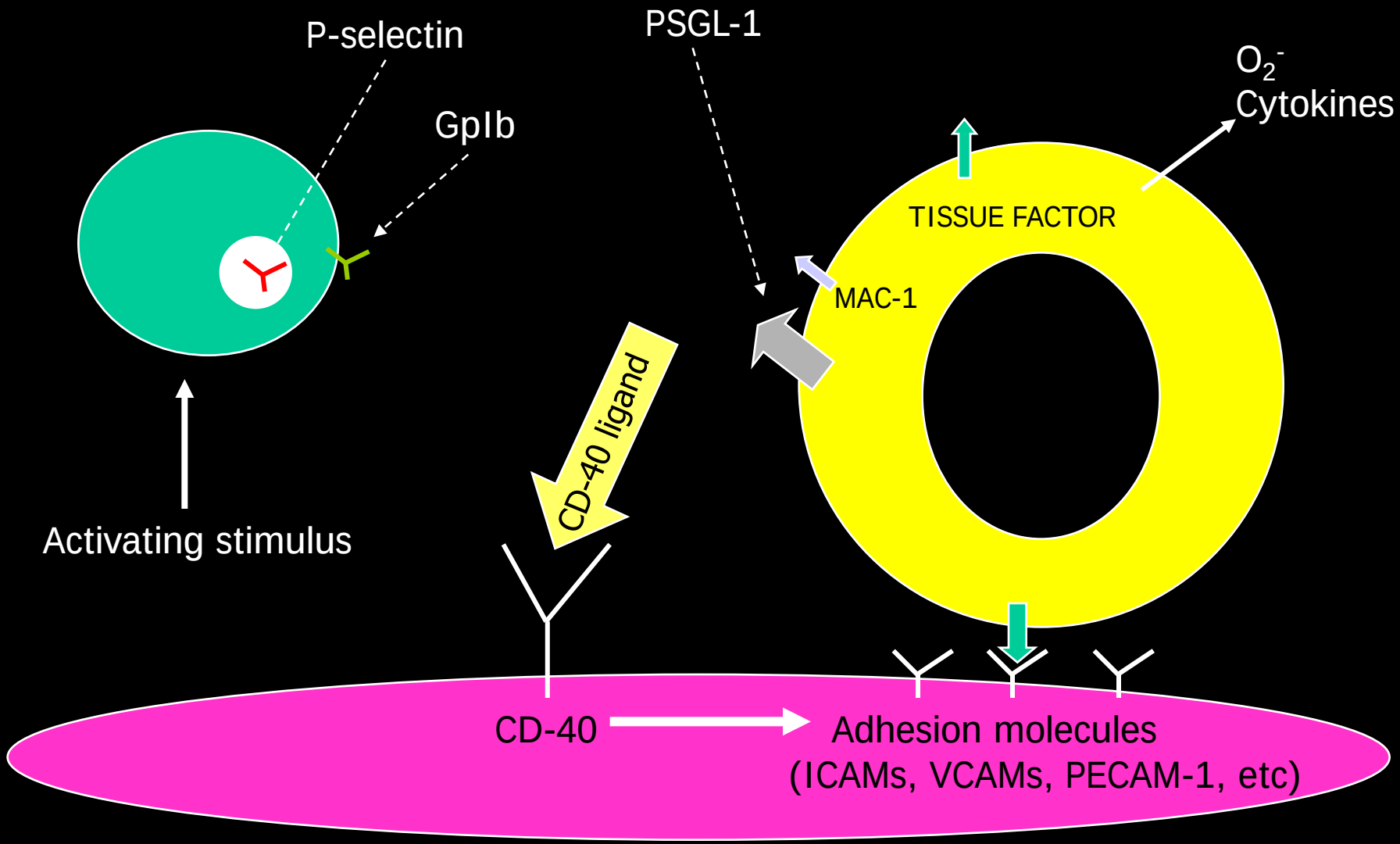
Huo Y. *et al. Nature Med* 2003;9:61-7

Wild-type (but not *Selp*-/- activated platelets or the supernatant of activated wild-type platelets) exacerbate atherosclerosis in *ApoE*-/- mice



Huo Y. *et al. Nature Med* 2003;9:61-7

Interaction of platelets, monocytes and endothelial cells



Can anti-platelet therapy prevent platelet-leucocyte interaction?

- Clopidogrel inhibits
  - platelet aggregation
  - expression of P-selectin
  - platelet-PMN adhesion
  - platelet-dependent ROS production in mouse PMN

- Pretreatment of human platelets with the active metabolite of clopidogrel in vitro inhibits
  - platelet P-selectin expression
  - platelet-PMN adhesion
  - production of ROS by PMN

Evangelista V. *et al. Thromb Haemost* 2005;94:568-77

Clopidogrel but not aspirin reduces formation of MPA in patients with atherosclerotic disease

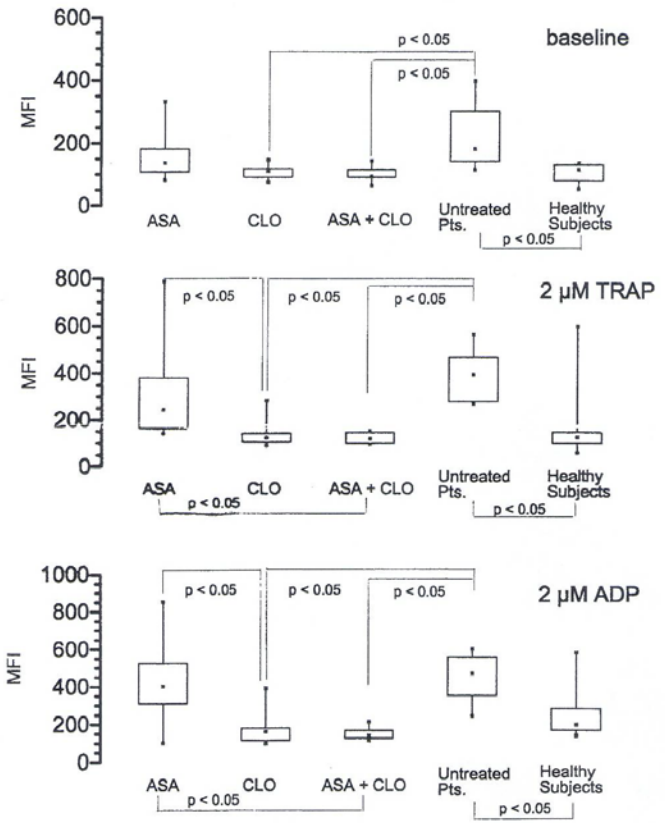


Fig 1. Formation of monocyte-platelet aggregates (mean fluorescence intensity of platelet mass attached to monocytes [MFI]) at baseline or after stimulation with 2-µmol/L thrombin receptor-activating peptide (TRAP) or 2-µmol/L adenosine diphosphate (ADP) in healthy subjects and patients with atherosclerotic vascular disease, either untreated or receiving treatment with aspirin (ASA) or clopidogrel (CLO) or their combination. *Box plots* indicate minimum, first quartile, median, third quartile, and maximum.

Klinkhardt U. *et al. Clin Pharmacol Ther* 2003;73:232-41

Conclusions (1): platelet activation in hypertension

- Circulating platelets show evidence (morphological, biochemical and functional) of activation, in essential hypertension
- One important manifestation is the formation of MPA
- Increased MPA formation in hypertension is at least partly related to deficient platelet NO, and anti-hypertensive treatment may reverse this
- MPA may contribute to progression of atherosclerosis and/or thrombosis

Conclusions (2): relevance of MPA

- ✓ Markers
  - Are sensitive marker of platelet activation
  - Are increased in patients with CV risk factors or with established atherosclerosis
- ✓ Mediators
  - Evidence that interaction of platelets with monocytes promotes adhesion to endothelium and activation of monocytes and endothelial cells
  - Prevention of such interaction inhibits atherogenesis in animal model of atherosclerosis

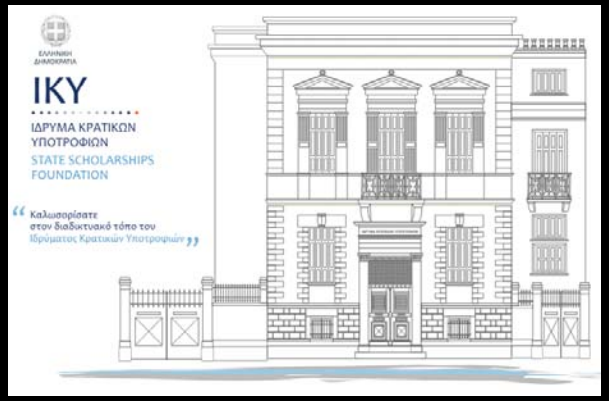
Conclusions (3): therapeutic opportunities

Inhibition of MPA formation may help prevent / retard atherosclerosis

- Aspirin – evidence weak (non-existent)
- Clopidogrel – more promising, needs further investigation

Grateful thanks to…

- Dr Eugenia Gkaliagkousi
- Dr Lindsay Queen
- Professor Ji Yong
- Iya Goubareva
- Dr Ashish Shah
- Dr Valerie Corrigall



British Heart Foundation