

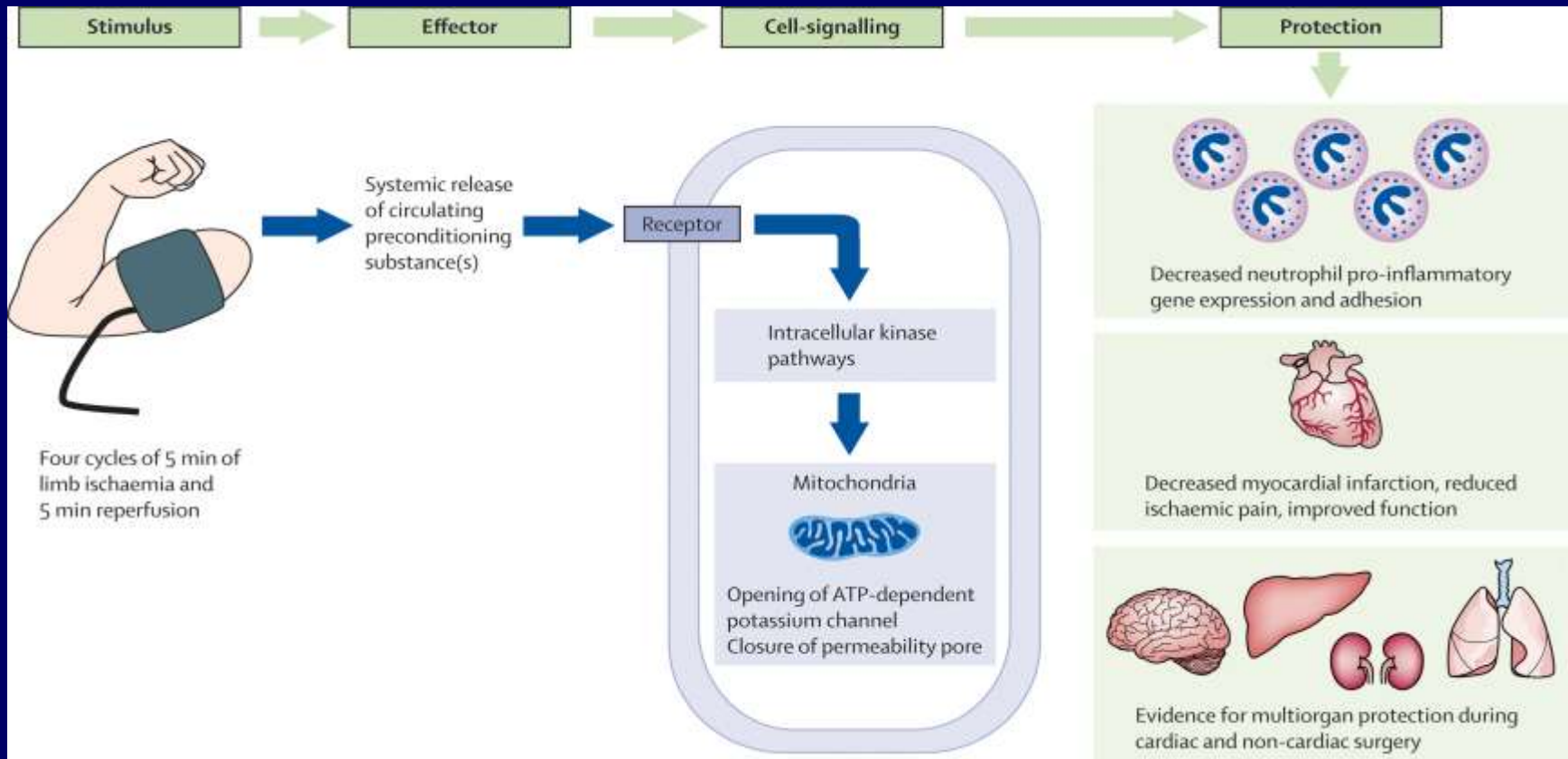
Remote Ischemic Preconditioning:

Current aspects of mechanisms

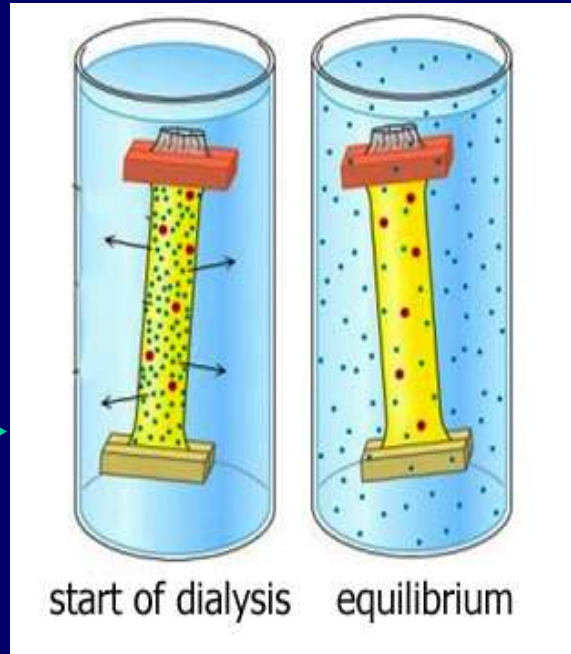
Hans Erik Bøtker,
Aarhus University Hospital, Skejby, Denmark,



Mechanisms

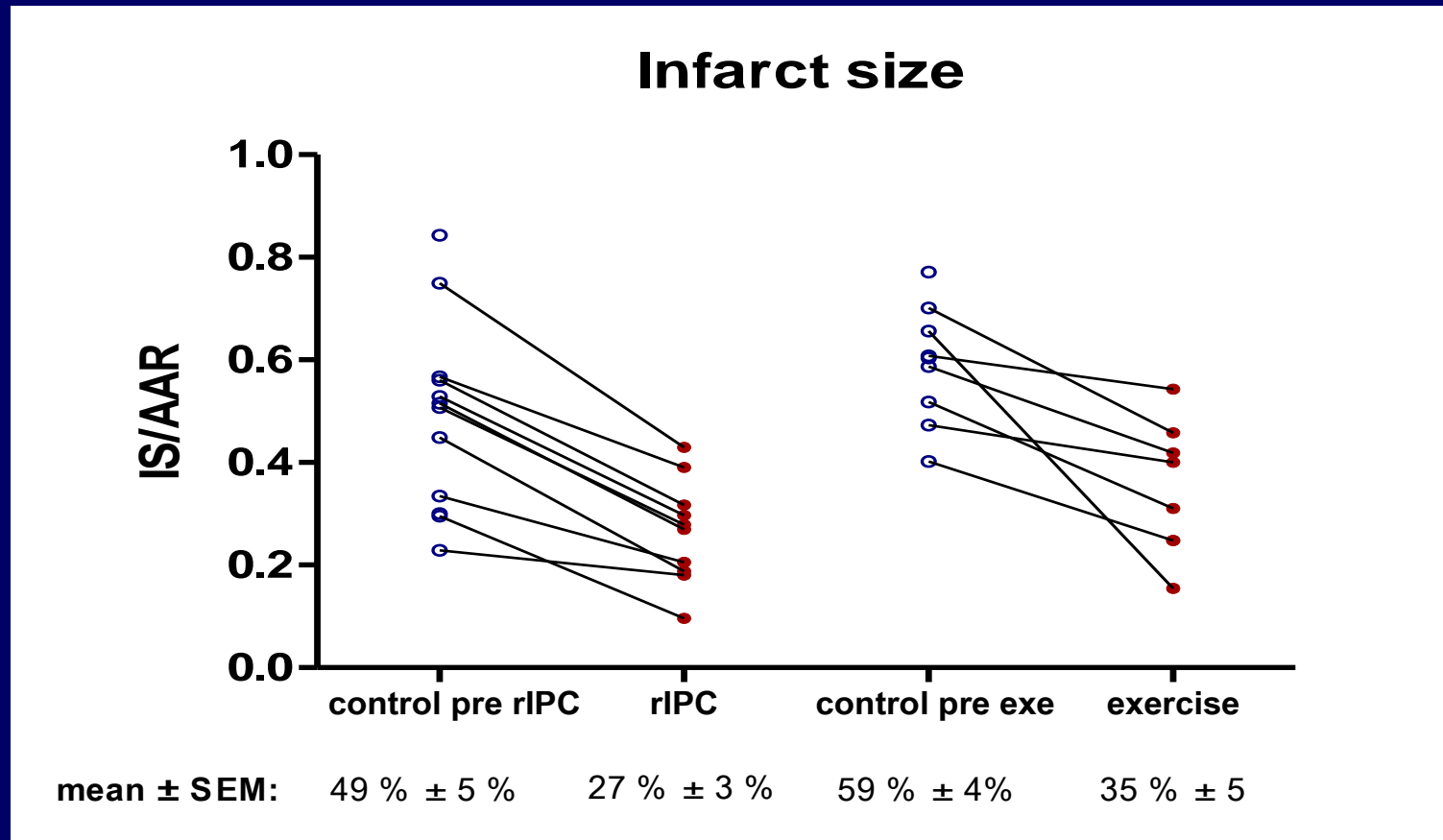


Dialysate as a bioassay for rPerC

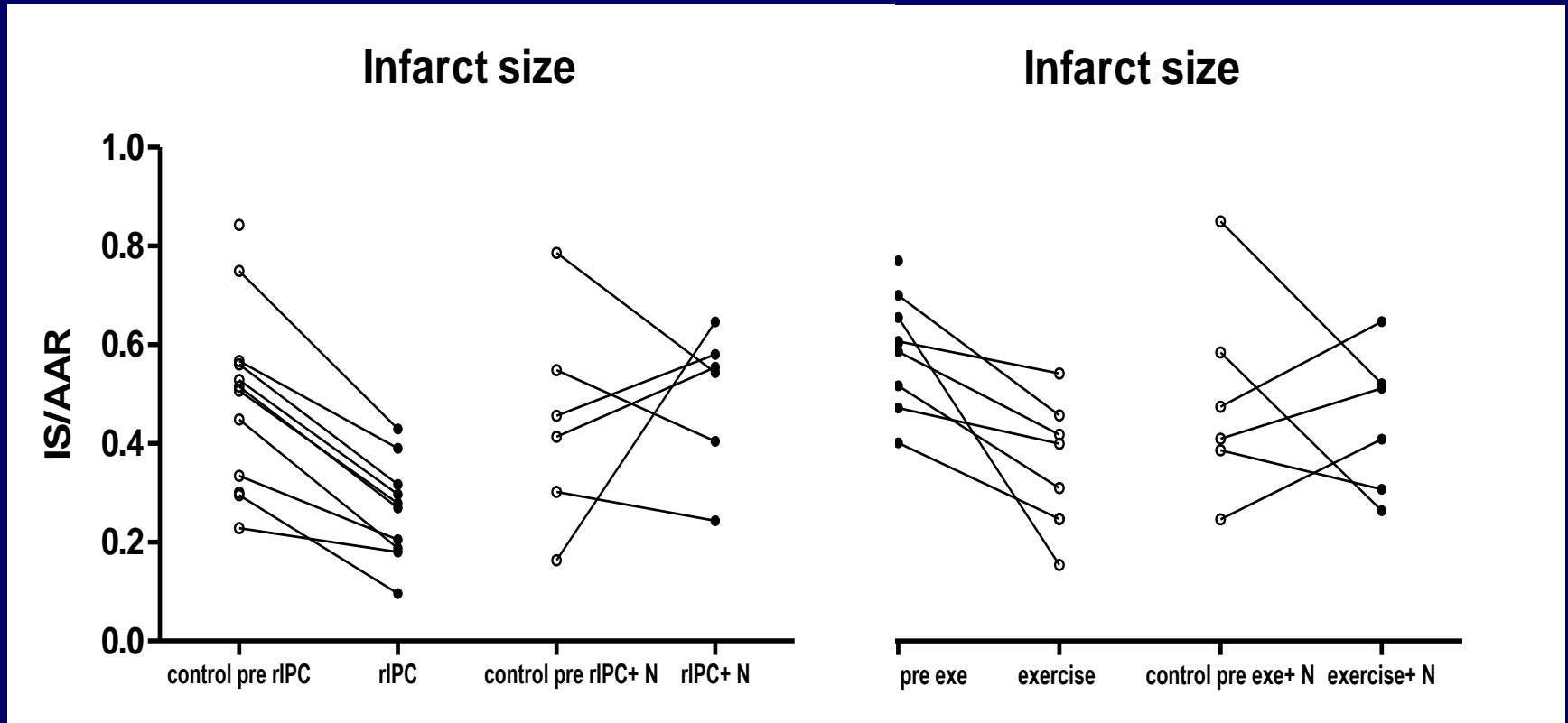


Shimizu et al. Clin Sci 2009;117:191-200
Michelsen et al. 2011 submitted

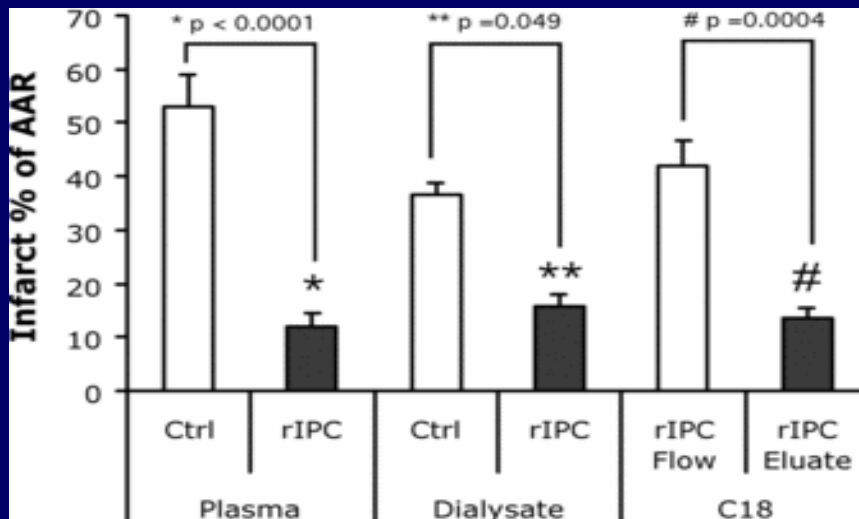
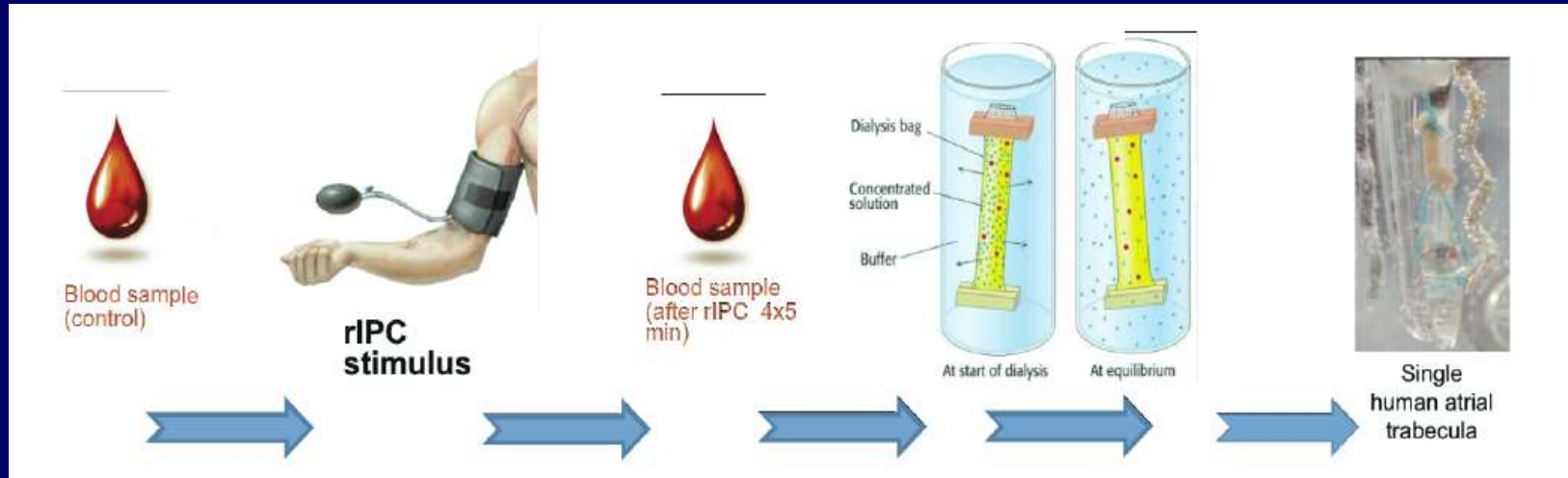
Cardioprotection with exercise and rIPC



Factor X: endogenous opioids?



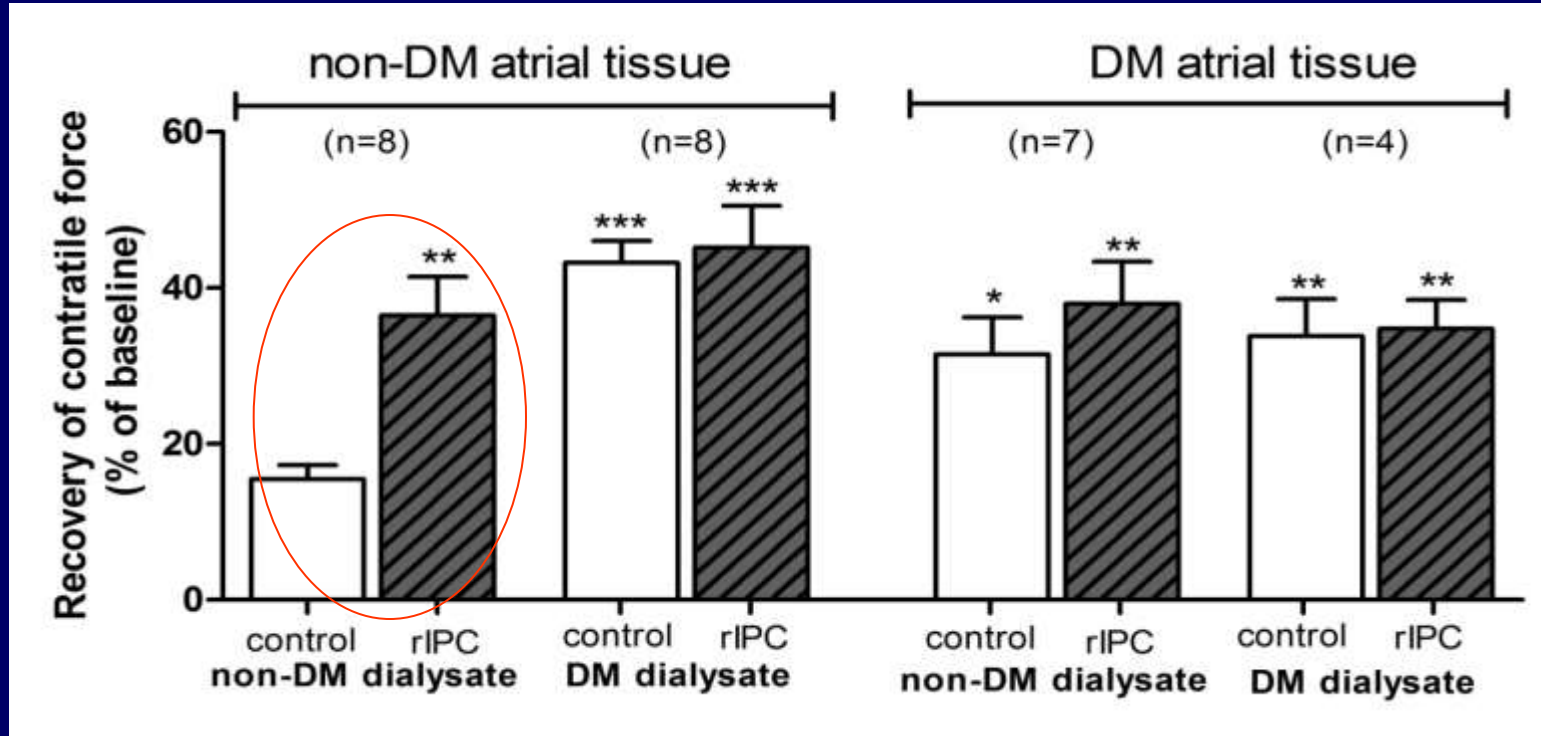
Dialysate as a bioassay for rIPerC



Cardioprotective circulating factor(s) can be dialysed from human plasma and transferred to atrial trabeculae

Human diabetic type 2 patients – heart tissue (atrial strips) Remote conditioning

Atrial tissue exposed to **simulated ischemia** (no glucose, no oxygen)



Diabetic plasma dialysate: protective humoral mediator(s)
Diabetic heart tissue: protective mechanisms present

The Malate-aspartate shuttle (MAS)

Discovery: P. Borst 1962 (**Biochem Biophys Acta**)

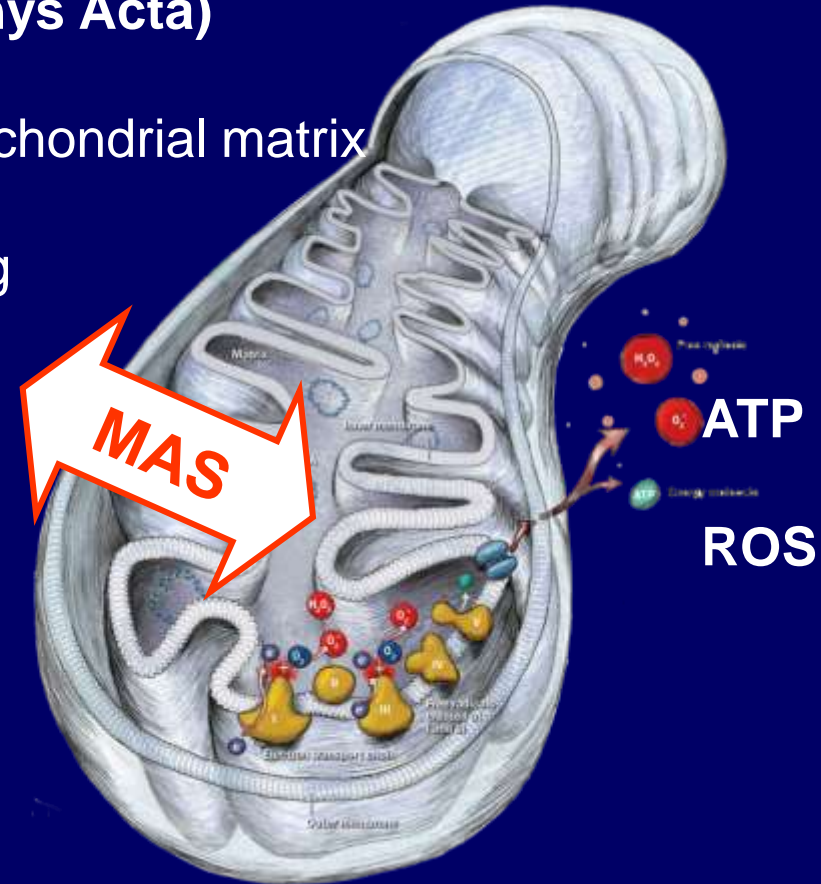
Communication between cytosol and mitochondrial matrix

No known relation to ischemic conditioning

No technology for dynamic MAS flux

Glucose metabolism (ischemia)

Opening of mitochondrial permeability transition "pore" (mPTP) includes NAD/NADH (Di Lisa J Biol Chem 2001)



Hypothesis:

Ischemic damage of mPTP includes disturbed MAS flux

Any link between signaling pathways of cardioprotection and myocardial energy metabolism ?

Proteomic and metabolomic analysis of cardioprotection: Interplay between protein kinase C epsilon and delta in regulating glucose metabolism of murine hearts

Manuel Mayr^{a,*}, David Liem^d, Jun Zhang^d, Xiaohai Li^d, Nuraly K. Avliyakov^d, Jeong In Yang^d, Glen Young^d, Tom M. Vondriska^d, Christophe Ladroue^c, Basetti Madhu^b, John R. Griffiths^b, Aldrin Gomes^d, Qingbo Xu^a, Peipei Ping^d

^a Cardiovascular Division, BHF Centre, King's College, London, 125 Coldharbour Lane, London SE5 9NU, UK

^b Cancer Research UK Cambridge Research Institute, Cambridge, UK

^c University of Warwick, UK

^d Department of Physiology, David Geffen School of Medicine, UCLA, Los Angeles, US

J Mol Cell Cardiol 2009;46:268

*“Among differentially expressed proteins were cytosolic enzymes of the **malate-aspartate shuttle**.....these enzymatic changes were dependent on **PKC ε,δ***”

“..first evidence that PKCε,δ activity modulates cardiac glucose metabolism..”

MAS – myocardial ischemia

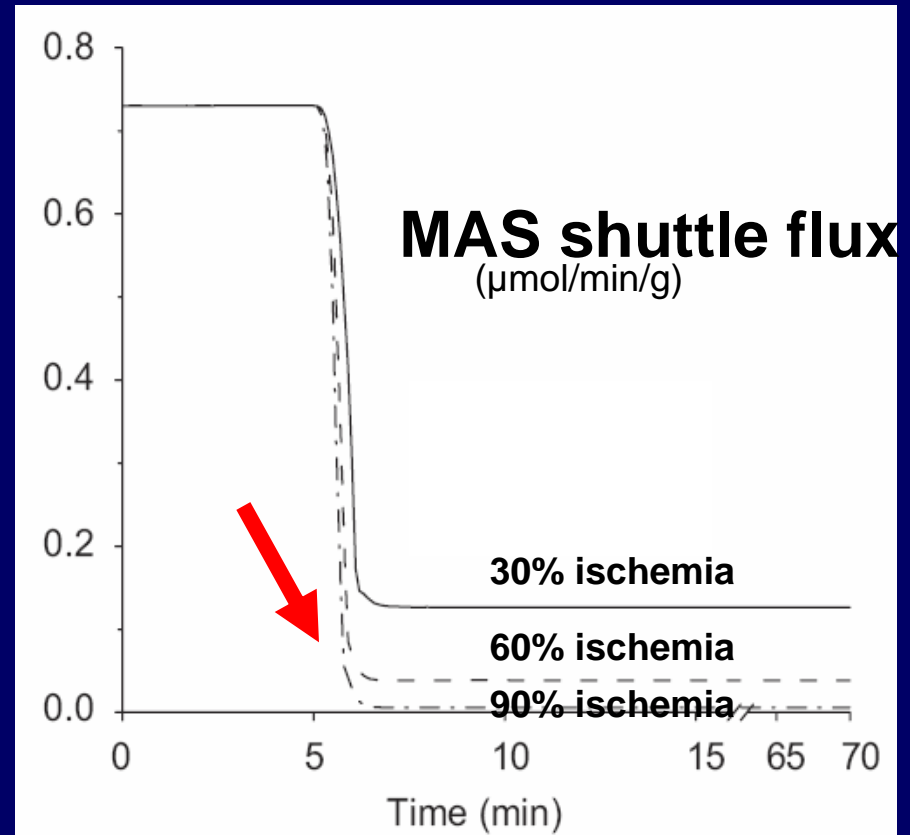
In silico studies: computerized flux estimation

MAS flux - transfer of NADH decreases within seconds during myocardial ischemia

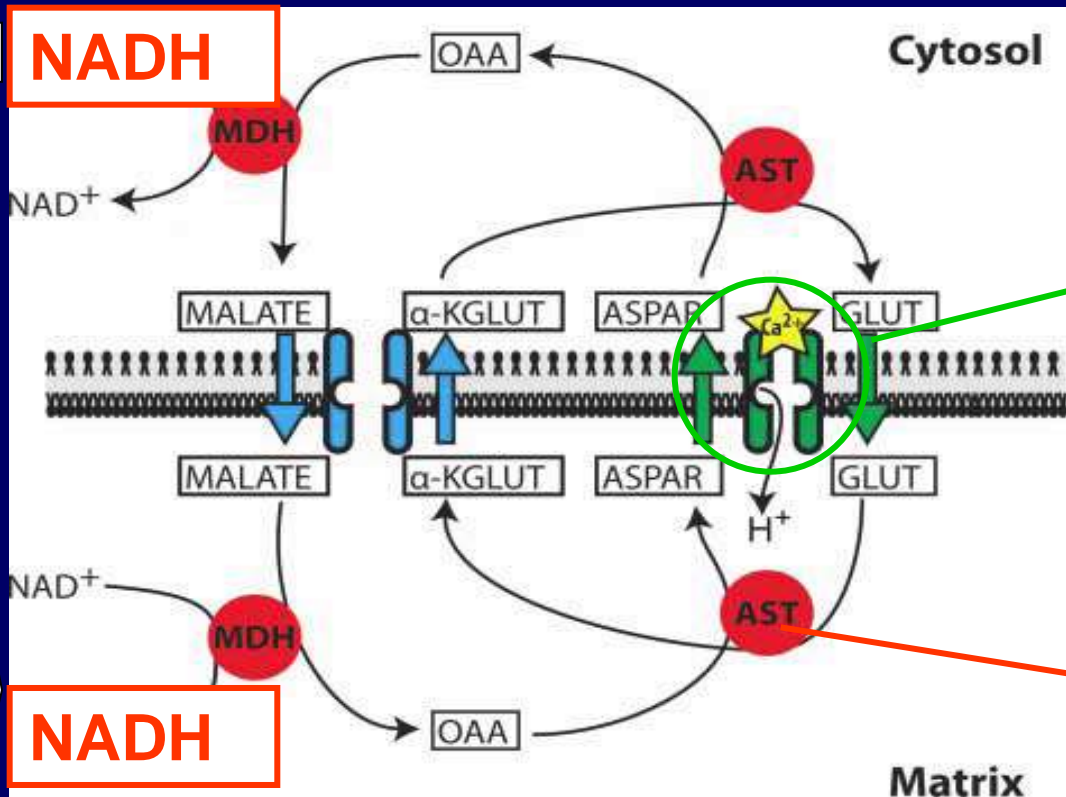
Lu et al. J Theoretical Biol 2008;254

Ischemic conditioning:

Repetitive occlusion of MAS during brief ischemic periods ?



The Malate-Aspartate shuttle (NADH transfer)



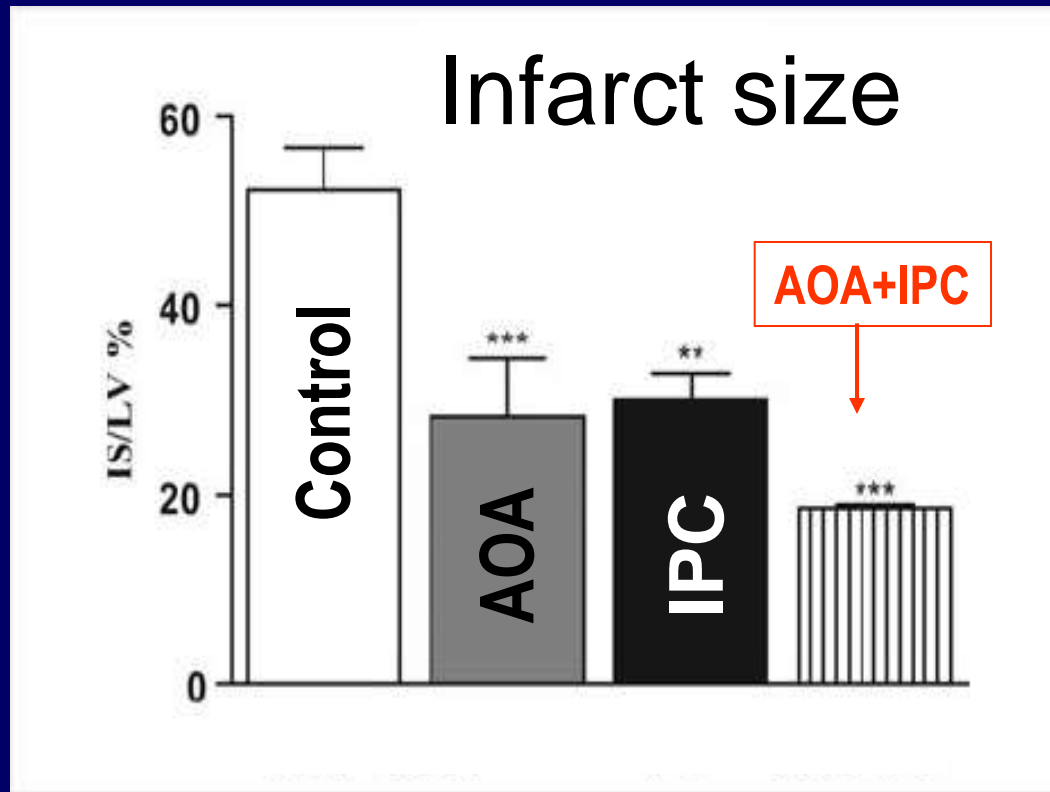
AGC- Carrier:
Proteins: Aralar,
Citrin, AAAT1
Glutamate uptake
Ca²⁺ uptake
H⁺ exchange

**Aspartate
amino transferase**

**MAS is blocked by Aminooxyacetate (AOA),
- chemical inhibitor of AST reaction**

Preischemic loading of the heart with AOA (0.1 mM) during ischemic and start of reperfusion

Isolated perfused rat hearts (Langendorff)
40 min global no flow ischemia – 120 min reperfusion

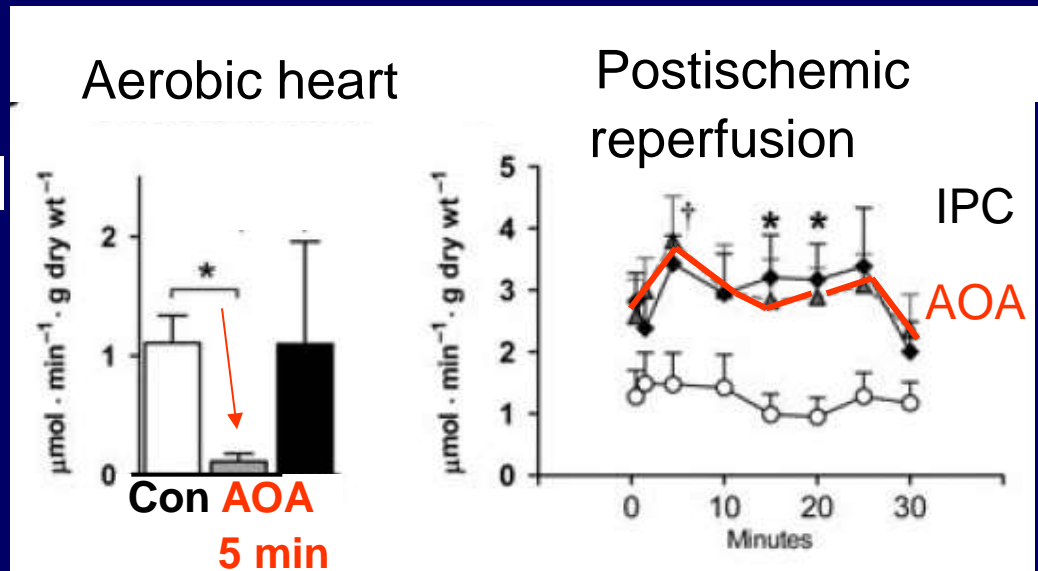


Additive protection of IPC and inhibition of MAS flux

AOA inhibition of MAS

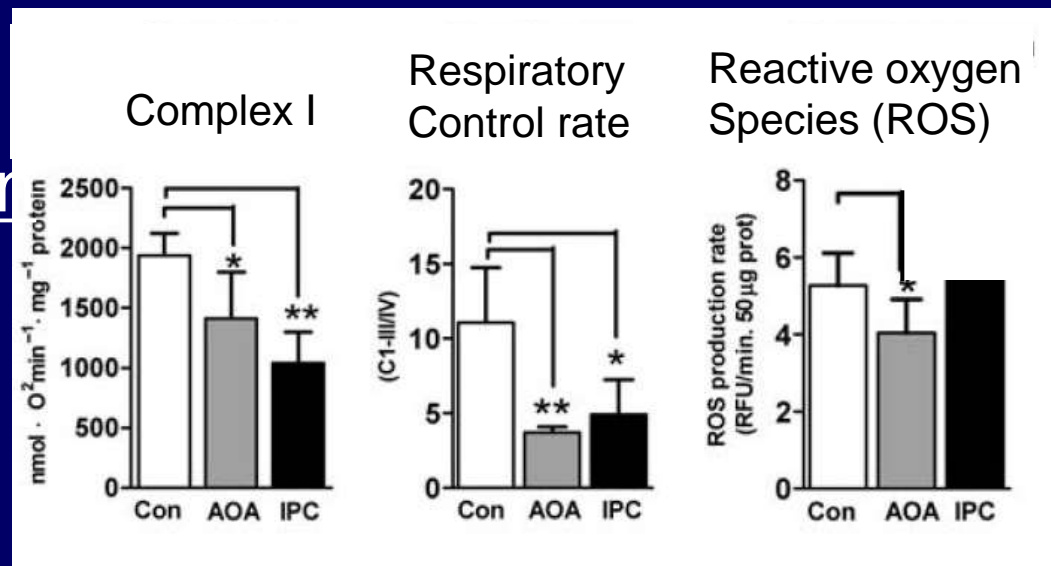
Effects on myocardial Glucose oxidation

Isolated perfused rat heart



Mitochondrial Oxygen consumption

Isolated rabbit mitochondria
Aerobic heart



Transient inhibition of the Malate-Aspartate shuttle is a potential mechanism of protection against ischemia-reperfusion injury (like IPC)

Reversible transient inhibition of myocardial glucose oxidation may be a key phenomenon of ischemic conditioning.

A tool for testing protective effect in experimental studies ?

Animal model (rats):

Remote ischemic preconditioning in evolving stroke

Hahn CD et al. Stroke 2011;42:2960-2

Focal cerebral ischemia:

Transient middle cerebral artery
occlusion 120 min

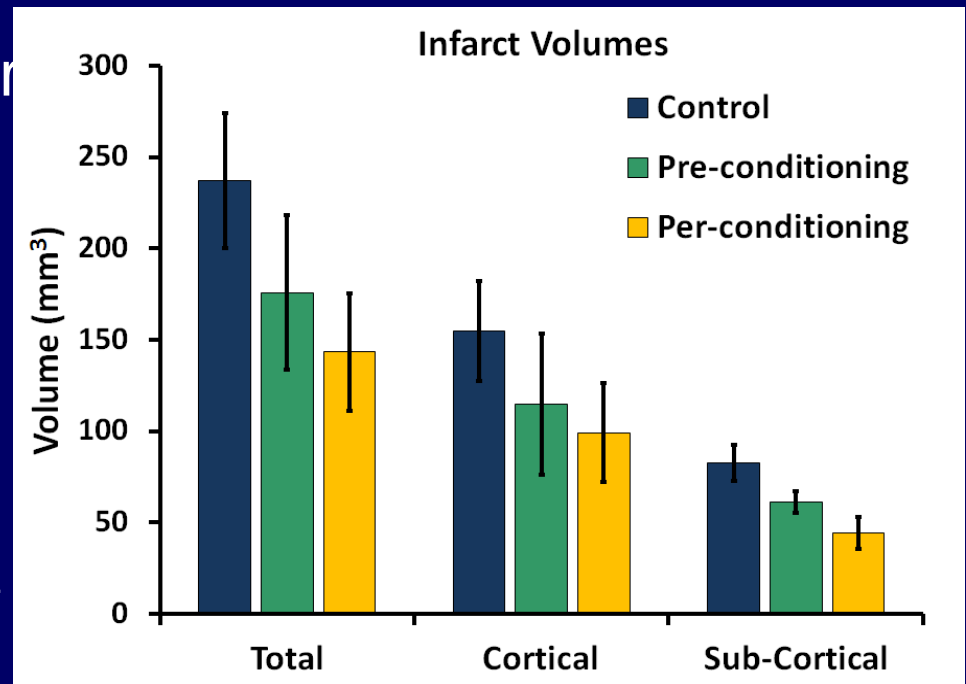
Reperfusion: 24 hours

Pre-conditioning:

40 min before ischemia

Remote Per-conditioning:

40 min before reperfusion



Protection by both IPC and rIPerC ($p < 0.001$) – rIPerC > local IPC

Remote ischemic preconditioning in thrombolysed patients with evolving stroke

Clinical Trials gov: NCT 00975962
Inclusion completed May 2011

Andersen G, Østergaard L, Department of Neurology,
Aarhus University Hospital , NBG, Denmark

Results available within approx two months

Clinical stroke study - rPerC

120 patients:

Randomized to **thrombolysis + rPerC** vs **thrombolysis alone**

Prehospital rPerC in the ambulance - (as in myocardial infarct study)

Inclusion criteria:

Suspicion of stroke (NIHSS 4-24), paresis of an extremity

Thrombolysis < 4.5 hrs from symptom onset

MRI: DWI lesion consistent with stroke

Advanced MR imaging : admission, - 24 hours and 1 month

Primary endpoint:

Salvage index %, difference in infarct growth (PWI-DWI)

Secondary endpoint:

Final infarct size, T2 FLAIR after 1 month

Case – advanced MRI technology

Young girl – embolism of the right middle cerebral artery (MR angio) due to persistent oval foramen – severe paresis of left sided extremities

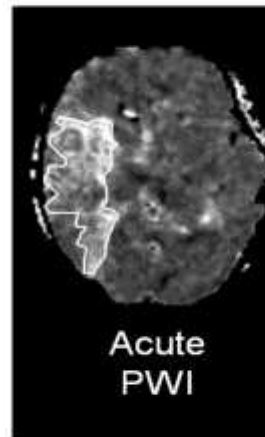
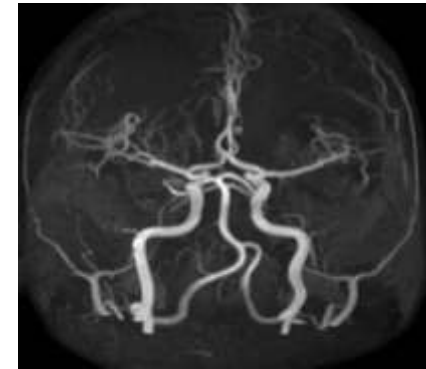
Treated with thrombolysis

DWI: diffusion weighted imaging:
"necrotic area at admission"

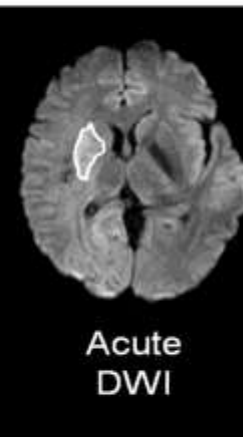
PWI: perfusion weighted imaging
"area at risk at admission"

Advantage: Calculation of correct amount of salvageable tissue or "**true area at risk**" at admission

Complete clinical remission



Acute
PWI



Acute
DWI



1 month
T2-FLAIR

Admission

Final infarct
Size

Thank you!

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