

P014 Mitochondrial Permeability Transition Inhibition by
Preconditioning Does Not Involve Protein Kinase C ϵ
Translocation

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Inhibition of the mitochondrial permeability transition pore by ischemic preconditioning can protect hearts from reperfusion injury. However, inhibition of pore opening in these hearts is secondary to a decrease in oxidative stress or calcium overload. Other workers have reported a protection of mitochondrial function by translocation of protein kinase C epsilon (PKC ϵ) to the mitochondria following preconditioning. We carried out careful sub-fractionation of preconditioned hearts with the measurement by Western blotting of PKC ϵ and PKC α in the cytosol, a crude particulate fraction and mitochondria freed of plasma membrane contamination. Sample loading and purity of each fraction was confirmed using Western blotting for glyceraldehyde-3-P dehydrogenase (cytosol), the adenine nucleotide translocase (mitochondria) and mono-carboxylate transporter 1 (plasma membrane). We found no evidence for any PKC translocation following preconditioning with transient ischemia, diazoxide or urocortin, and most, if not all of the PKC found in the mitochondrial fraction could be accounted for by plasma membrane contamination. In contrast, phorbol ester treatment gave a substantial translocation of PKC to the particulate fraction of which a small proportion was associated with mitochondria. Thus, PKC translocation to mitochondria is not involved in the mechanism of MPTP inhibition by preconditioning.