Pyschological Stress can induce Oxidative Stress-mediated Vascular Injury

Ick-Mo Chung, MD, PhD

Cardiology Division
Ewha Womans University

Contents

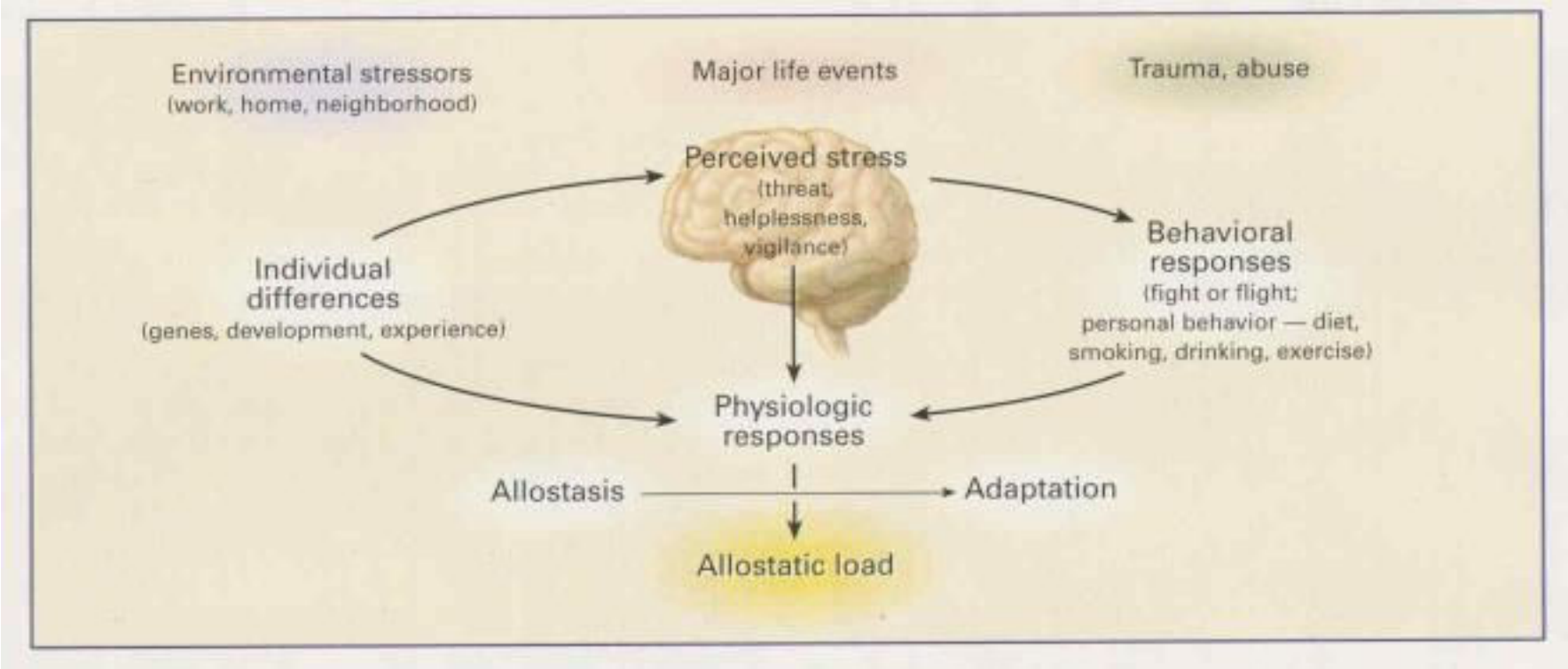
- Evidence of psychological stress as a risk factor for CAD
- Possible underlying mechanism by which psychological stress injure the vessel

  - Oxidative stress : ROCK, RGS

  - Inflammation

Allostatic Load

Allostasis: ability to achieve stability through change
Allostatic load: long term effect of physiological response to Stress



McEwen BS. N Engl J Med 1998;338:171

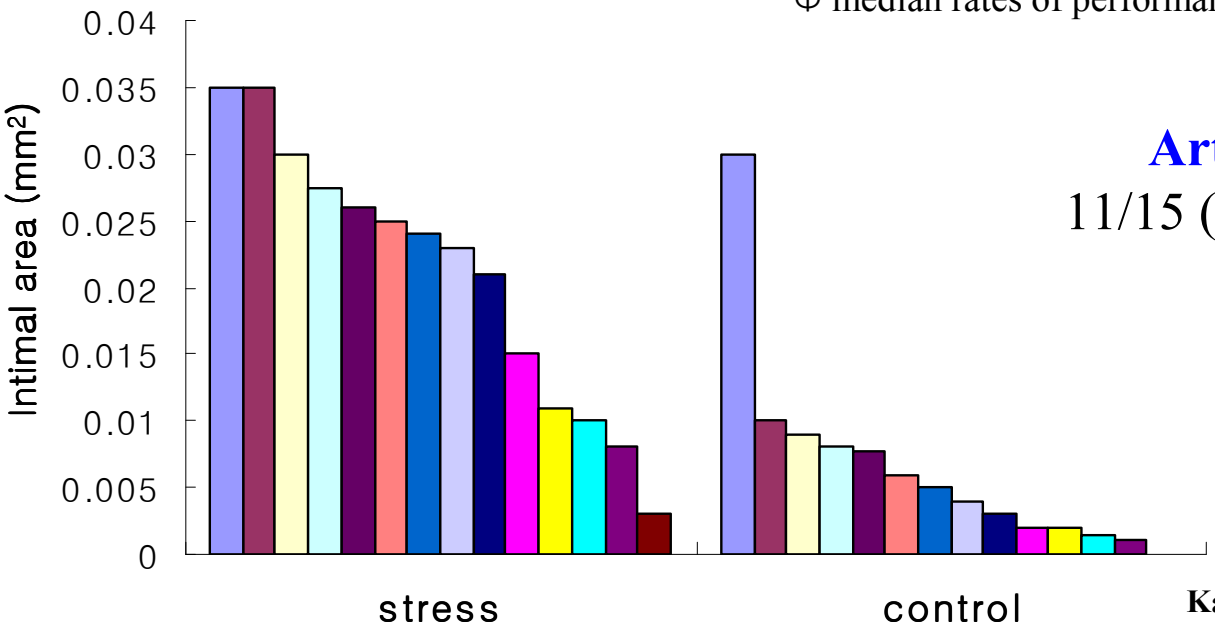
Psychological stress as a risk factor for CVD

- **Psychological stress is estimated as an emerging risk factor for atherosclerosis.**
  - **Stress: threatened homeostasis provoked by stressor.**
  - **Psychological factors contribute to the development and progression of CAD: depression, anxiety, personality and character traits, social isolation, and chronic life stress**
- **Physiological response to stress is mediated by the activation of sympathetic nervous system (SNS) and hypothalamic pituitary adrenal (HPA) axis.**
- **Stress can induce inflammation in peripheral organ through neuroendocrine system**

Social stress and Atherosclerosis in normocholesterolemic monkeys

	Rate of aggressionΦ	Severe aggression %	Rate of submission	Severe submission %	Time in affiliation %
Stress (n=15)	**5.16**	**30.0***	**6.59**	**27.0***	**21.0***
Control (n=14)	**6.69**	**20.0**	**7.10**	**19.0**	**26.0**

Φ median rates of performance per hour per monkey, * $p<0.05$



Arteries with plaques

11/15 (stress) vs 4/14 (control)

$p<0.05$

Kaplan JR. Science.1983;220:733-735

Anger proneness predicts CHD risk

Prospective analysis from the Atherosclerosis Risk In Communities (ARIC) Study

Williams JE. Circulation. 2000;101:2034-2039

Aim

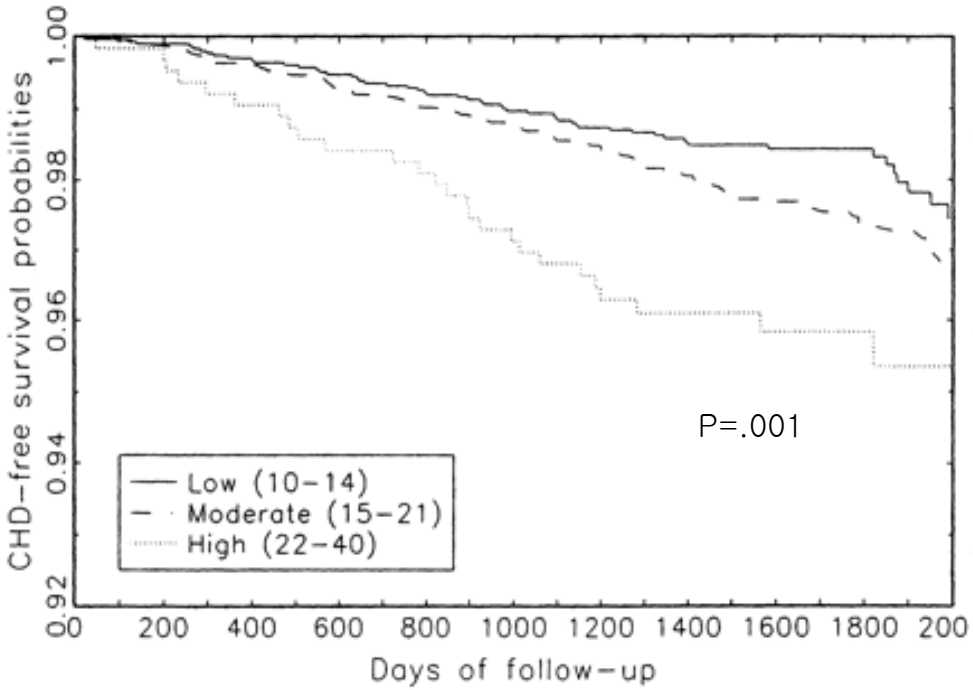
1. dose-response relation between anger and CHD risk
2. relation of CHD free survival with anger level

Methods

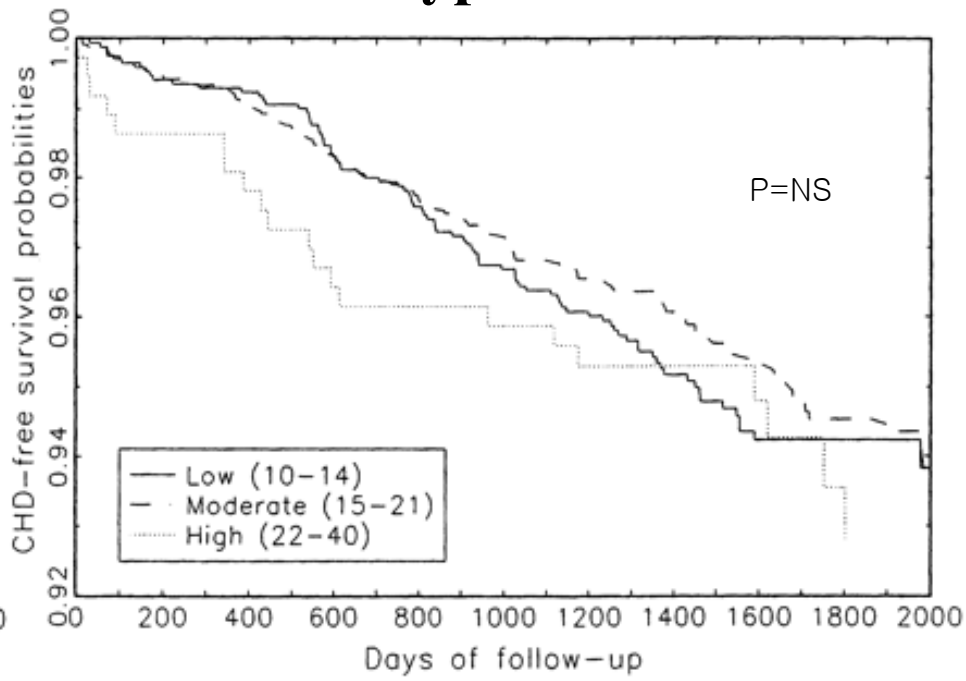
- Prospective design (median 53 mo, 6yr Max. f/u)
- ARIC study (n=13,208)
  white and black men and women (45-64 yr) without CHD
- 10-item Spielberger Trait Anger Scale
- CHD events: acute MI/fatal CHD (hard event), silent MI, revascularization procedure

CHD event free survival probabilities by Trait Anger scores

Normotensive



Hypertensive



Williams JE. Circulation. 2000;101:2034-2039

Intensive lifestyle changes for reversal of CHD

Ornish D. JAMA. 1998;280:2001-7

- **Lifestyle Heart Trial** (JAMA1983;249:54-9)

  Comprehensive lifestyle changes (1 yr) without lipid lowering drug could reduce LDL-chol by 37% and reduce frequency of angina attack by 91%

- **Aims**

  Effect of sustained lifestyle changes on risk factors, coronary atherosclerosis, and cardiac events after 4 additional yr.

- **Patients**: angiography-proven CAD Pts

- **Intensive Lifestyle Program**

  10% fat vegetarian diet, moderate aerobic exercise, stress management, smoking cessation, and group psychosocial support for 5 yrs

  avoid simple sugars, emphasize intake of complex carbohydrates and other whole foods

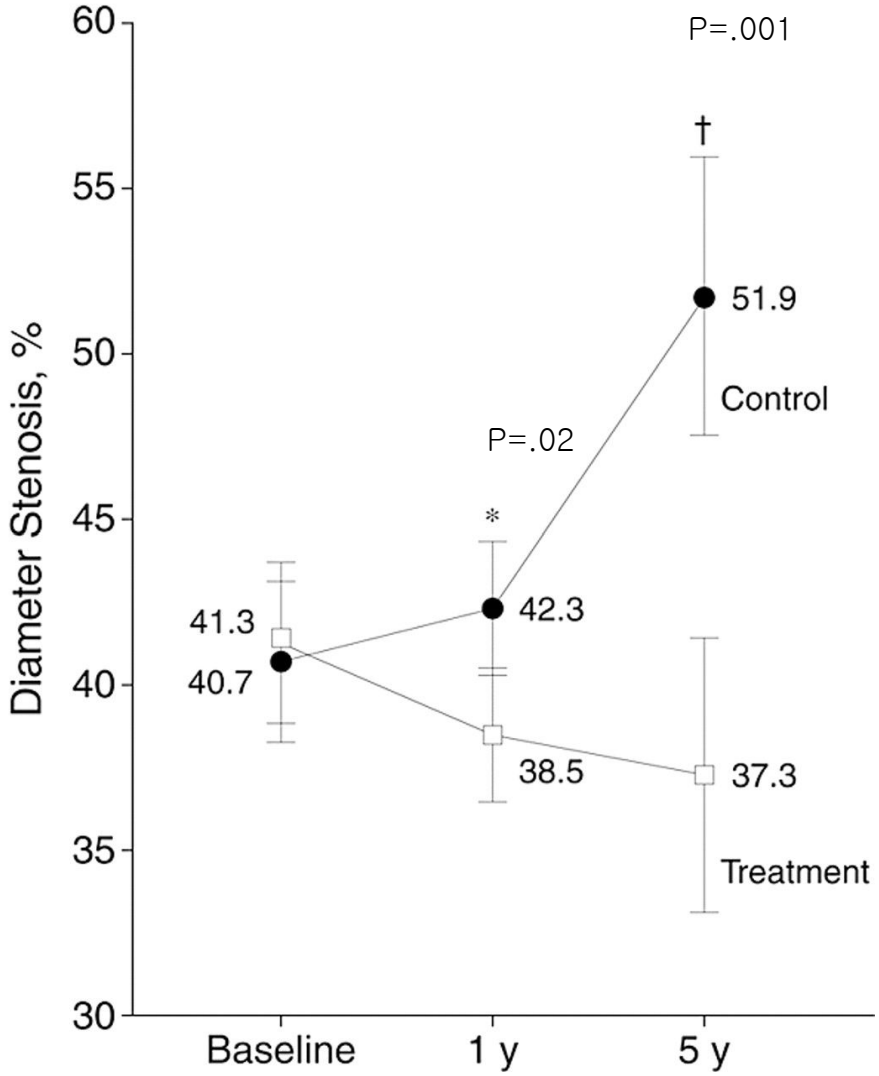
Angina symptoms

	Mean (SD) at Baseline		Mean (SD) at 1 Year			Mean (SD) at 5 Years		
	Experimental (n = 18)	Control (n = 14)	Experimental (n = 18)	Control (n = 14)	*P* Value* Baseline-1 Year	Experimental (n = 18)	Control (n = 14)	*P* Value* Baseline-5 Years
Chest pain frequency, times per week	5.8 (14.7)	1.4 (1.8)	0.5 (0.8)	4.0 (9.3)	.08	1.6 (2.7)	0.9 (1.9)	.32
Chest pain duration, min	3.1 (4.8)	3.2 (8.4)	1.8 (4.7)	7.6 (15.9)	.11	0.9 (1.3)	1.0 (2.7)	.93
Chest pain severity (1-7 scale)	1.5 (1.5)	0.6 (0.8)	0.7 (1.2)	1.4 (1.2)	<.001	0.9 (1.4)	0.6 (1.1)	.29

Cardiac events during 5 yr f/u

		No. of Events				
		Experimental* (n = 28)	Control† (n = 20)	Risk Ratio	95% Confidence Interval	*P* Value
	Myocardial infarction	2	4	2.74	0.393-30.3	.26
*	Percutaneous transluminal coronary angioplasty	8	14	2.40	0.939-6.60	<.05
	Coronary artery bypass graft	2	5	3.43	0.561-36.0	.14
*	Cardiac hospitalizations‡	23	44	2.62	1.55-4.55	<.001
	Deaths	2	1	0.685	0.012-13.2	.81
*	Any event	25	45	2.47	1.48-4.20	<.001

Mean percentage diameter stenosis



Ornish, D. et al. JAMA 1998;280:2001-2007.

Reduction in the incidence of type II DM with lifestyle intervention or metformin

Diabetes Prevention Program Research Group

N Engl J Med 2002;346:393

- **Question**

  Does a lifestyle intervention or Tx with metformin prevent or delay the onset of DM?

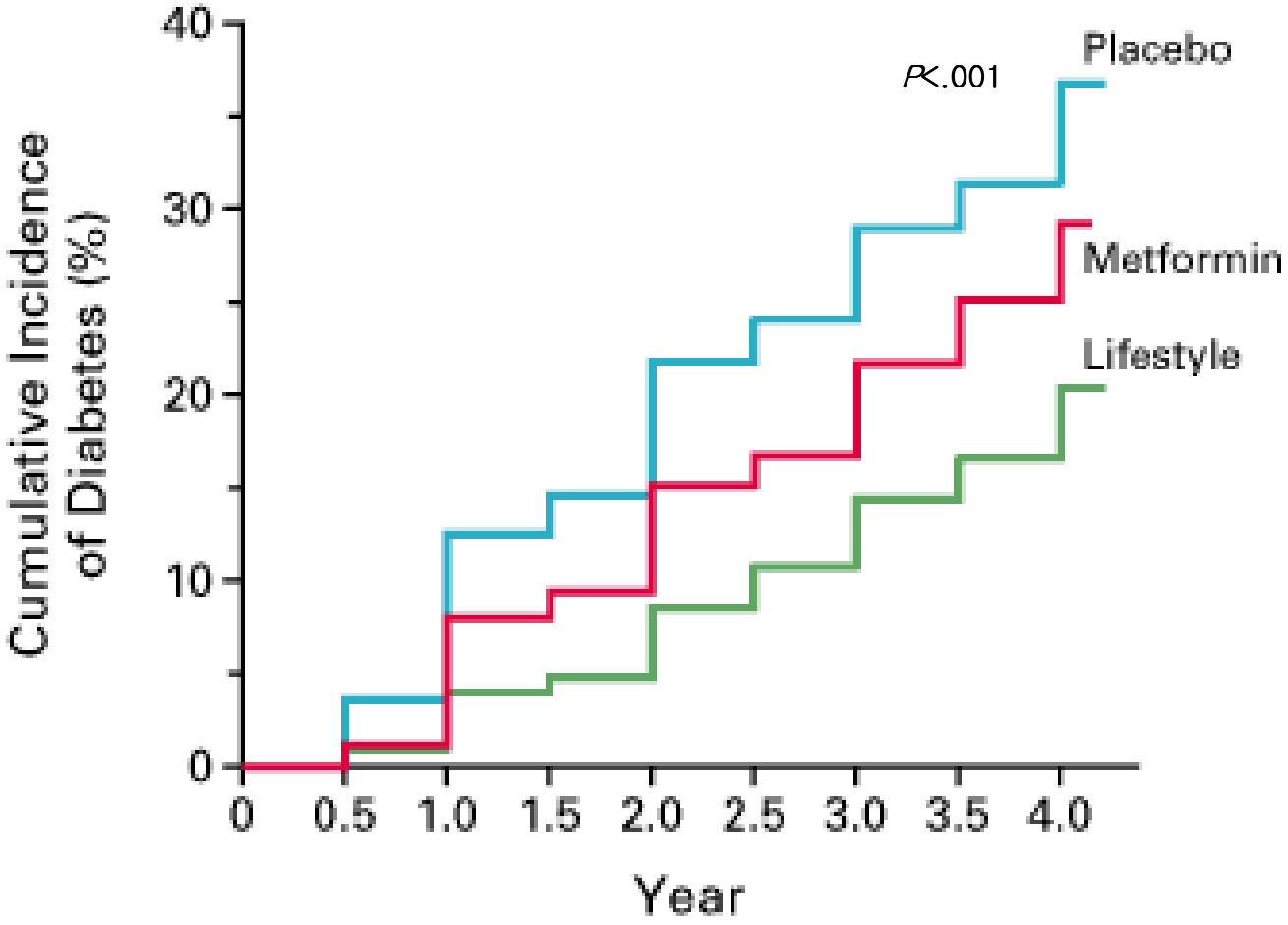
- **Inclusion criteria**

  BMI: ≥24,

  plasma glucose: (AC) 95-125 mg/dl, PC 140-199 mg/dl

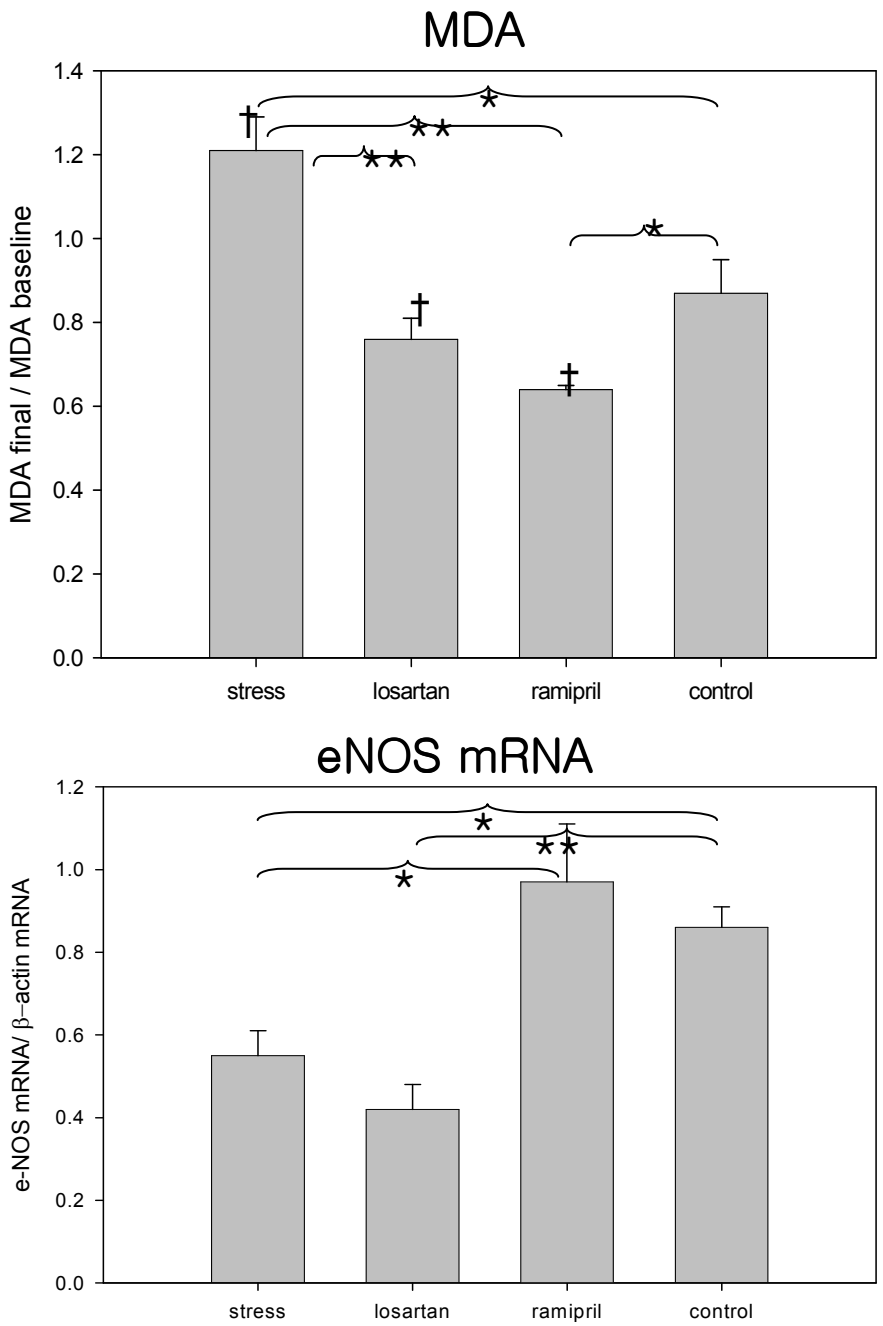
  - Standard lifestyle with metforrmin 850 mg bid
    : food guide pyramid NCEP step 1 diet
  - Intensive lifestyle modification
    16-session curriculum covering diet, exercise, and **behavior modification**, weight reduction >7% through a healthy low calorie, low fat diet, moderate physical activity s/a brisk walking 150 min /wk

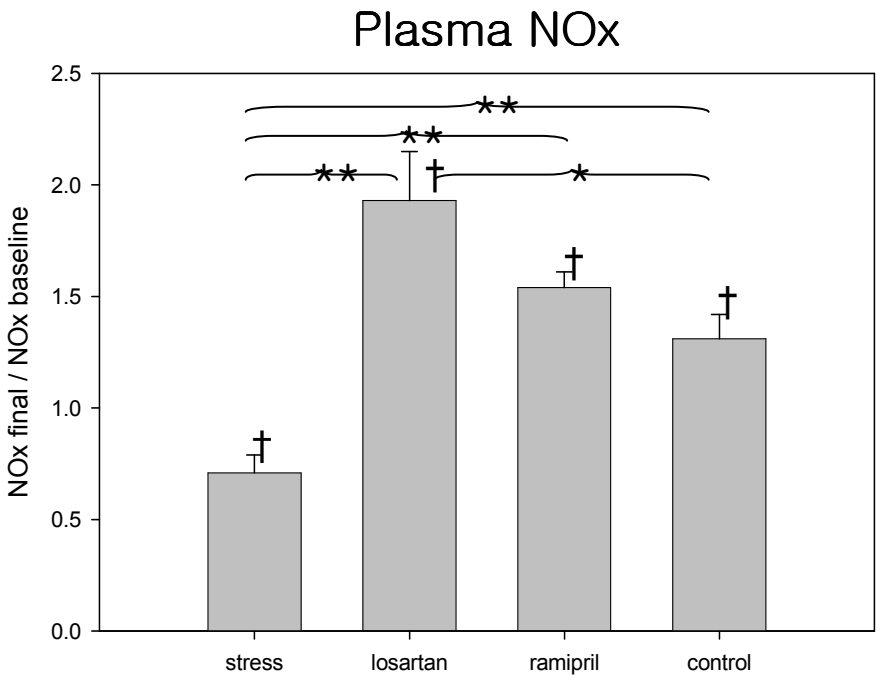
Cumulative incidence of DM



Diabetes Prevention Program Research Group
NEJM. 2002;346:393-403

Immobilization stress induces changes in ROS

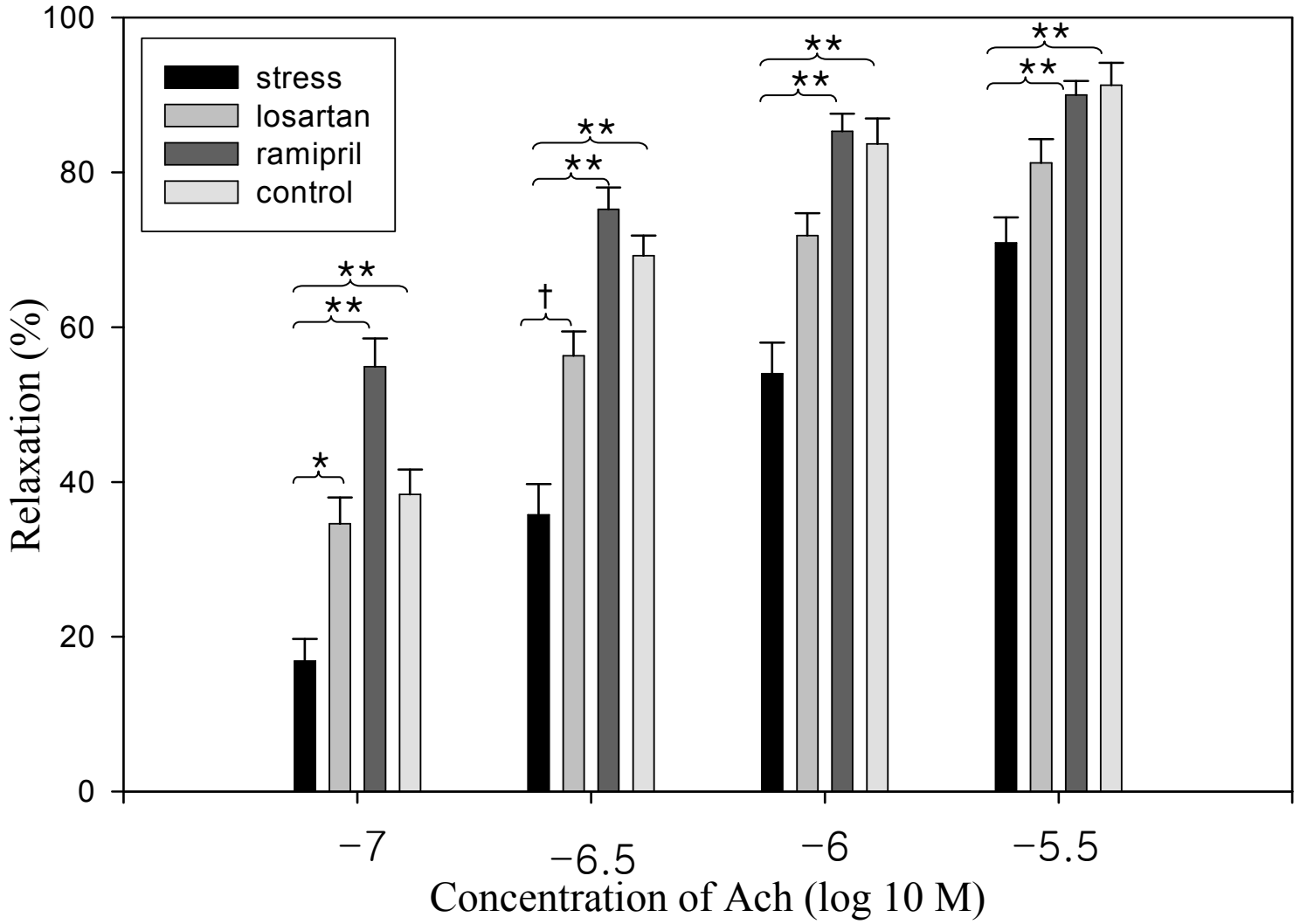




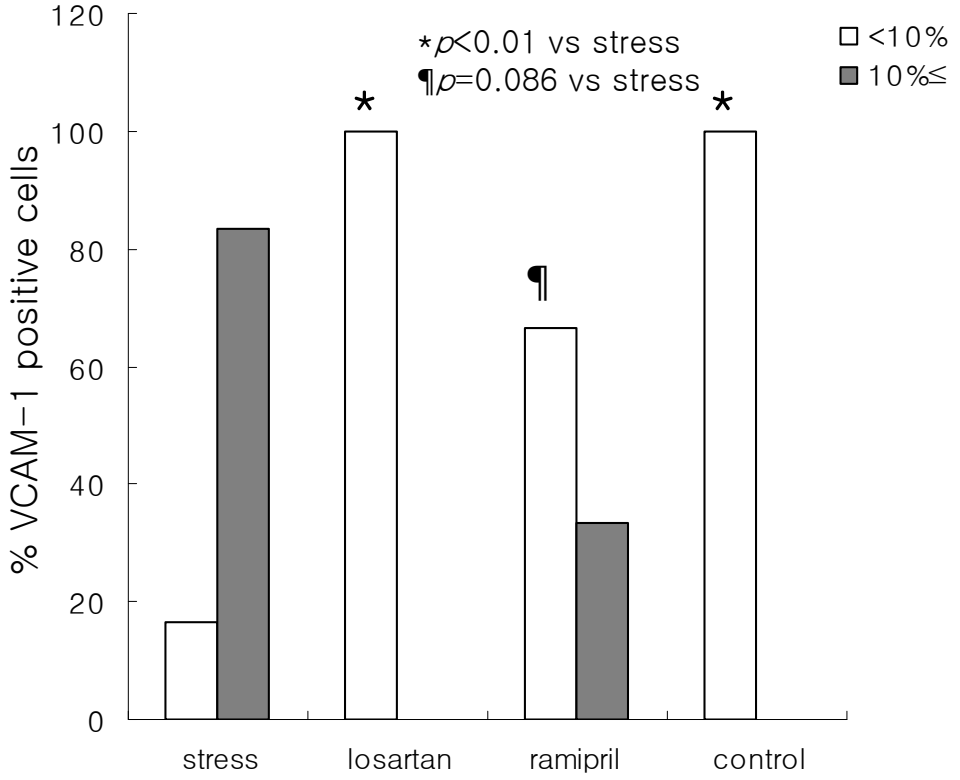
*p<0.05, **p<0.01,†p<0.05 for final vs baseline
M±SEM

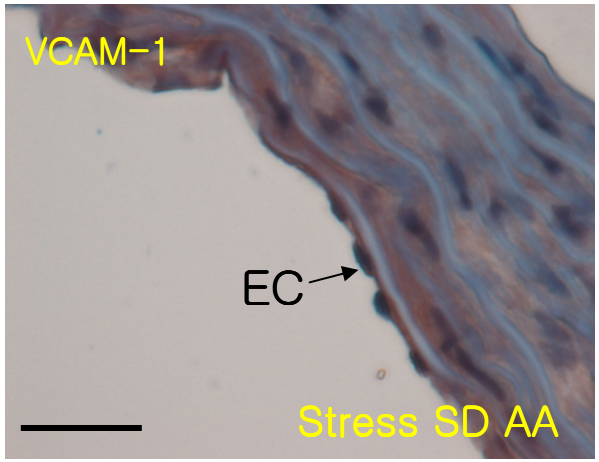
Chung I-M & Kim YM Circ Res 2004;93:1523 (abstr)
http://circres.ahajournals.org/cgi/data/94/12/1523/DC1/1

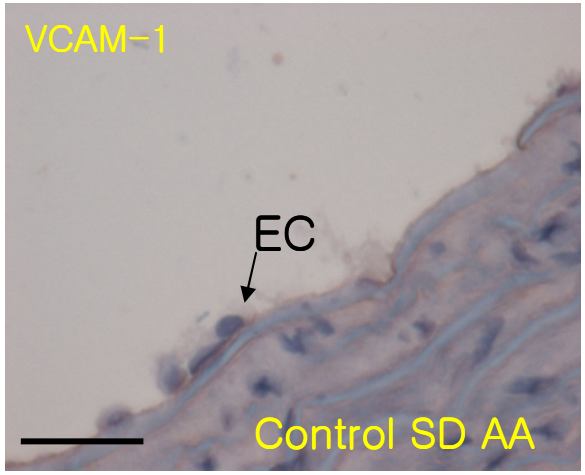
Changes in Ach-induced arterial relaxation



Immobilization stress can enhance expression of VCAM-1

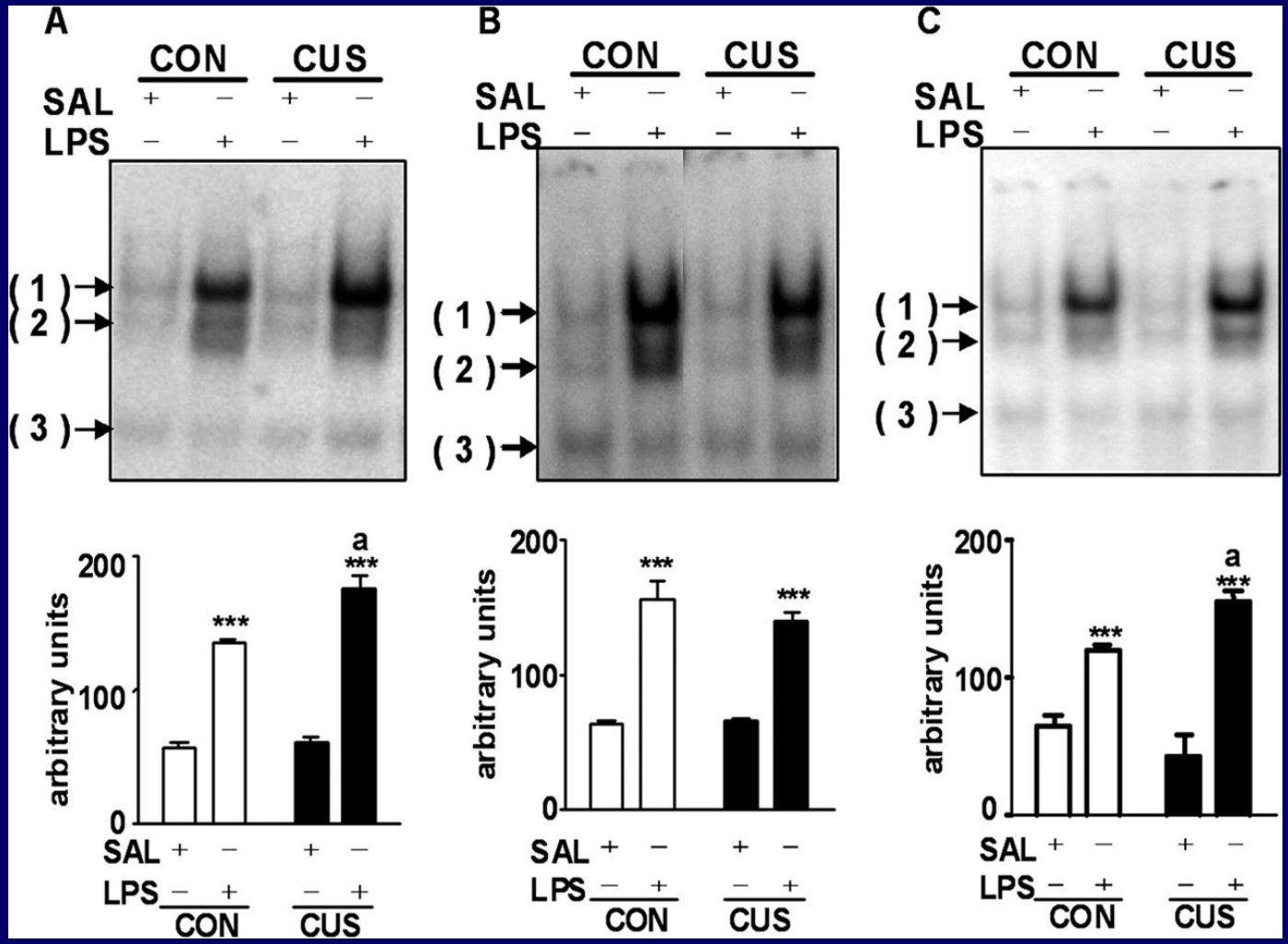






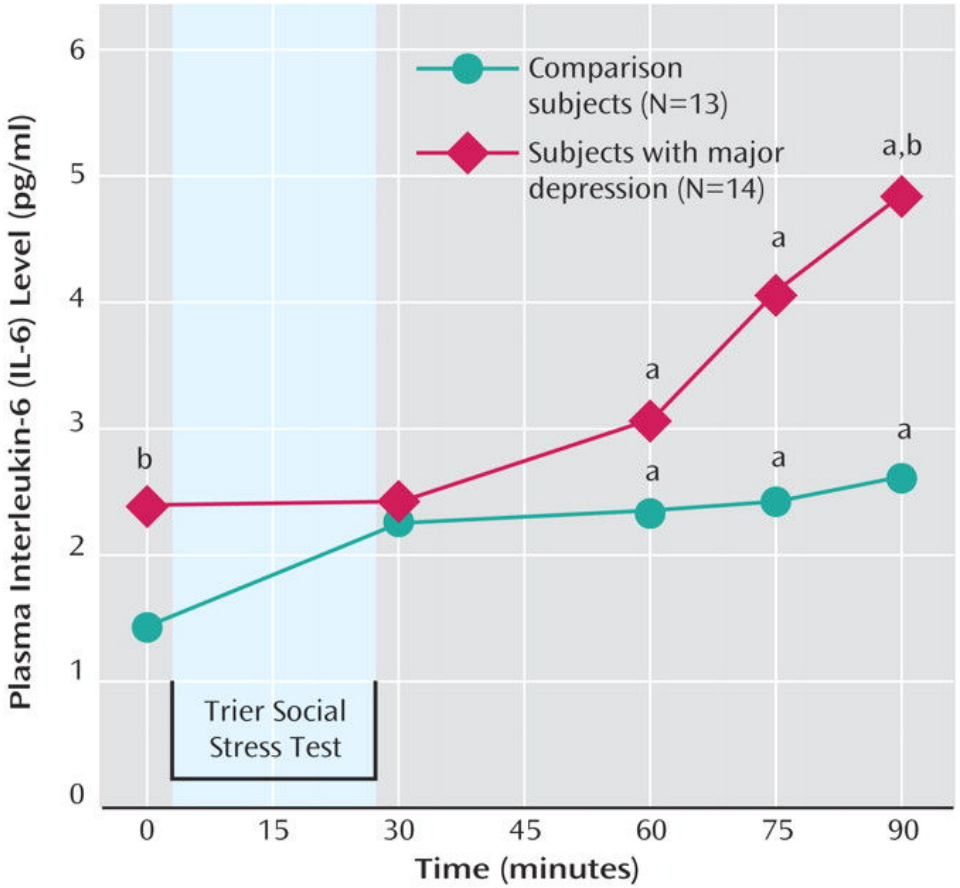
Bar: 50 μm

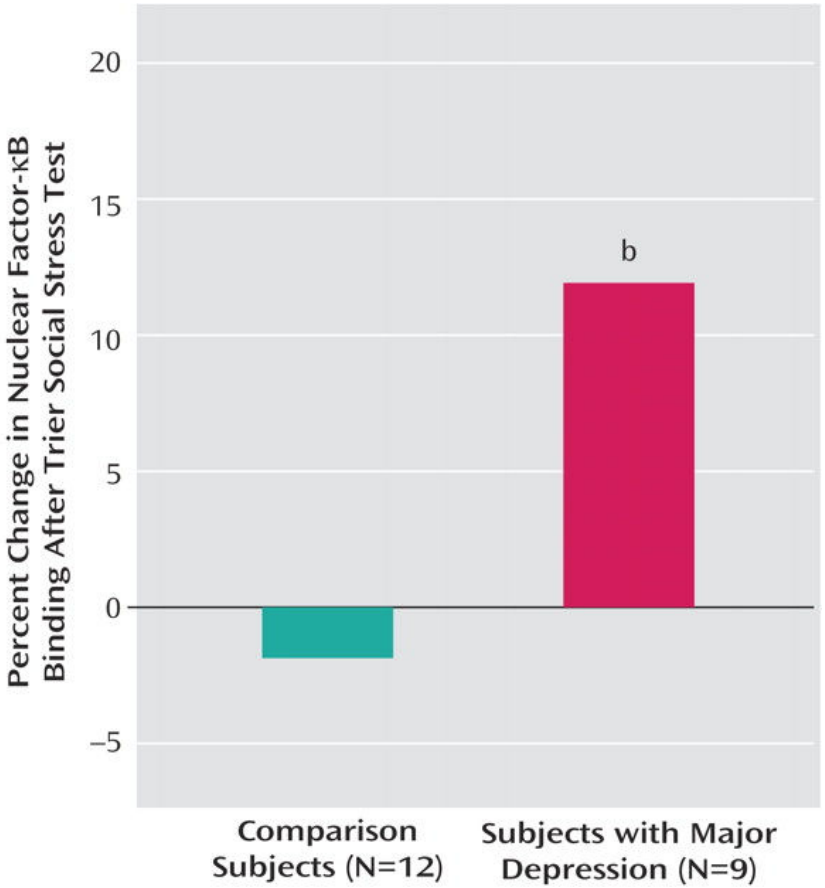
Effect of CUS on NF-κB activation induced by LPS or saline (SAL) in the frontal cortex (A), hypothalamus (B), or hippocampus (C)



Munhoz, C. D. et al. J. Neurosci. 2006;26:3813-3820

Plasma IL-6 and NF-κB DNA Binding in PBMC of Patients With Major Depression After a Psychosocial Stressor Challenge





Miller AH, Am J Psychiatry 2006;163:1630

Neurogenic Inflammation in Stress

- Both somatic and autonomic nerves are associated with inflammatory cells, and nervous transmission may result in neurogenic inflammation
  : Nervous innervation is a requirement for establishing certain inflammatory reaction: eg. rhematoid arthritis

- Direct linkage between neurogenic stimulation and proinflammatory cytokine release
  - contributing inflammatory mediators in stress
    PGE2: + SN ending
    Neuropeptide Y: cotransmitter of SN innervation
    promotes SMC proliferation, enhances leukocyte adhesion, platelet aggregation, and MΦ activation
    CRF: + in SN ending, sensory N, spinal cord
    Substance P: + in autonomic N, ganglia, spinal cord,
    IL-6: + in sensory N, autonomic N

Vascular Injury mediated through Oxidative Stress

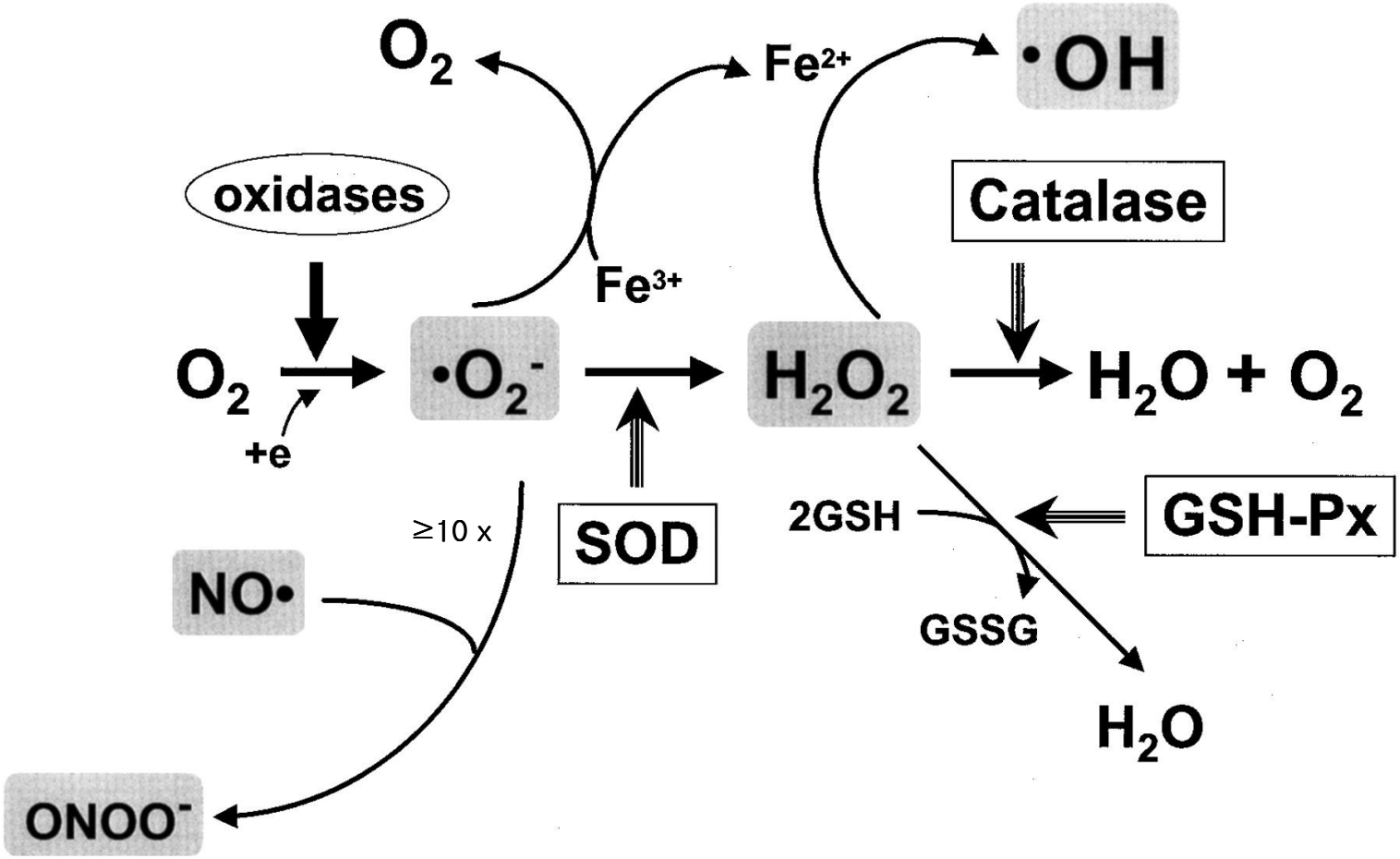
- **Response to injury Hypothesis**

  Injury to the endothelium might precipitate the atherosclerotic process (Virchow R, 1856)

  Atherosclerotic lesions result from an excessive inflammatory-fibroproliferative response various forms of insult to the endothelium and smooth muscle of the artery wall (Ross R,Nature. 1993)

- **Oxidative stress**, an excessive production of ROS outstripping antioxidant defense systems, has been implicated in many pathophysiological conditions in cardiovascular systems (atherosclerosis, hypertension, restenosis, smoking, hypercholesterolemia, DM, hypertension, heart failure), cancer, and neurodegeneration.

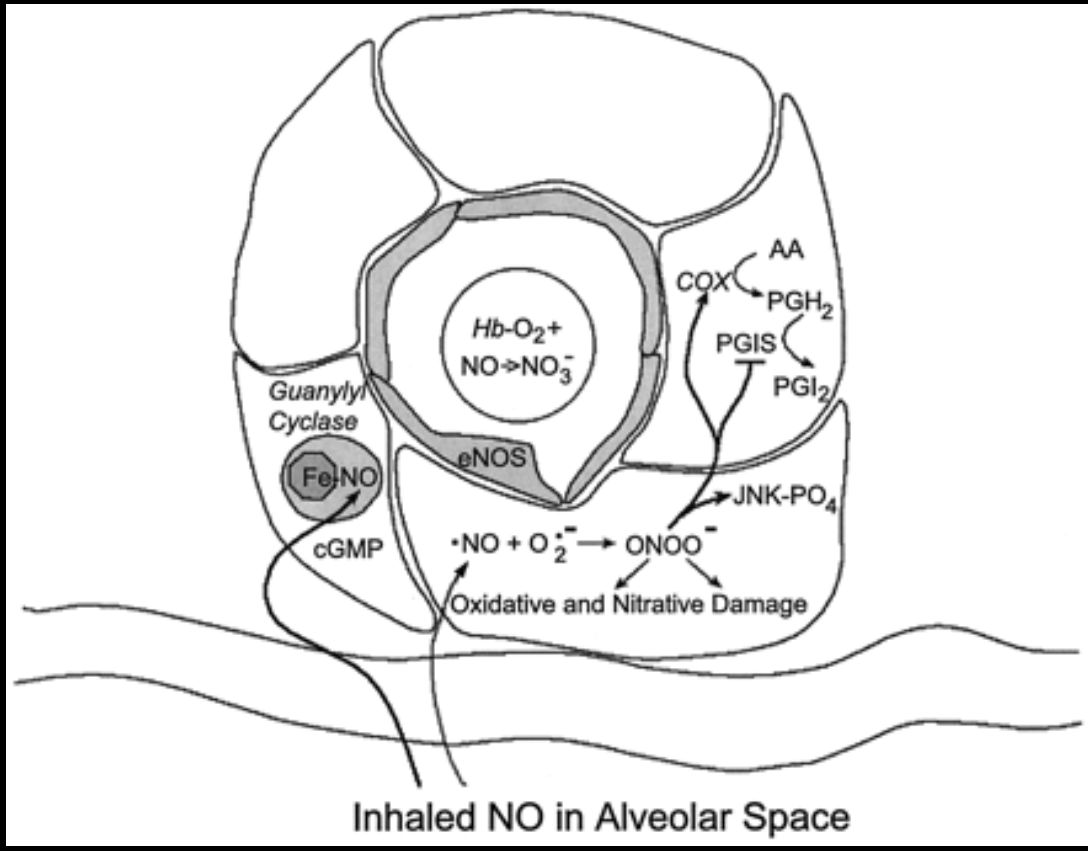
Vascular ROS



Griendling KK et al. Circulation.2003;108:1912-6

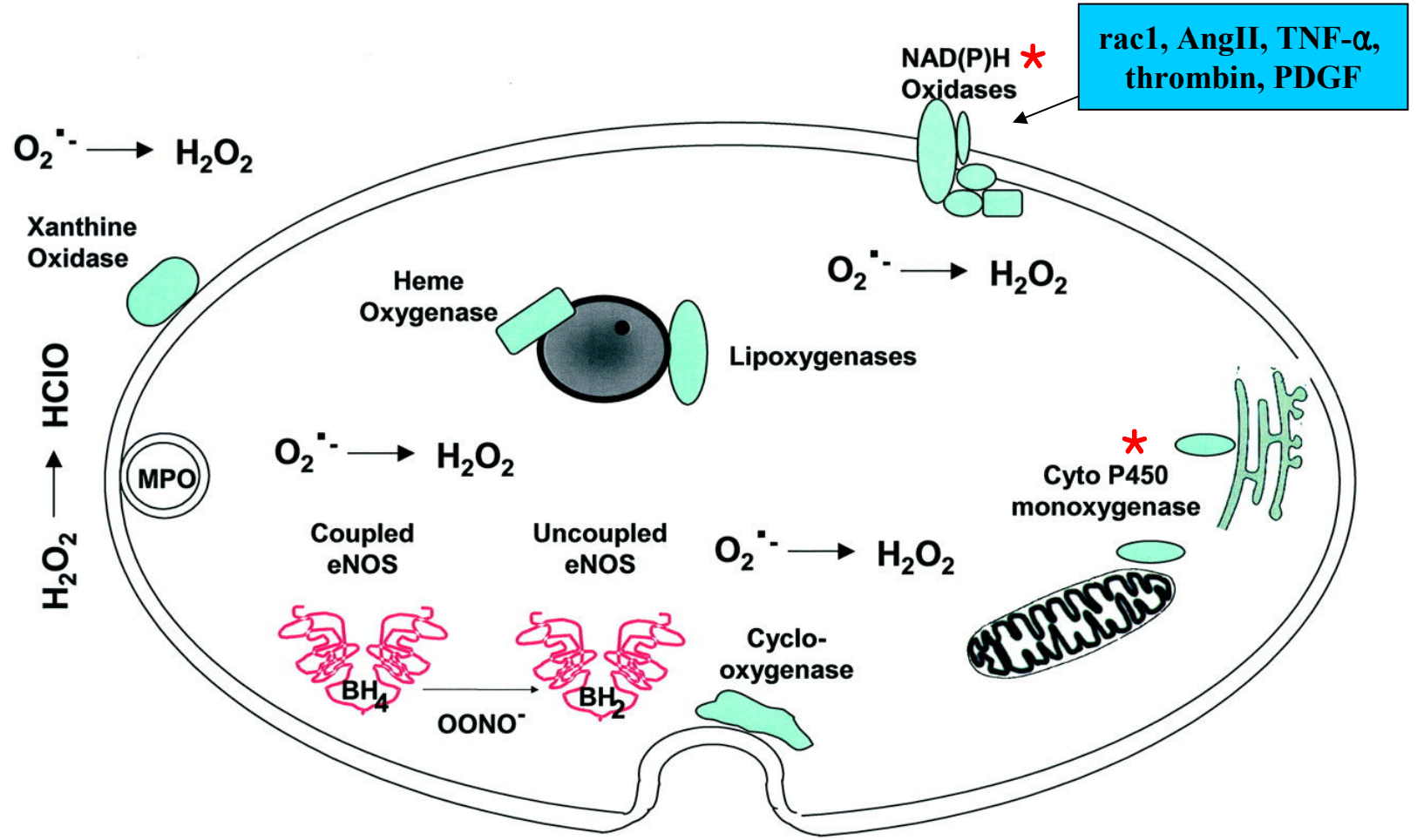
Peroxynitrite ($ONOO^-$)

- strong oxidant: oxidative damage of DNA, RNA, proteins, lipids
- Mediator of protein nitration
- Promotes inflammatory synthesis of prostaglandin by activating cyclooxygenase (COX)
- Attack prostacyclin synthase (Tyr nitration ?): Accumulating PGH_2 can activate thromboxane receptors, thereby promoting vasoconstriction
- Deactivate anti-inflammatory agents
- Activate c-Jun-NH_2-terminal kinase (JNK) thereby inducing a wide range of stress-related responses including apoptosis
- Protective role: microbicidal agent, limit bleeding



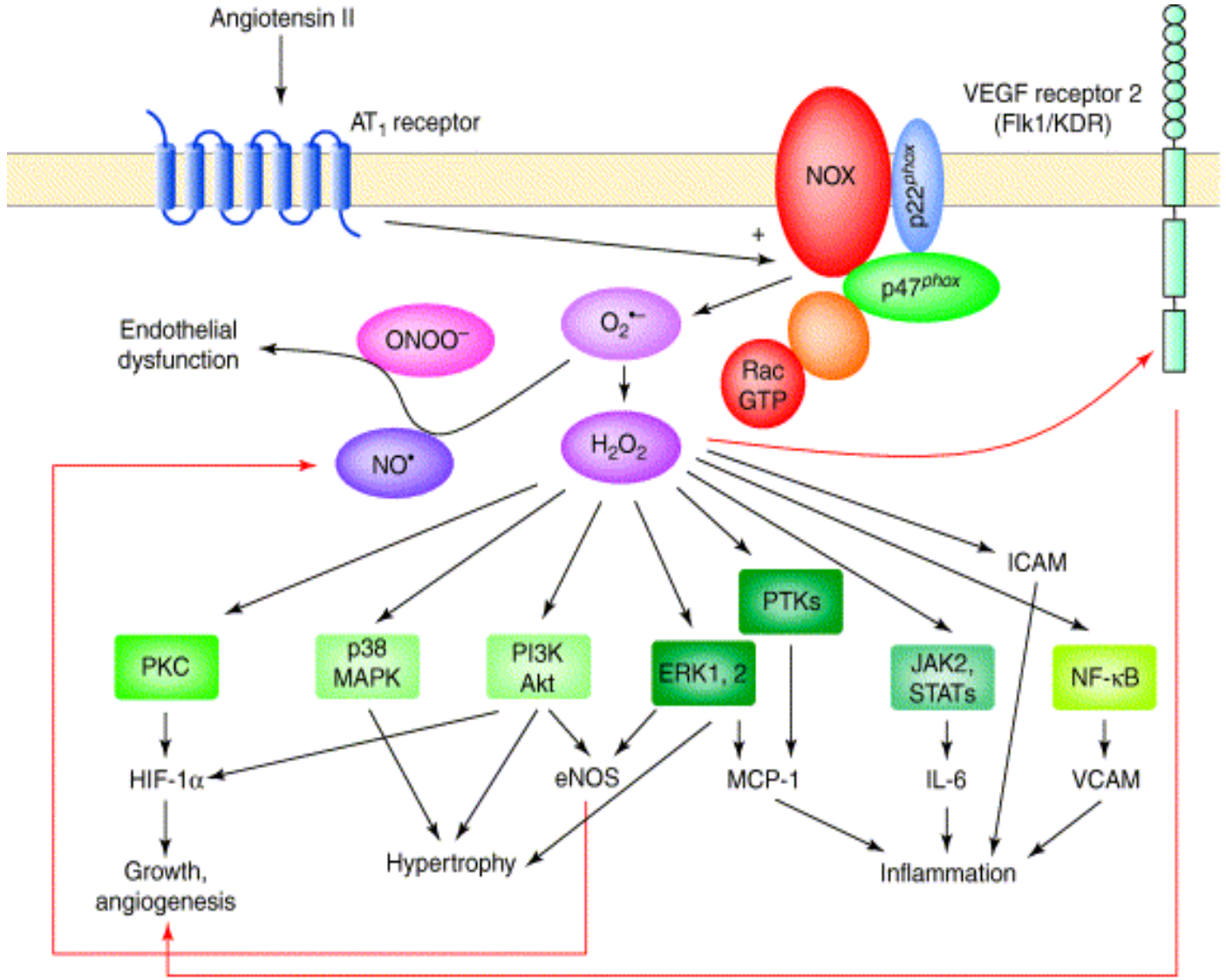
Beckman JS, Circ Res 2001;89:295

Potential sources of ROS



Griendling KK et al. Circulation.2003;108:1912-6

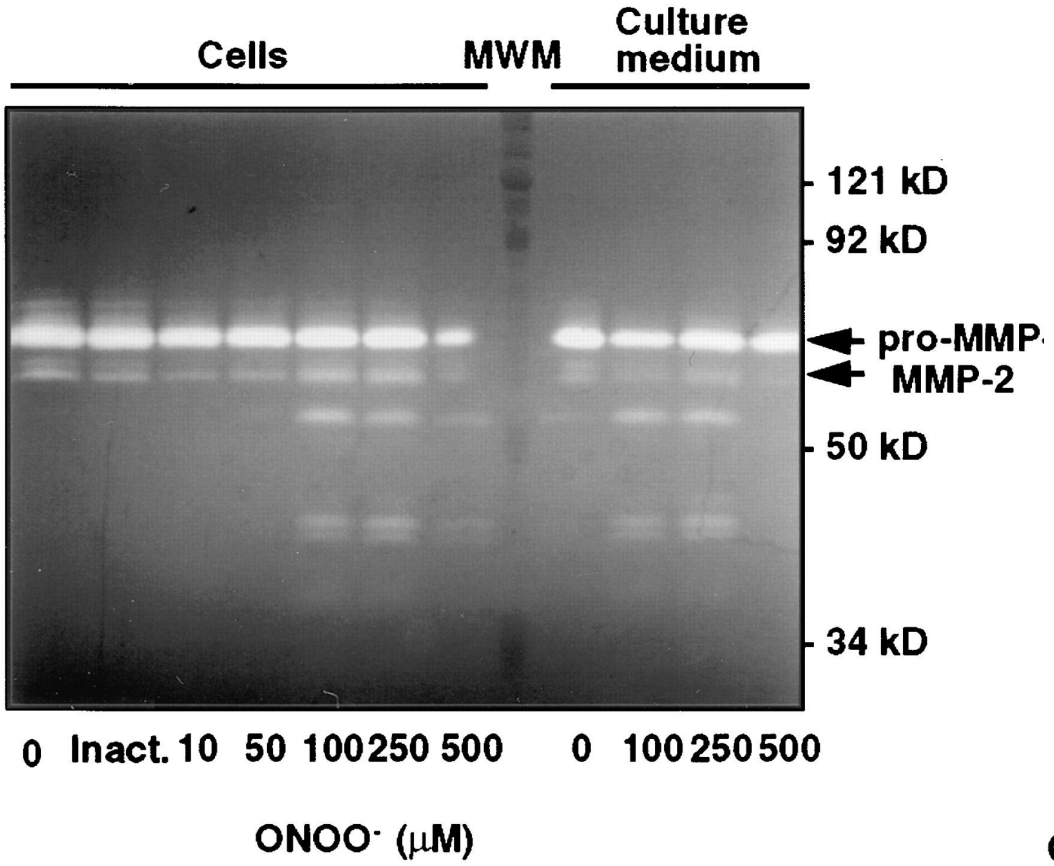
Consequences of NAD(P)H oxidase activation



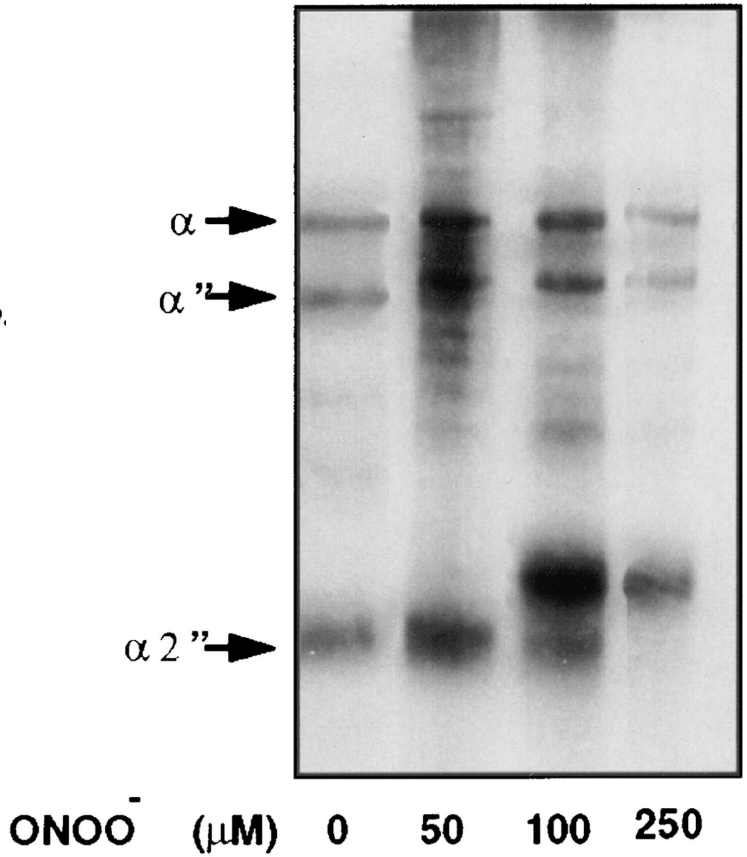
Harrison DG, 2003;24:471-478

Peroxynitrite can increase MMP2 activity

Effect of $ONOO^-$ on gelatinolytic activity in human SMCs



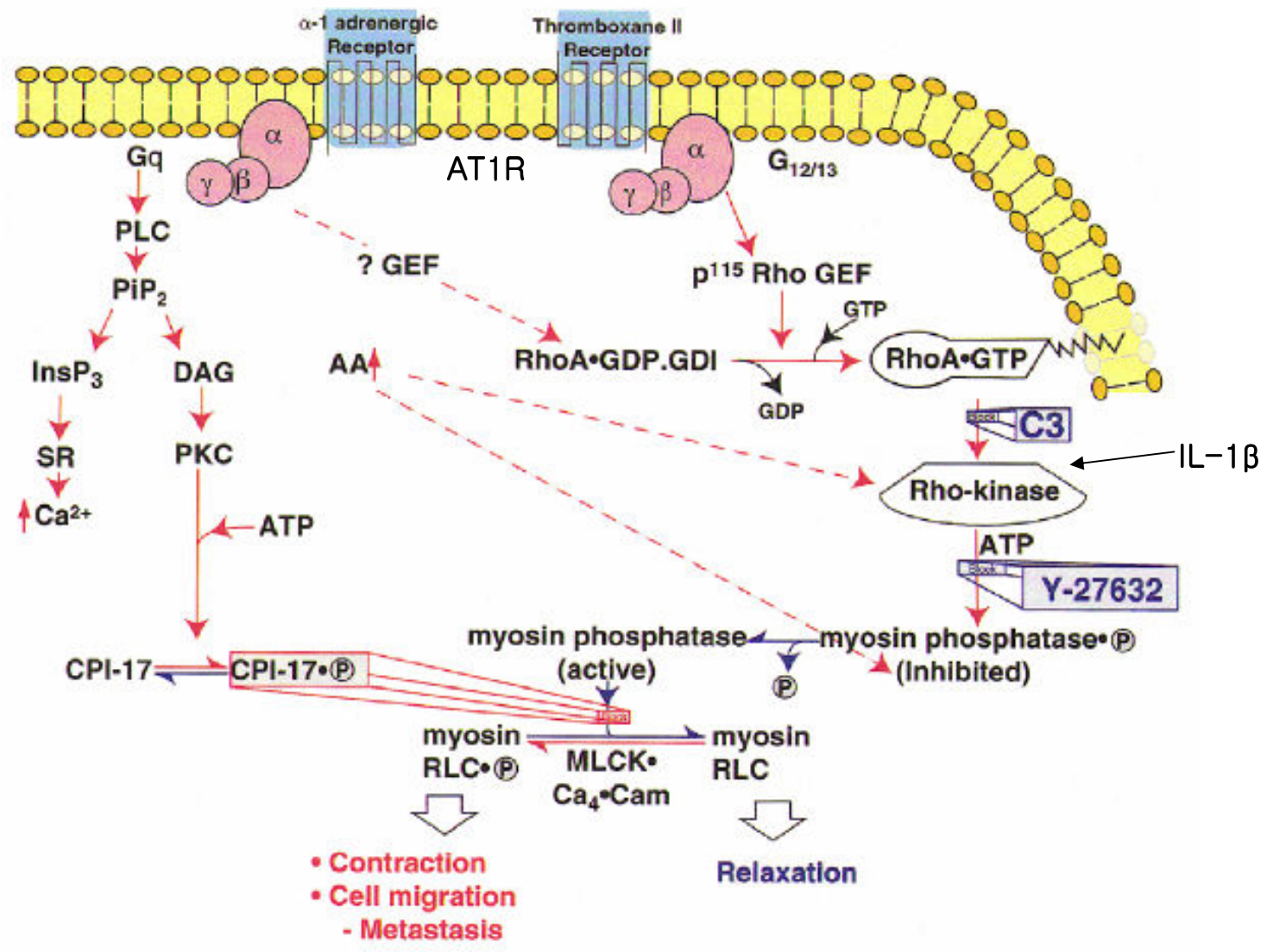
Effect of $ONOO^-$ on collagenolytic activity in human SMCs



Fluorographic image of ^3H-type IV collagen

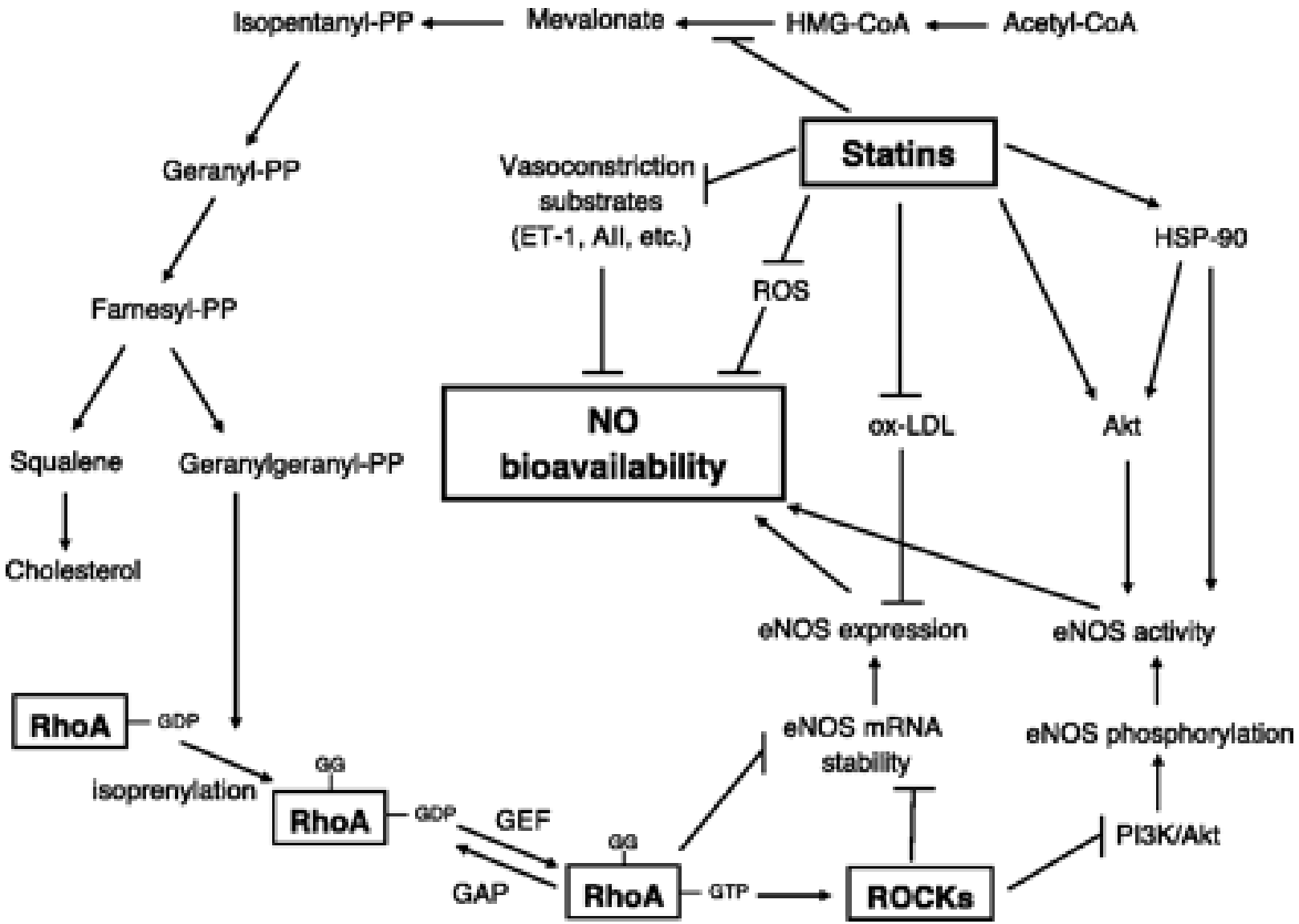
Gallis ZS, J Clin Invest 1996;98:2572

Regulation of myosin II in SMC and non-muscle cells



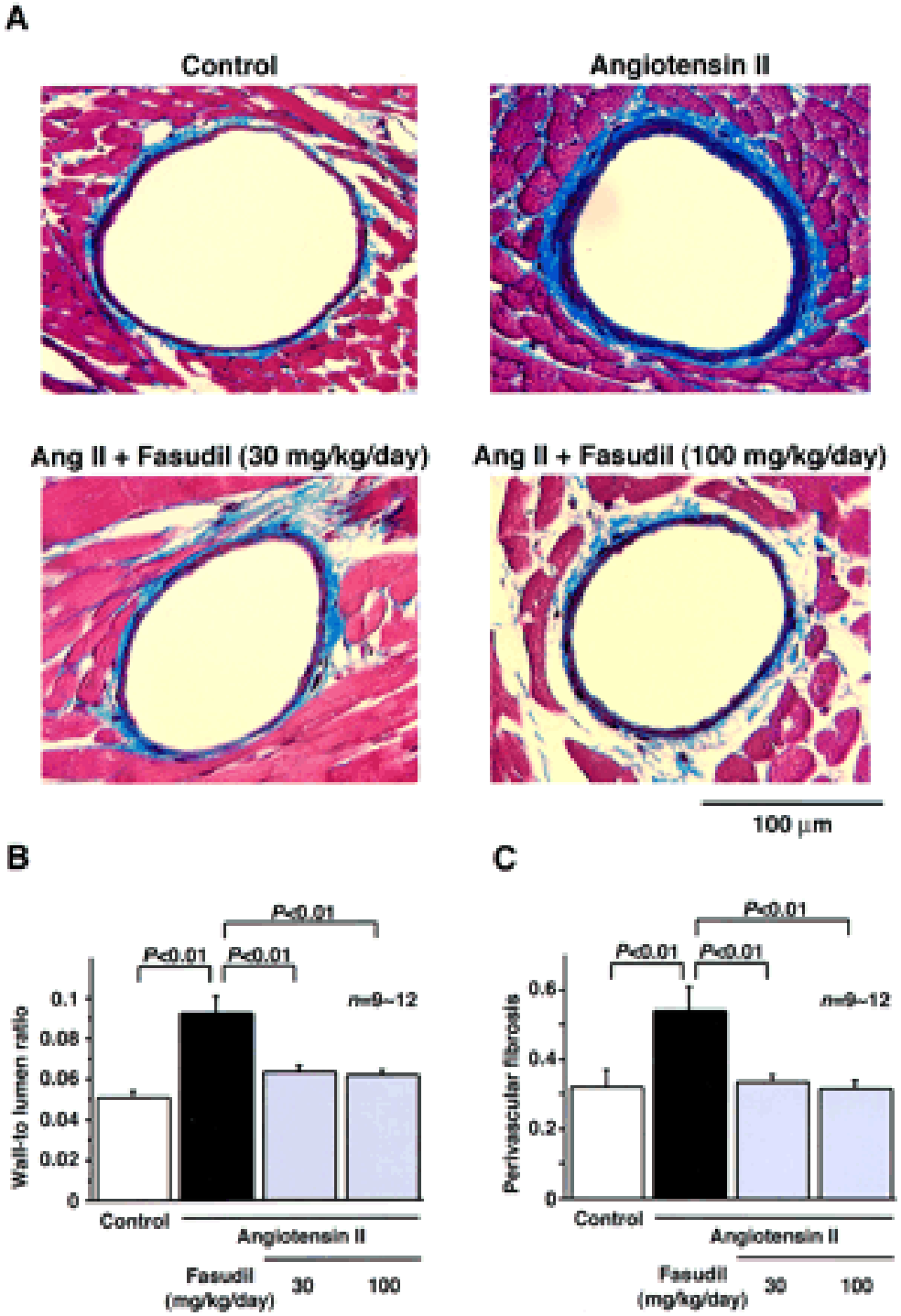
Somlyo AP. J Phys 2000;522:1777

Regulation of eNOS by ROCK



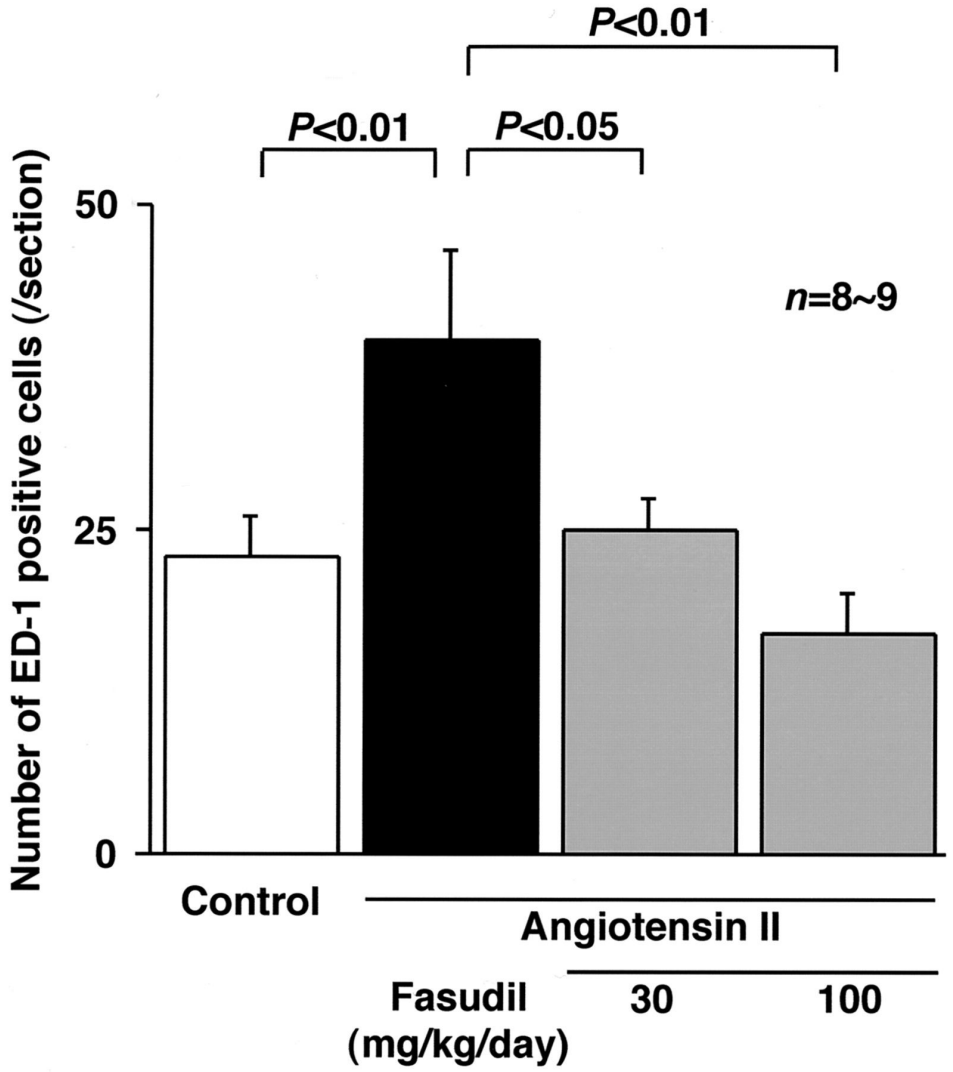
Liao JK, Am J Physiol Cell Physiol 290: C661-C668, 2006

Ang II-mediated coronary vascular hypertrophy: role of Rho-kinase



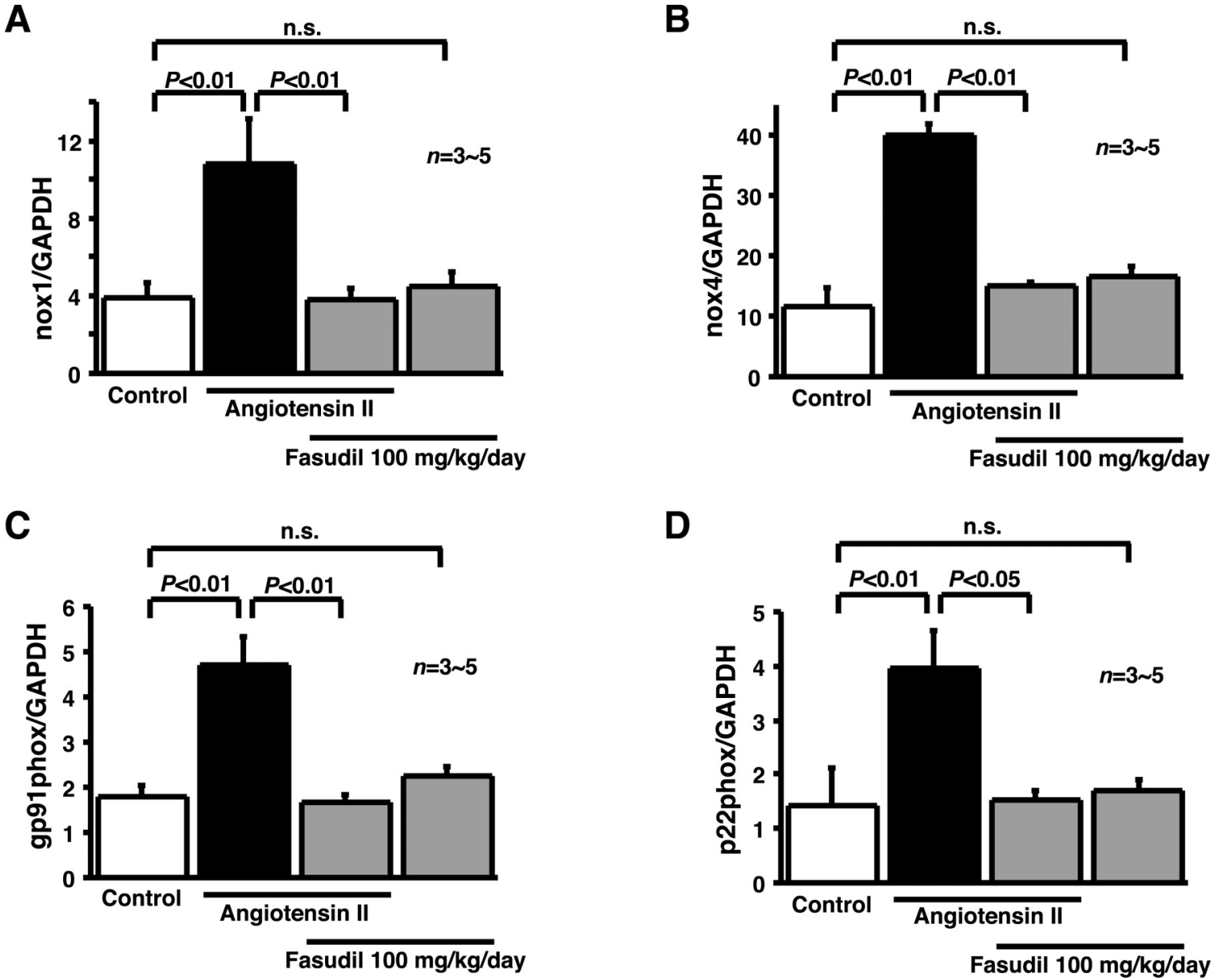
Shimokawa H. Circ Res 2003;93:767-75

Ang II-mediated vascular macrophage accumulation
Role of Rho-kinase



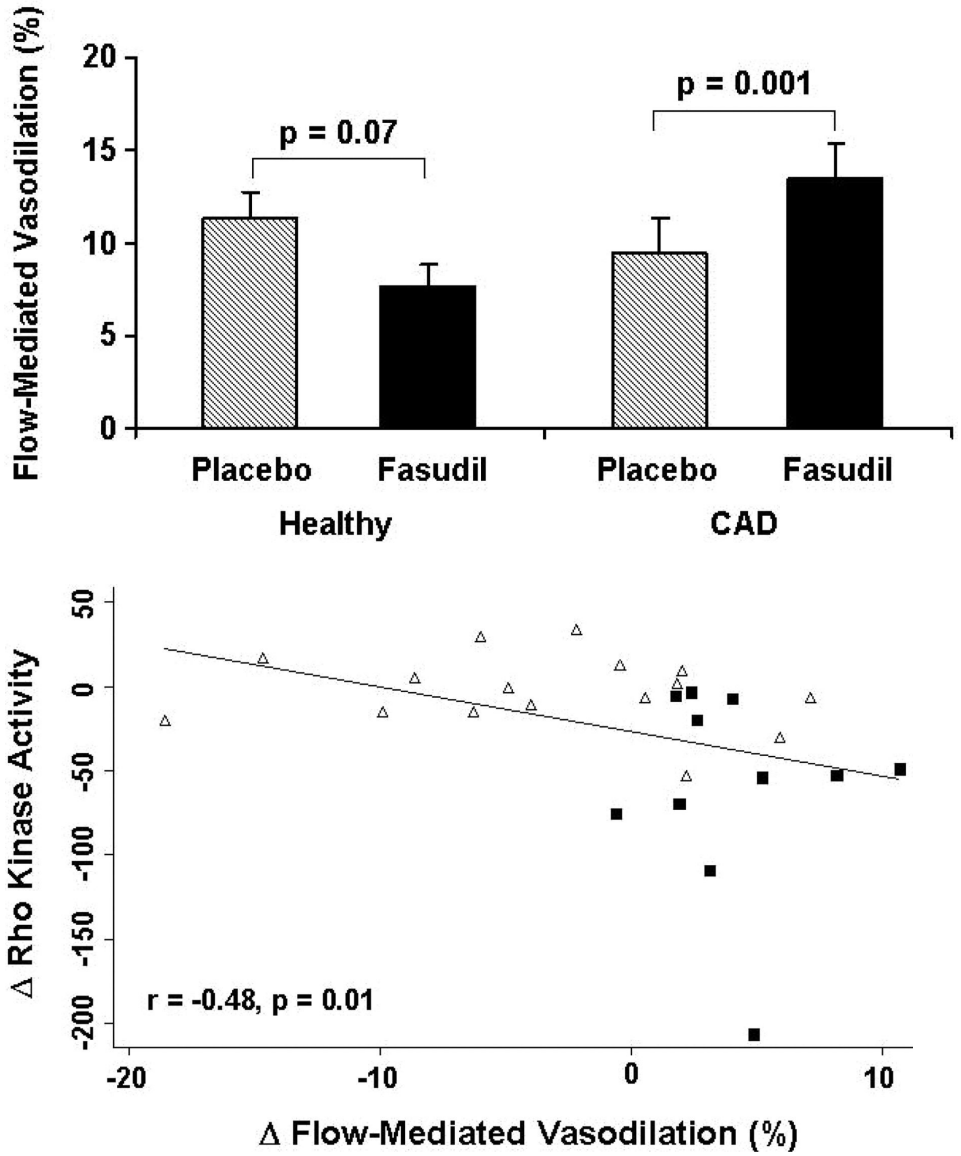
Shimokawa H. Circ Res 2003;93:767-75

Ang II-mediated NADPH oxidase: Role of Rho-kinase



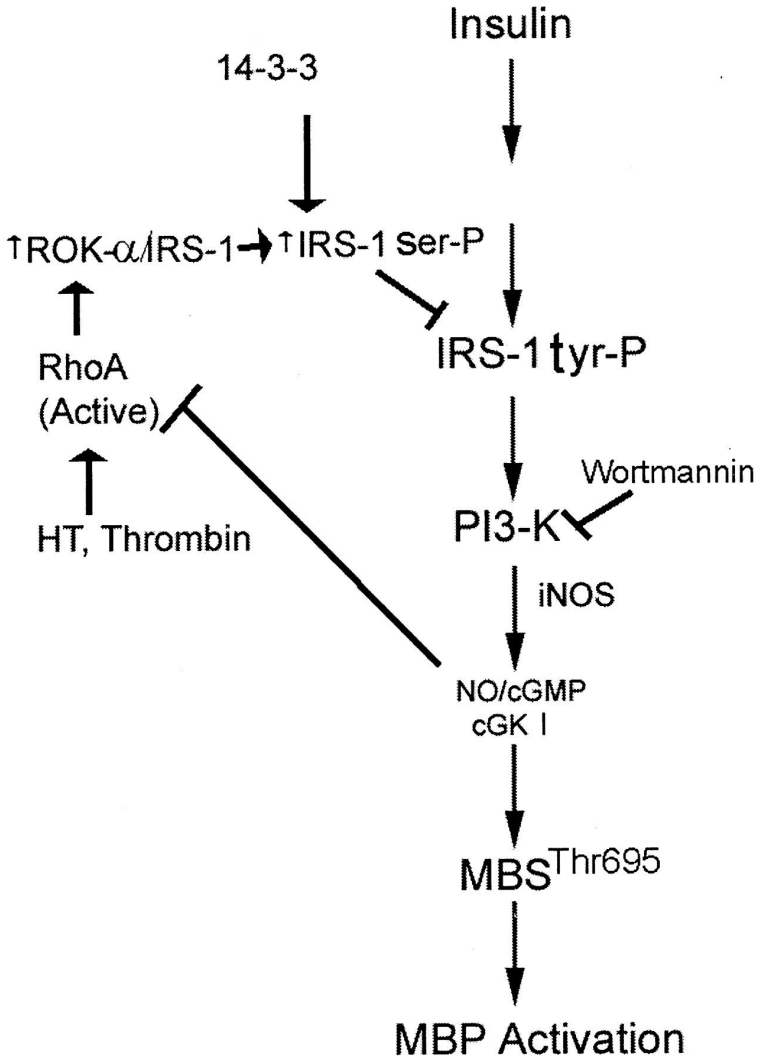
Shimokawa H. Circ Res 2003;93:767-75

ROCK inhibition improves endothelial function in CAD



Nohria A. et al. Circ Res. 2006;99:1426

Active ROCK-α associates with Insulin Receptor Substrate-1 and inhibits insulin signaling in VSMCs



Begum N, J Biol Chem 2002;277:6214

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Journal of Molecular and Cellular Cardiology 37 (2004) 537–546

www.elsevier.com/locate/yjmcc

Original Article

Inflammatory stimuli upregulate Rho-kinase in human coronary vascular smooth muscle cells

Junko Hiroki [a], Hiroaki Shimokawa [a,*], Midoriko Higashi [a], Keiko Morikawa [a], Tadashi Kandabashi [a], Natsumi Kawamura [a], Toru Kubota [a], Toshihiro Ichiki [a], Mutsuki Amano [b], Kozo Kaibuchi [b], Akira Takeshita [a]

[a] *Department of Cardiovascular Medicine, and the 21st Century COE Program on Lifestyle-Related Diseases, Kyushu University Graduate School of Medical Sciences, 3-1-1 Maldashi, Higashi-ku, Fukuoka 812–8582, Japan*

[b] *Department of Cell Pharmacology, Nagoya University Graduate School of Medicine, Nagoya, 466–8550, Japan*

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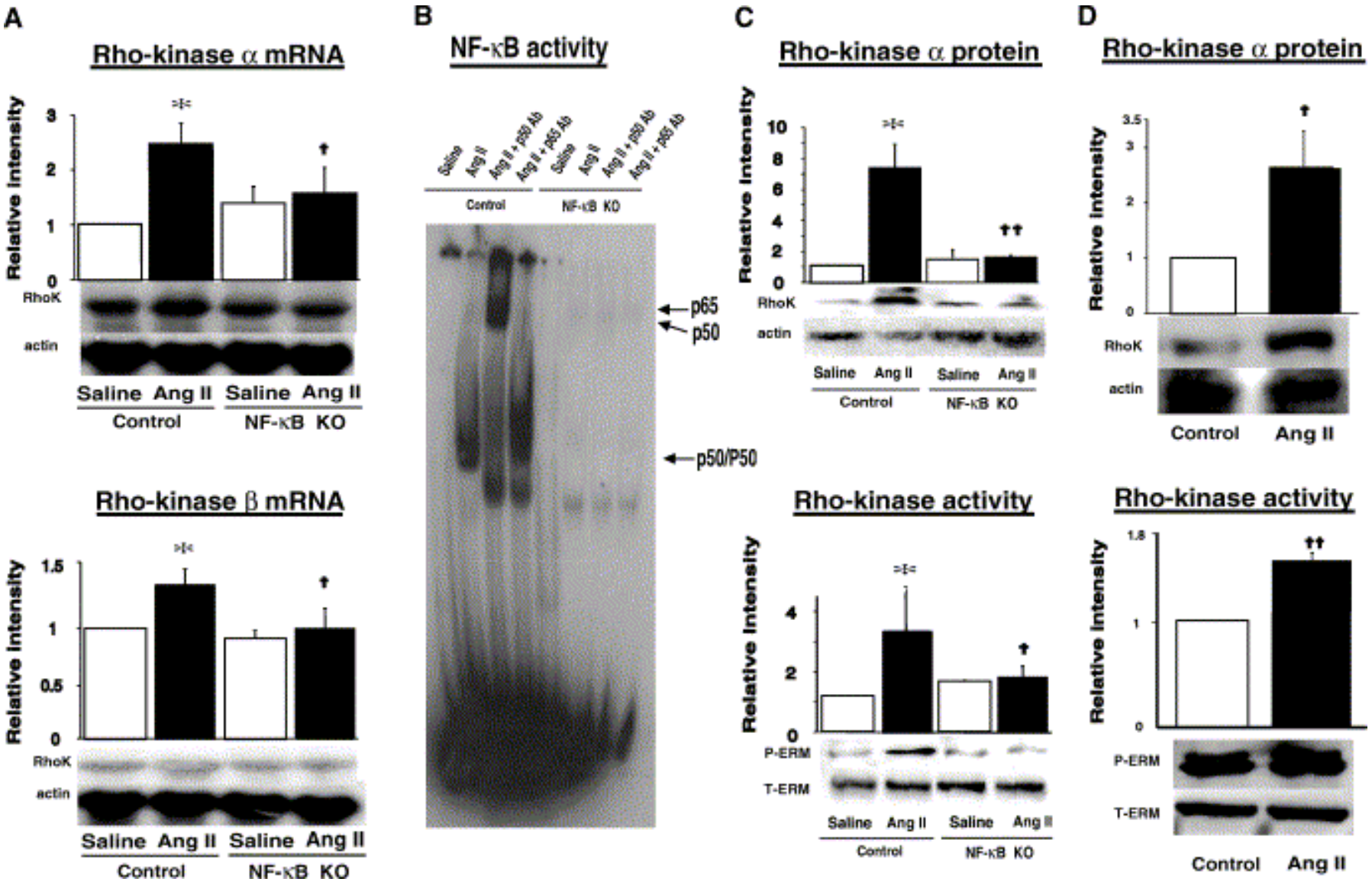
Abstract

Recent studies have demonstrated that upregulated Rho-kinase plays an important role in the pathogenesis of arteriosclerosis and vasospasm in both animals and humans. However, little is known about the molecular mechanism(s) involved in the Rho-kinase upregulation. Since inflammatory mechanisms have been implicated in the pathogenesis of arteriosclerosis and vasospasm, we examined whether inflammatory stimuli upregulate Rho-kinase in vitro and in vivo. In cultured human coronary vascular smooth muscle cells (hcVSMC), inflammatory stimuli, such as angiotensin II and interleukin-1β, increased Rho-kinase expression (at both mRNA and protein levels) and function (as evaluated by the extent of the phosphorylation of the ERM (the ezrin/radixin/moesin) family, substrates of Rho-kinase) in a time- and concentration-dependent manner. The expression of Rho-kinase was inhibited by blockades of protein kinase C (PKC) (by either GF109253 or prolonged treatment with phorbol myristate acetate for 24 h) and an adenovirus-mediated gene transfer of dominant-active Iκ-B, suggesting an involvement of PKC and NF-κB in the intracellular signal transduction pathway for the Rho-kinase expression. Furthermore, coronary vascular lesion formation (characterized by medial thickening and perivascular fibrosis) induced by a long-term administration of angiotensin II was markedly suppressed in NF-κB$^{-/-}$ mice with reduced expression and activity of Rho-kinase in vivo. These results indicate that the expression and function of Rho-kinase are upregulated by inflammatory stimuli (e.g. angiotensin II and IL-1β) in hcVSMC with an involvement of PKC and NF-κB both in vitro and in vivo.

Ang II induce expression and activation of ROCK via NF-κB *in vivo*



Shimokawa H, J Mol Cell Cardiol 2004;37:537

Cell, Vol. 119, 343–354, October 29, 2004, Copyright ©2004 by Cell Press

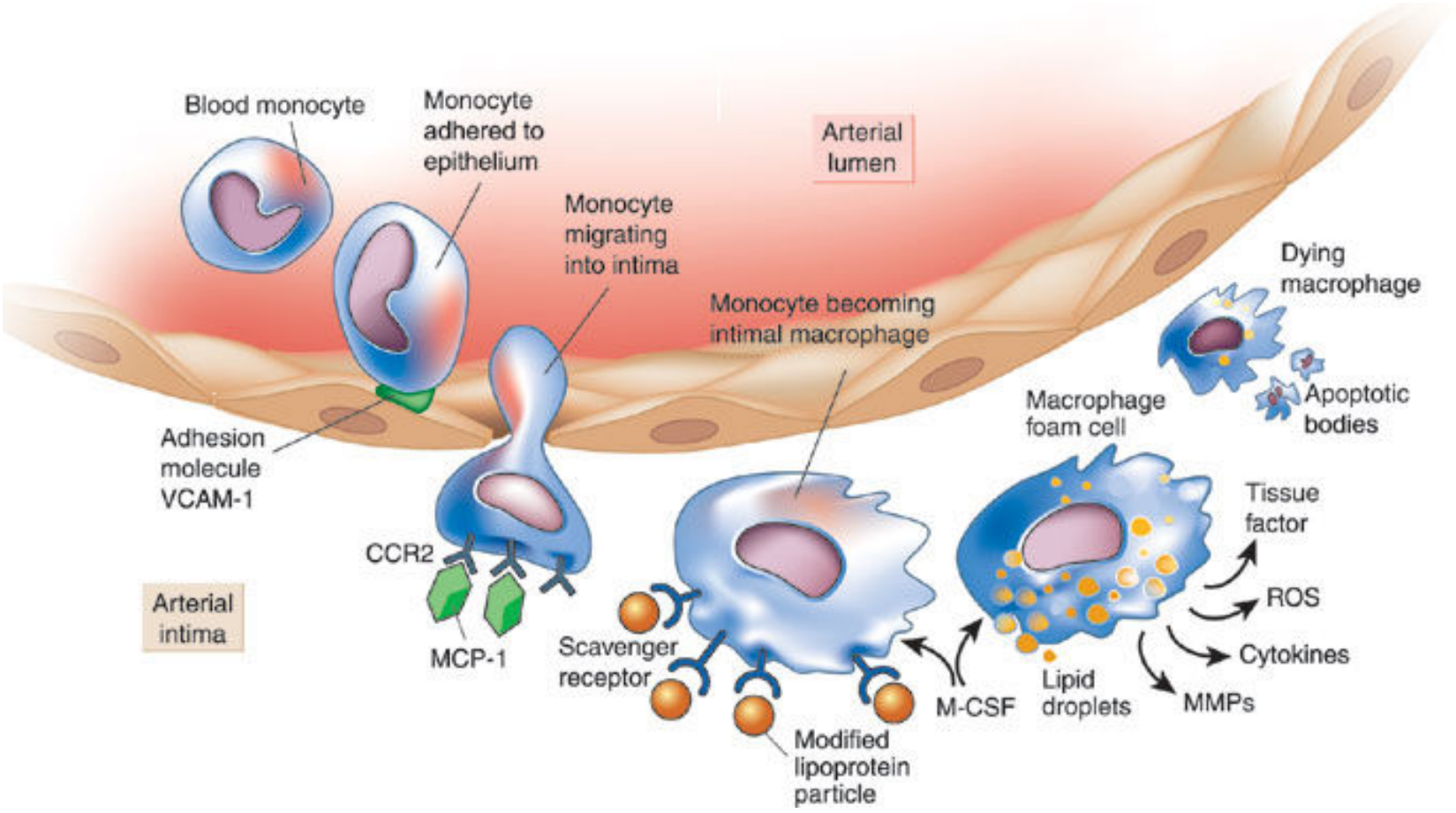
Factor XIIIA Transglutaminase Crosslinks AT_1 Receptor Dimers of Monocytes at the Onset of Atherosclerosis

Summary

Many G protein-coupled receptors form dimers in cells. However, underlying mechanisms are barely understood. We report here that intracellular factor XIIIA transglutaminase crosslinks agonist-induced AT_1 receptor homodimers via glutamine^{315} in the carboxyl-terminal tail of the AT_1 receptor. The crosslinked dimers displayed enhanced signaling and desensitization in vitro and in vivo. Inhibition of angiotensin II release or of factor XIIIA activity prevented formation of crosslinked AT_1 receptor dimers. In agreement with this finding, factor XIIIA-deficient individuals lacked crosslinked AT_1 dimers. Elevated levels of crosslinked AT_1 dimers were present on monocytes of patients with the common atherogenic risk factor hypertension and correlated with an enhanced angiotensin II-dependent monocyte adhesion to endothelial cells. Elevated levels of crosslinked AT_1 receptor dimers on monocytes could sustain the process of atherogenesis, because inhibition of angiotensin II generation or of intracellular factor XIIIA activity suppressed the appearance of crosslinked AT_1 receptors and symptoms of atherosclerosis in ApoE-deficient mice.

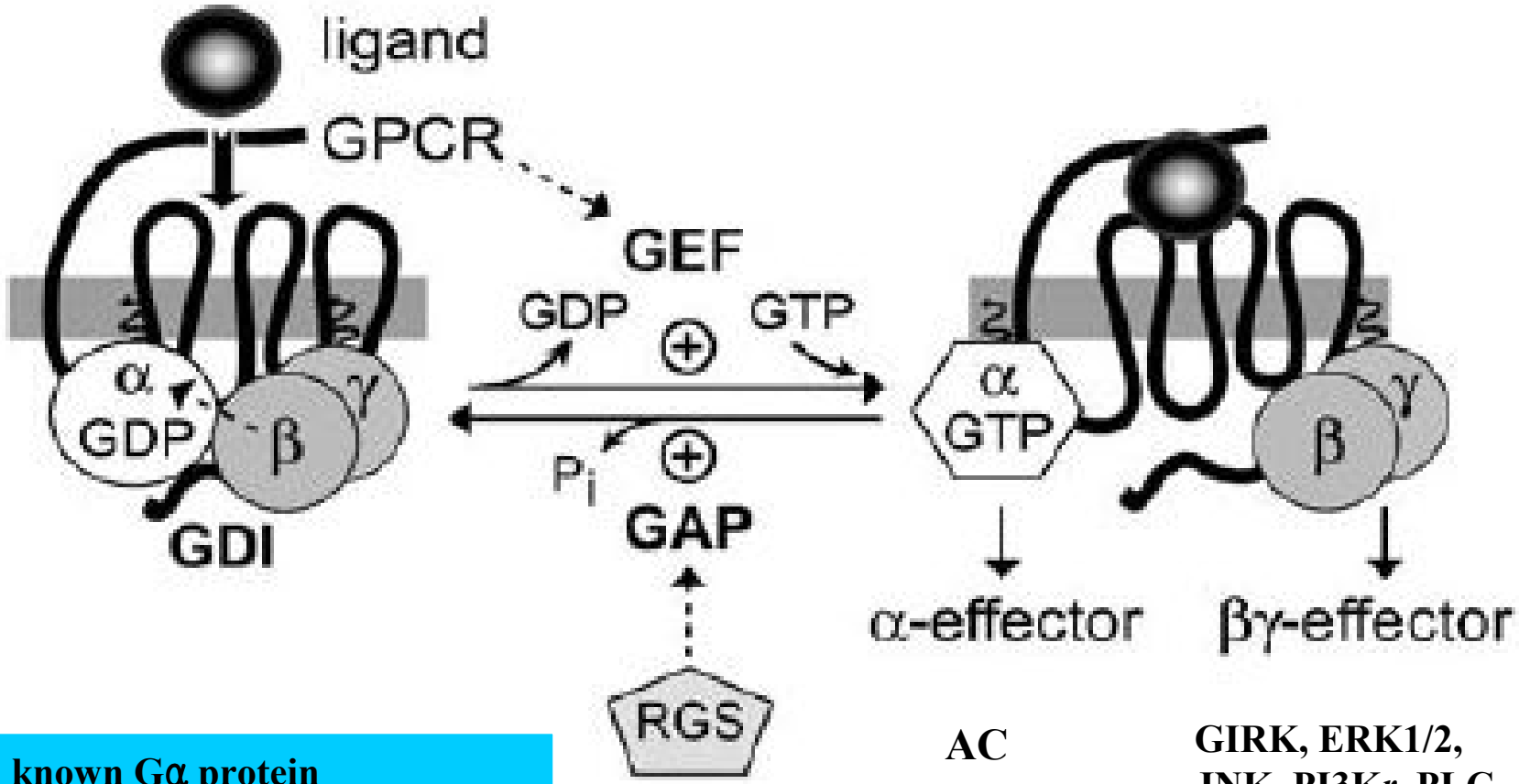
AbdAlla S et al. Cell 2004;119:343-54

Mononuclear phagocytes in atherogenesis



Libby P. Nature 2002;420:868

G-Protein coupled Receptor (G-PCR) Signaling Model



23 known Gα protein
Gα(s/olf)
Gα(i1/i2/i3/t-rod/t-con/gust/z)
Gα(q/11/14/16)
Gα(12/13)

AC

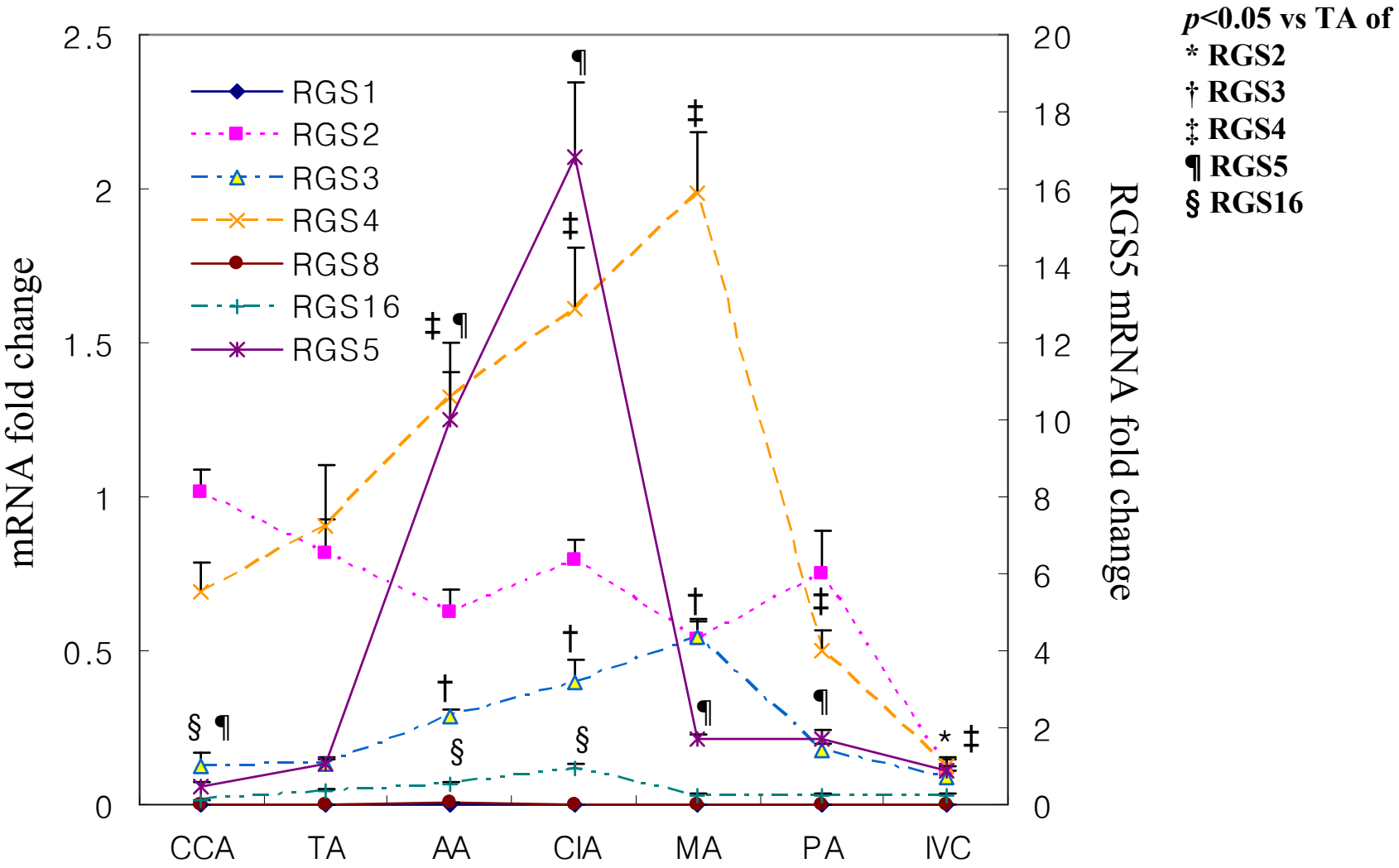
GIRK, ERK1/2, JNK, PI3K*r*, PLC-β/-ε
MAPKs etc.

McCudden CR. CMLS 2005;62:551

GPCR signaling & Cariovascular disease

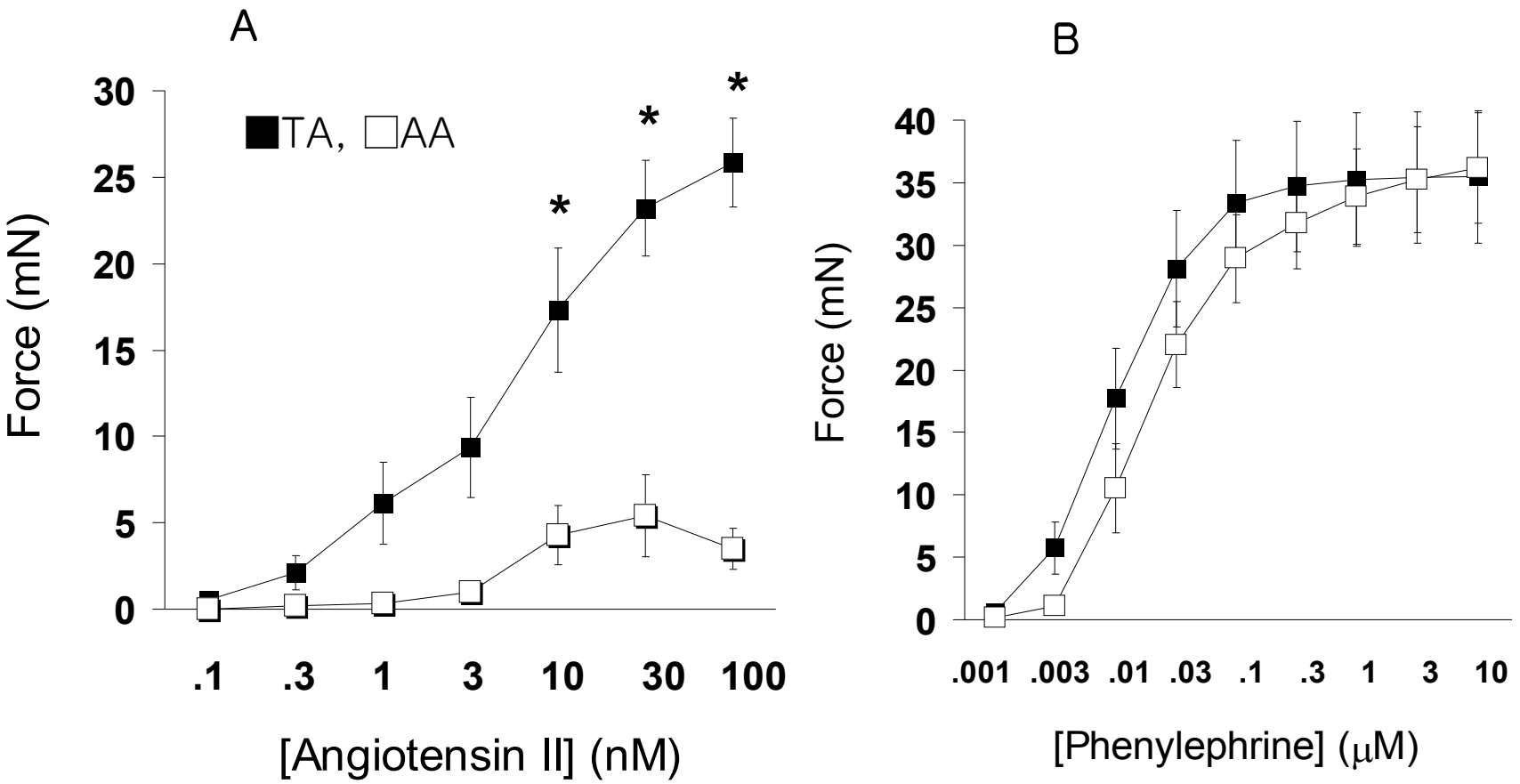
- **$G_{\alpha q}$ transgenic overexpression mediates changes in heart function leading to hypertrophy** (D'Angello D. PNAS197;94:8121).
- **RGS2+/- and RGS2-/- are hypertensive and show prolonged response of the vasculature to vasoconstrictor *in vivo*.** (Heximer SP, J Clin Invest 2003)
- **Transgenic overexpression of RGS4 in aortic coarctation mice model reduced ventricular hypertrophy and rapid decompensation** (Rogers JH. J Clin Invest 1999)

Differential expression of RGS proteins in each vascular segments

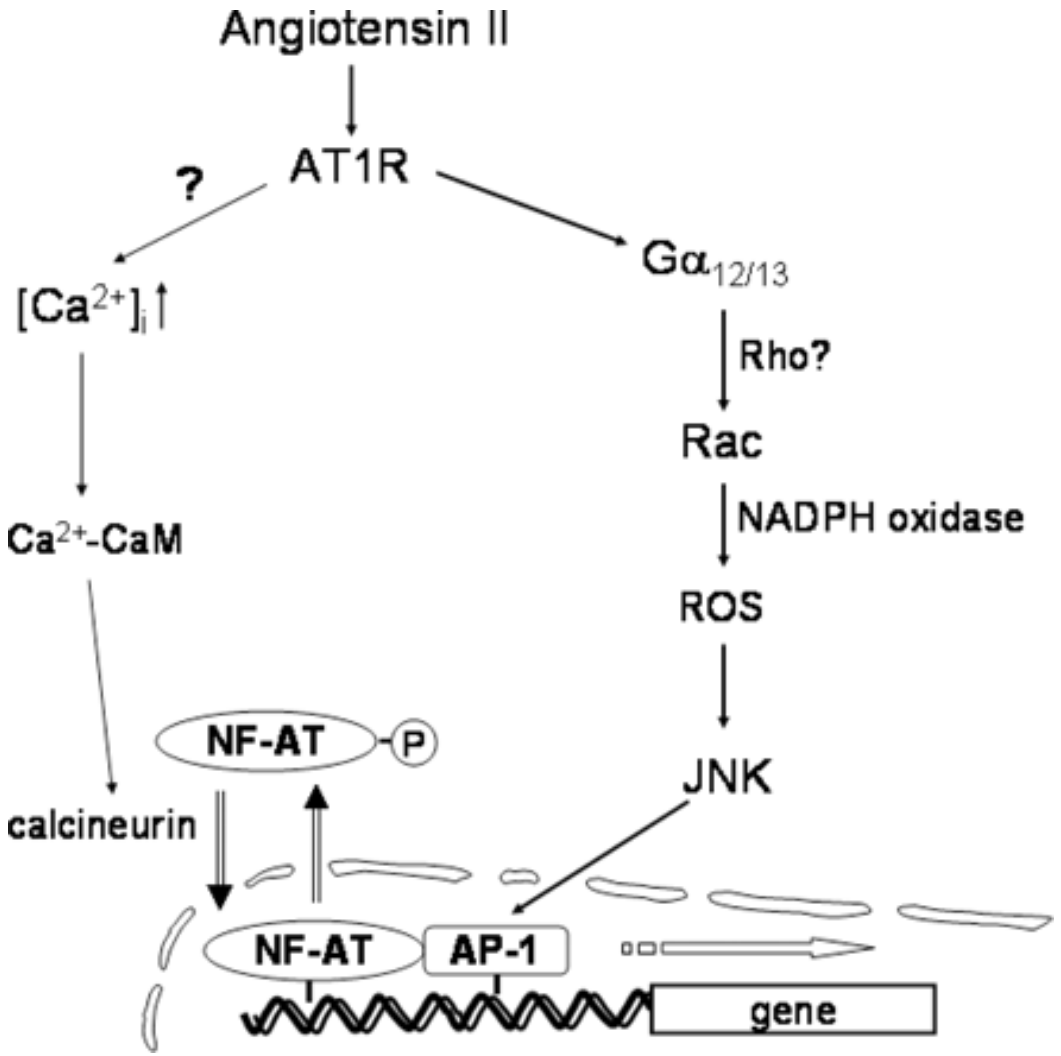


Chung I-M & Schwartz SM. AHA 2006

Decreased arterial contraction in abdominal vs thoracic aorta

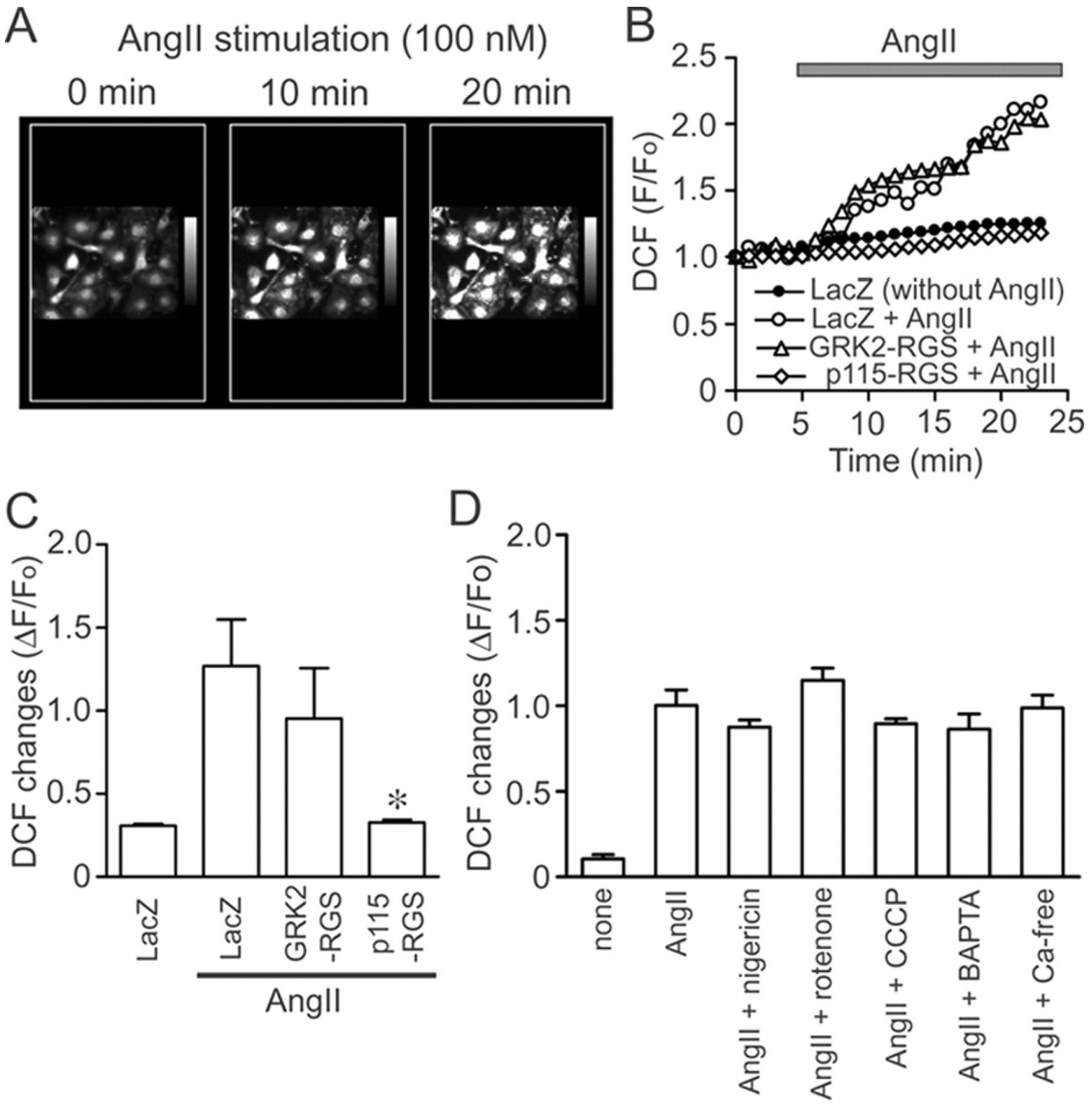


AT1R-stimulated NFAT Activation in Cardiac Fibroblast

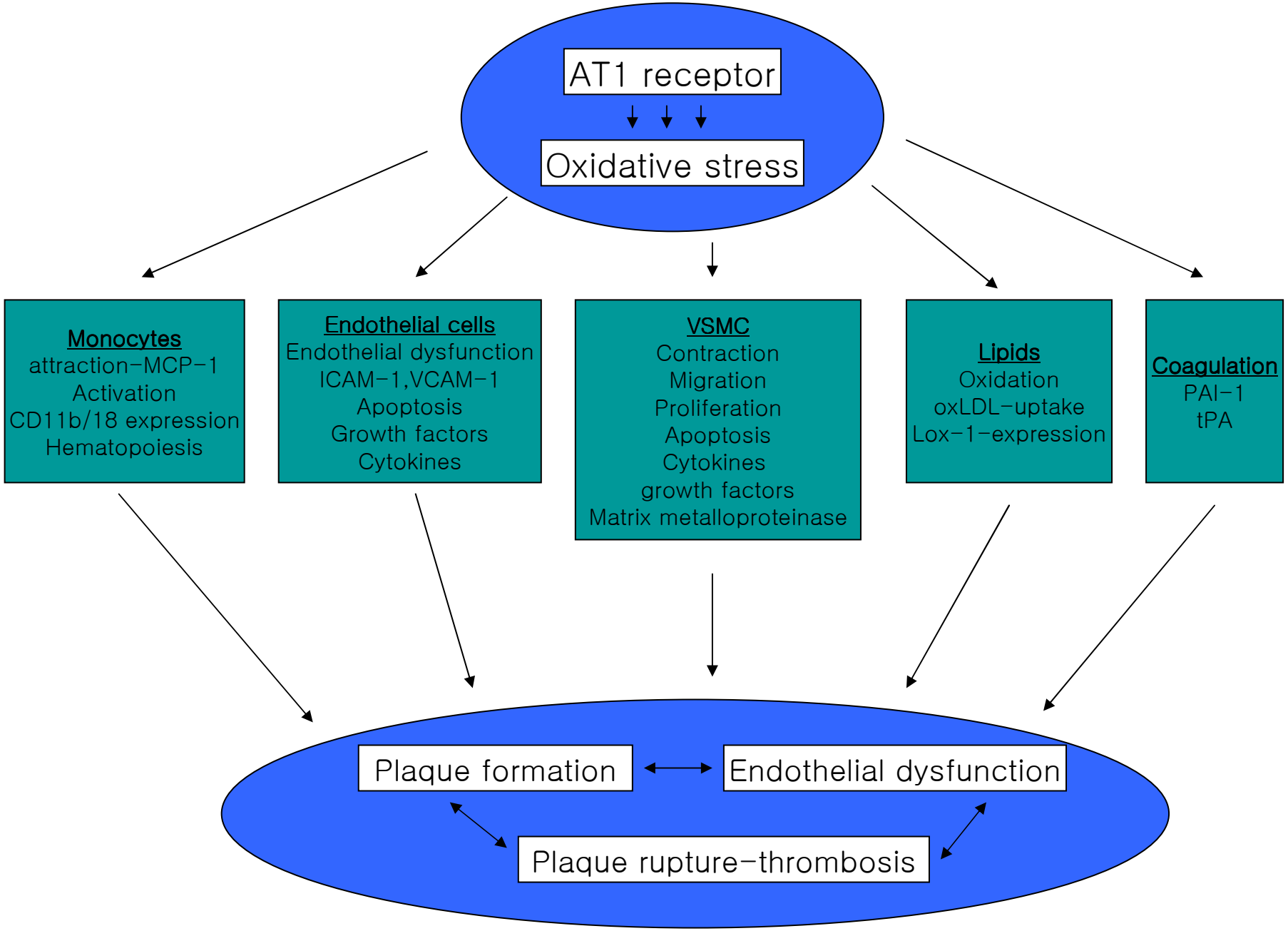


Kurose H. J Biol Chem. 2005;280:23041-7

G12/13 mediate Ang II-induced ROS production



Fujii, T. et al. J. Biol. Chem. 2005;280:23041-23047


AT1 receptor
Oxidative stress
Monocytes
attraction-MCP-1
Activation
CD11b/18 expression
Hematopoiesis
Endothelial cells
Endothelial dysfunction
ICAM-1,VCAM-1
Apoptosis
Growth factors
Cytokines
VSMC
Contraction
Migration
Proliferation
Apoptosis
Cytokines
growth factors
Matrix metalloproteinase
Lipids
Oxidation
oxLDL-uptake
Lox-1-expression
Coagulation
PAI-1
tPA
Plaque formation
Endothelial dysfunction
Plaque rupture-thrombosis

Psychological Stress-mediated CV disease