Pharmacological study of therapeutic outcomes of carvedilol and carnitine combination in isoprenaline-induced myocardial infarction in rats

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Abstract

Mortality associated with acute myocardial infarction (AMI) is still a leading cause of death in both developing and developed countries. The effect of early treatment with ß-blockers in reducing mortality and ischemic events after AMI is well known. The beta blocker; carvedilol; is an effective antianginal and antihypertensive agent and significantly limits infarct size in animal models. Carvedilol has previously been demonstrated to be beneficial in patients with AMI. However, the overall success of beta blockers is limited, and usually adverse reaction is clear. As such, it is important to evaluate new therapeutic modalities in addition to standard medical therapy.

Metabolic agents are a new class of drugs that directly modify the use of energy substrates in the heart, lessening ischaemic injury and improving cardiac performance during ischaemia. One of these metabolic agents is carnitine. Increasing evidences also demonstrate that oxidative stress plays an important role in experimental models of MI. Thymoquinone, the main volatile oil constituent of *Nigella sativa* seed, is reported to possess strong antioxidant properties. The protective effect of carvedilol-carnitine combination, and carvedilol-thymoquinone combination on myocardium was investigated using isoprenaline (ISO)-induced myocardial infarction (MI) model in rats.

Firstly, the effect of a single dose of drugs on ISO (250 mg/kg, S.C.)-induced MI was investigated; resembling acute MI treatment. Image analysis was carried out and levels of diagnostic marker enzymes in plasma, cardiac glutathione and lipid peroxides were measured. Injection of ISO caused animal mortality (46%), 60% infarction size, significant reduction in cardiac GSH level, and increased levels of diagnostic marker enzymes in plasma and lipid peroxidation in heart tissue.

Treatment with carvedilol, carnitine, or thymoquinone significantly prevented ISO-induced cardiac toxicity as evidenced by reduced mortality, reduced myocardial infarction size, decreased serum enzymes, and maintained myocardial antioxidant status at near normal status. While combination of carvedilol with either carnitine, or thymoquinone, didn't show significant difference from carvedilol group.

Secondly, the effect of daily administration of drug combination for 2 weeks started 24 hours following ISO injection (250 mg/kg S.C.) was investigated. Tissue pathology, heart weight, collagen content, and serum TNF-α were assessed. ISO induced focal area of fibrosis, increased heart weight, increased collagen content, but didn't affect TNF-α level. Thymoquinone didn't show any significant difference from ISO group. Carnitine, and carvedilol showed significant protection against post-MI change. Combination of carvedilol with thymoquinone didn't show significant difference

from carvedilol group, while, carnitine showed histopathological improvement when added to carvedilol.

From the results we conclude that, adding carnitine or thymoquinone to carvedilol didn't show additional beneficial effect compared to carvedilol alone against ISO-induced AMI. Regarding post MI remodeling, carvedilol-carnitine shows potential additive effect, while combination of carvedilol with thymoquinone didn't show beneficial effect compared to carvedilol alone. In addition, our results show that thymoquinone represents a potential cardioprotective agent aginst acute ISO-induced MI, while no protective effect was observed aginst post-MI remodeling.

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Myocardial infarction

Cardiovascular diseases form a major health concern in recent years **(Dhandapani et al., 2007)**. It is the leading cause of mortality and a major cause of morbidity. Coronary heart disease (CHD) accounts for nearly half of all CVD deaths **(Nasir et al.; 2006)**. CHD kills over 6.5 million people worldwide each year. CHD is a condition in which the vascular supply to the heart is impeded by atheroma, thrombosis or spasm of coronary arteries. This may impair the supply of oxygenated blood to cardiac tissue sufficiently to cause myocardial ischaemia which, if sever or prolonged, may cause the death of cardiac muscle cells; myocardial infarction (MI) **(Scott and Dwight, 2007).**

The consequences of MI are not benign. According to the statistics given by WHO in 2004 about 16.7 million people around the globe die of MI every year, which forms about one- third of the total global deaths. Among those who survive to reach hospital alive, approximately 12% of patients with ST segment elevation MI will die in the succeeding six months, and 13% of those with non-ST segment elevation MI. The frequency of new stroke is between 1.5–3%, and

rehospitalisation for a further MI is between 17–20% in the same time interval **(Fox, 2004).** It is predicted that heart disease and stroke will become the leading cause of death and disability world-wide by the year 2020, with the number of fatalities projected to increase more than 20 million a year and to more than 24 million a year by 2030 **(Dhandapani et al., 2007).**

1- Pathogenesis of MI

a- Pathogenesis of acute MI (AMI)

MI is the acute condition of necrosis of the myocardium that occurs as a result of imbalance between coronary blood supply and myocardial oxygen demand, resulting in myocardial ischaemia and accumulation of waste metabolites **(Rajadurai and Prince, 2007 and Devika and Stanely Marinez Prince, 2008).** MI can be regarded as a metabolic problem involving disruption of mitochondrial oxygen consumption and subnormal aerobic ATP resynthesis, resulting in ATP depletion, decrease of mitochondrial NADH oxidation, activation of glycolysis and accumulation of lactate and H+ **(Wang et al., 2007).** Membrane fragmentation and cell swelling are observed. Myofibril contracture bands are usually

considered characteristic **(Piper et al., 2006).** Necrosis is not the only aspect associated with irreversible lesions. Both necrosis and apoptosis are present in hearts subjected to permanent total ischaemia **(Monassier, 2008).**

MI is often due to atherosclerotic disease of the coronary arteries. It is a consequence of disruption of a vulnerable coronary artery plaque, complicated by intraluminal thrombosis, embolisation, and varying degrees of obstruction to perfusion. The severity of coronary arterial obstruction and the volume of affected myocardium determine the characteristics of clinical presentation **(Bertrand et al., 2002, Braunwald et al., 2002; and Van de Werf et al., 2003 and).** Additionally, coronary spasm, emboli, or dissection of the coronary artery are causes of infarction in the absence of occlusive atherosclerosis, and are reported in 5–10% of patients **(White and Chew, 2008).** Incomplete occlusion at the site of a disrupted arterial plaque may produce ischaemia or microinfarction, depending on the volume of myocardium affected and the extent of distal embolisation **(Van de Werf et al., 2003).**