Pretreatment with a Novel Tri-Peptide Elicits Cardioprotective Effects in Myocardial Ischemia/Reperfusion Injury

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INTRODUCTION

Reperfusion of coronary blood flow to the ischaemic heart following an acute myocardial infarction (MI), although necessary, may lead to myocardial ischemia/reperfusion (IR) injury, resulting in cardiomyocyte death and compromised cardiac function [1]. The major cause of IR injury is due to reactive oxidative species (ROS), which damage the mitochondria that comprise up to one-third of the heart volume and is one of the key producers of ROS [2] (figure 1). The generation ROS leads to the loss of mitochondrial membrane potential and opening of the mitochondrial permeability transition pore (MPTP), leading to cardiac contractile dysfunction and increased infarct size [2]. There are currently no pharmacologic tools that have been shown to clinically improve cardiac function and reduce infarct size in patients who have suffered from reperfusion-induced MI injury. Due to the role of mitochondria in IR injury, treatment modalities targeting the mitochondria are a growing field in cardiovascular research.

METHODS

Male Sprague-Dawley rats (275-325 g, Charles River, Springfield MA) were anesthetized i.p. with sodium pentobarbital (60 mg/kg) and anesthetized with heparin 1000 units. Hearts were isolated and studied using a modified Langendorff heart preparation as previously described [3]. All tri-peptide treated hearts received a dose of 50 μM peptide in plasma during the first five minutes of reperfusion via a syringe pump at 1 ml/min. Posttreatment trials received only tri-peptide prepared in plasma, prepared trials additionally received tri-peptide prepared in Krebs’ buffer a naïve (Nix, 10μM) during the last five minutes of baseline (pretreatment). Control hearts did not receive tri-peptide or noloxone. All hearts were frozen at -20°C for 30 min, sectioned into 2mm slices and incubated at 37°C in 1% 2,3,5-Triphenyltetrazolium chloride (TTC) to determine infarct size (figure 2).

RESULTS

Male Sprague-Dawley rats (275-325 g, Charles River, Springfield MA) were anesthetized i.p. with sodium pentobarbital (60 mg/kg) and anesthetized with heparin 1000 units. Hearts were isolated and studied using a modified Langendorff heart preparation as previously described [3]. All tri-peptide treated hearts received a dose of 50 μM peptide in plasma during the first five minutes of reperfusion via a syringe pump at 1 ml/min. Posttreatment trials received only tri-peptide prepared in plasma, prepared trials additionally received tri-peptide prepared in Krebs’ buffer a naïve (Nix, 10μM) during the last five minutes of baseline (pretreatment). Control hearts did not receive tri-peptide or noloxone. All hearts were frozen at -20°C for 30 min, sectioned into 2mm slices and incubated at 37°C in 1% 2,3,5- Triphenyltetrazolium chloride (TTC) to determine infarct size (figure 2).

Prepared tri-peptide + Nix (10μM, 10μM) Pre-treatment tri-peptide + Nix (10μM, 10μM)

Conclusions

Pretreatment with a novel tri-peptide significantly restored both post-reperfusion cardiac function and reduced infarct size compared to untreated control (IR) hearts, IR + posttreated tri-peptide hearts, and pretreated tri-peptide + noloxone + IR hearts. The improvement in post-reperfusion heart function is mostly due to a decrease in UDEP.

Noloxone blocked the cardioprotective effects of tri-peptide during pretreatment, suggesting that the mechanism of tri-peptide likely involves preconditioning via opioid receptor activation which in part involves opening of mitochondrial channels. The results from this study suggest that novel tri-peptide would be an effective treatment that can be given to heart transplant patients to restore heart function and reduce reperfusion injury in heart tissue.

Future studies will aim to identify the opioid receptor subtype mediating the preconditioning effect of tri-peptide. Additional studies will delineate whether the preconditioning mechanism of opioid receptor activation involves the opening of mitoKATP channels by blocking the channel with 5-hydroxydecanoate (5-HD).

REFERENCES