Mechanism of Late Preconditioning

ISCHEMIC STRESS

TRIGGER OF LATE PC

SIGNAL TRANSDUCTION CASCADE

TRANSCRIPTION FACTORS

GENE UPREGULATION

MEDIATOR OF LATE PC

PROTECTION

- NO (eNOS)
- ROS
- Adenosine
- PKC (ε isoform)
- Tyrosine kinases (Src/Lck)
- JAK1/2
- NF-κB
- STAT1/3
- Nrf2
- iNOS
- COX-2
- HO-1
- Antioxidant enzymes (aldose reductase, Mn SOD, and ecSOD)
- HSPs?

K_ATP Channels

Day 1

Days 2-4
iNOS is an obligatory mediator of late preconditioning induced by diverse stimuli, including ischemia, physical exercise, NO donors, adenosine A₁ agonists, opioid delta₁ agonists, alpha₁ adrenergic agonists, and endotoxin derivatives, indicating that this enzyme plays a central role in cardioprotection.

However, it is unknown whether an increase in iNOS activity via gene transfer can replicate the salubrious effects of late PC.
Ischemic region

Region of transfection

Coronary occluder
iNOS expression in myocardium 8 weeks after Av3-mediated iNOS gene transfer

Av3/LacZ  Av3/iNOS

A  B

x400  x400
iNOS expression and activity 1 year after rAAV-mediated *iNOS* gene transfer in mice

![Graph showing iNOS protein and activity](Image)

- **iNOS protein** (% of LacZ group)
  - rAAV/LacZ
  - rAAV/iNOS
  - *P<0.05 vs. LacZ*

- **iNOS activity** (pmol L-citrulline/min/mg protein)
  - LacZ
  - iNOS
  - *P<0.05 vs. LacZ*
Myocardial infarction 1 year after rAAV-mediated LacZ gene transfer
Myocardial infarction 1 year after rAAV-mediated iNOS gene transfer
Infarct size is reduced up to 1 year after Av3- or rAVV-mediated iNOS gene transfer
• *iNOS* gene transfer affords effective cardioprotection. The magnitude is equivalent to that afforded by late preconditioning but the protection is essentially permanent (at least 1 year).

• The results provide proof of principle for gene therapy against ischemia/reperfusion injury which increases local myocardial NO levels without hemodynamic effects, thereby obviating the need for continuous intravenous infusion of NO donors.