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ISOPRENALINE: A TOOL FOR INDUCING MYOCARDIAL INFARCTION IN EXPERIMENTAL ANIMALS

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ABSTRACT

Cardiovascular Diseases (CVDs) remain the principal cause of death in both developed and developing countries, accounting for roughly 20% of all worldwide deaths per year. Due to changing lifestyles in developing countries, such as India and particularly urban areas, Myocardial infarction is making an increasingly important contribution to mortality statistics. Myocardial infarction is defined as an acute condition of necrosis of the myocardium that occurs as a result of imbalance between coronary blood supply and myocardial demand. Isoprenaline/Isoproterenol (ISO) is a synthetic catecholamine and beta adrenergic agonist, which causes severe stress in the myocardium, resulting in an infarct like necrosis of the heart muscle in experimental animal. ISO-induced myocardial infarction serves as a well standardized model because the pathophysiological changes in heart muscle of experimental animal, similar to that observed in human myocardial infarction. The present studies cover the cardioprotective activity of various drugs against Isoprenaline induced Myocardial infarction in animal model.

Keywords: Cardiovascular diseases, Myocardial infarction, Isoprenaline, Experimental animal, necrosis.

INTRODUCTION

Isoprenaline (isoproterenol) is a sympathomimetic that acts almost exclusively on beta-adrenergic receptors. It is listed in the 2004 WHO Model List of Essential Medicines. It is used to increase the heart rate for the treatment of patients with severe bradycardia that is unresponsive to atropine; for the short-term emergency treatment of heart block; for ventricular arrhythmias secondary to atrioventricular nodal block [1], during electrophysiological study, to facilitate the induction of supraventricular and ventricular tachycardias [2- 6]. Isoprenaline (Figure 1) is known to accelerate the sinus node and to enhance AV nodal conduction; the drug has no effect on His-Purkinje conduction time [7]. Paradoxical bradycardia is an unusual phenomenon. Pharmacological alternatives include atropine and for Torsades de Pointes magnesium sulfate. Cardiac pacing is an option for the treatment of patients with bradyarrhythmias or Torsades de Pointes [8, 9].

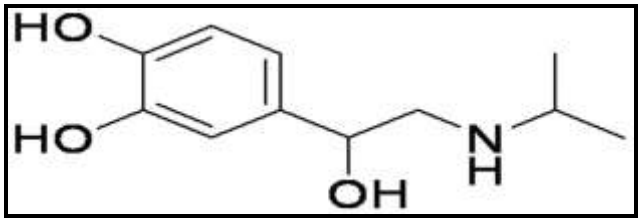

Figure 1: Structure of Isoprenaline

PHARMACOLOGY

Pharmacodynamic

- ***Cardiovascular system:*** Isoproterenol produces powerful stimulation of the heart to increase its rate andforce of contraction, causing increased cardiac output. It is as active as epinephrine in this action and, therefore, is useful in the treatment of atrioventricular block or cardiac arrest. Isoproterenol also dilates the arterioles of skeletal muscle (β2 effect), resulting in decreased peripheral resistance. Because of its cardiac stimulatory action, it may increase systolic blood pressure slightly, but it greatly reduces mean arterial and diastolic blood pressure [10].
- ***Pulmonary system:*** A profound and rapid bronchodilation is produced by the drug (β2 action). Isoproterenol is as active as epinephrine and rapidly alleviates an acute attack of asthma when taken by inhalation (which is the recommended route). This action lasts about 1 hour and may be repeated by subsequent doses [10].
- ***Other effects:*** Other actions on β- receptors, such as increased blood sugar and increased lipolysis, can be demonstrated but are not clinically significant [10].

Pharmacokinetics

Isoproterenol can be absorbed systemically by the sublingual mucosa but is more reliably absorbed when given parenterally or as an inhaled aerosol. It is a marginal substrate for COMT and is stable to MAO action [10].

Adverse effects: The adverse effects of Isoprenaline are similar to epinephrine.

- Cardiovascular: Angina, flushing, hyper-/hypotension, pallor, palpitation, paradoxical bradycardia (with tilt table testing), premature ventricular beats, Stokes-Adams attacks, tachyarrhythmia, ventrical ventricular arrhythmia.
- Central nervous system: Dizziness, headache, nervousness, restlessness, Stokes-Adams seizure.
- Endocrine & metabolic: Hypokalemia, serum glucose increased
- Gastrointestinal: Nausea, vomiting. Neuromuscular & skeletal: Tremor, weakness
- Ocular: blurred vision.
- Respiratory: Dyspnea, pulmonary edema [11].

Indications

- Mild or transient episodes of heart block that do not require electric shock or pacemaker therapy
- Serious episodes of heart block and Adams-Stokes attacks (except when caused by ventricular tachycardia or fibrillation)
- Cardiac arrest until electric shock or pacemaker therapy is available.
- Bronchospasm during anesthesia
- Adjunct to fluid and electrolyte replacement therapy and other drugs and procedures in the treatment of hypovolemic or septic shock.
- Low cardiac output states (eg, decompensated heart failure, cardiogenic shock) [11].

Isoprenaline forms available in market

Isoprenaline hydrochloride: Isoprenaline is available as an injection containing isoprenaline hydrochloride 20mcg/mL (1 - 3ml). Isoprenaline hydrochloride contains not less than 98.0 per cent and not more than the equivalent of 101.5 per cent of (1*RS*)-1-(3,4-dihydroxyphenyl)2 [(1methylethyl) amino]ethanol hydrochloride, calculated with reference to the dried substance. A white or almost white, crystalline powder, freely soluble in water, sparingly soluble in alcohol, practically insoluble in methylene chloride. Details are listed in table 1 [12].

Isoprenaline sulphate: Isoprenaline sulphate contains not less than 98.0 per cent and not more than the equivalent of 102.0 per cent of bis[(1RS)-1-(3,4-dihydroxyphenyl)-2-[(1-methylethyl)amino]ethanol] sulphate, calculated with reference to the anhydrous substance. A white or almost white, crystalline powder, freely soluble in water, very slightly soluble in alcohol. It melts at about 128 °C, with decomposition [12].

Dosage

- The Formulary recommends isoprenaline for the treatment of adults with *bradyarrhythmias*, for which it is administered by intravenous infusion at a dose of 1-4mcg/minute
- For the treatment of adults with *other cardiac disorders* administered by slow intravenous injection at a dose of 20 – 60 mcg adjusted according to ventricular rate
- For adults with *heart block* (acute Stokes-Adams attack) administered by intravenous infusion at a dose of 4-8 mcg/minute [12].

Table 1: Trade Names of Marketed Isoprenaline [13]

S. No.	Brand Name	Manufacturers	Type	Unit
1	Autohaler	Cipla Limited	Inhaler	400mcg/1mdi
2	Isolin	Samarth Pharma Pvt. Ltd	Injection	2mg/1ml
3	Isoprin	Unichem Laboratories Ltd.	Injection	2mg/2ml
4	Isosol	SG Pharma Pvt. Ltd.	Injection	2mg/3ml
5	Isuprin	Lemery	Injection	2mg
6	Neo –Epinine	Glaxo Smithkline Pharmaceuticals Ltd.	Tablet	20mg

MECHANISMS OF ISOPRENALINE INDUCED MYOCARDIAL INFARCTION

Myocardial infarction induced by ISO has been reported to show many metabolic and morphologic aberrations in the heart tissue of the experimental animals similar to those observed in human myocardial infarction [14]. ISO induced necrosis is maximal in the subendocardial region of the left ventrical and in the interventricular septum. Continuous infusion of ISO in rats elicits typical cardiac gene expression similar to that observed in cardiac hypertrophy caused by pressure overload. Amidst several mechanisms proposed to explain the isoproterenol-induced myocardial harm, one might say: an unbalance between oxygen supply to and demand from cardiomyocytes inwardly, which is related to myocardial hyperfunction due to increase both in chronotropism and inotropism as well as to hypotension in the coronary bed [15]. Secondly, it is also claimed that there is an elevation of Ca++ overcharge inside the cell [16]. In addition, that ion is related to the activation of the adenylate cyclase enzyme and the depletion of ATP levels on the course of the events [17]. Eventually, there is an oxidative stress augmentation because of several metabolic products originated from isoproterenol, not to mention free radicals genesis [18]. A schematic diagram is shown to explain the mechanism of action of Isoprenaline (Figure 2).

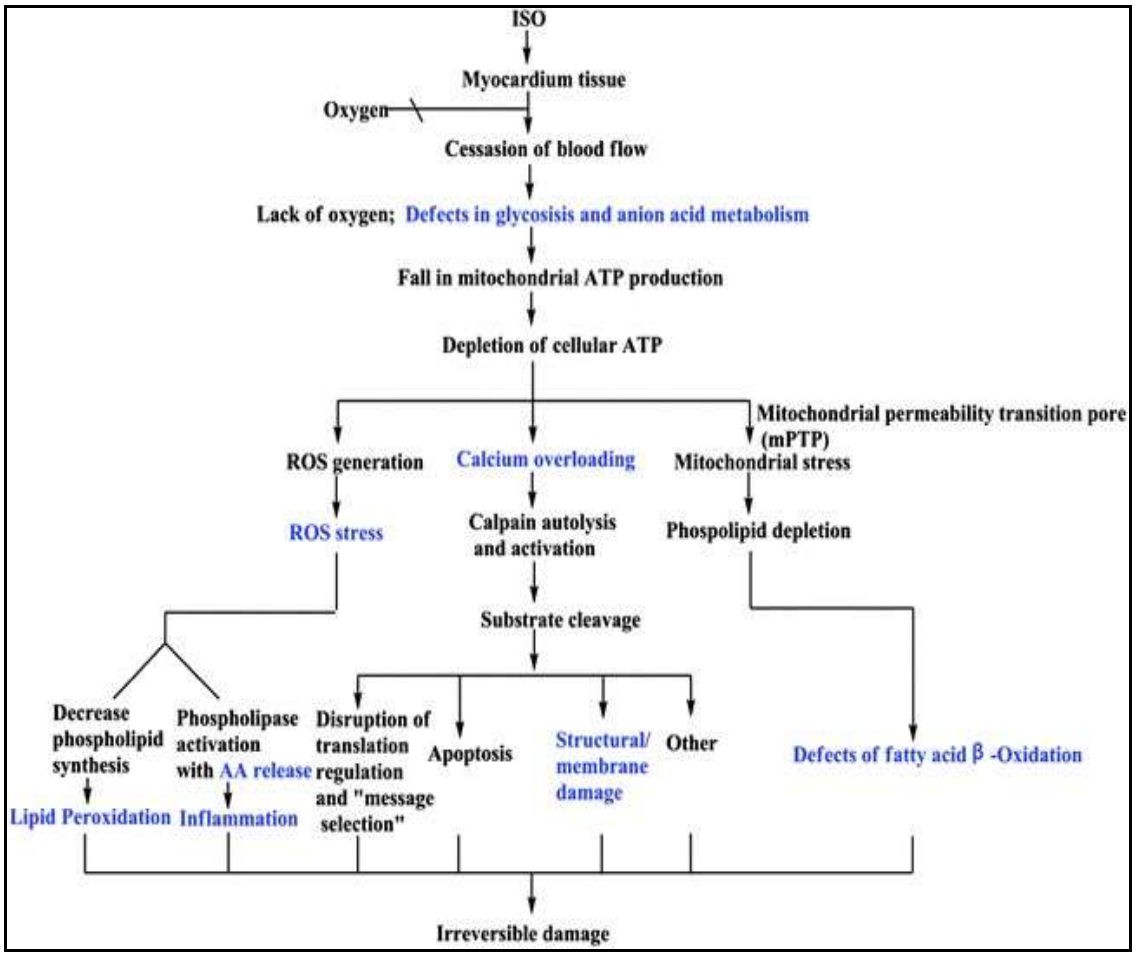

Figure 2: Mechanism of Isoprenaline induced myocardial infarction

DOSE OF ISOPRENALINE TO INDUCED MYOCARDIAL INJURY

Based on the available literature the ISO-induced effects on heart could be divided into 3 groups depending on the dose and duration of ISO administration:

• Low doses of isoproterenol (0.3–6 mg/kg body weight) administered acutely or repeatedly during 1–3 weeks

• Medium doses of isoproterenol (10–85 mg/kg body weight) applied in a single dose

• High doses of isoproterenol (150–300 mg/kg body weight) applied in a single dose or in two consecutive doses.

Low-dose ISO models

Very low doses of ISO 0.3 mg/kg applied for 7 days did not affect the blood pressure in rats [19]. However, it was shown that low doses of ISO (0.3 to 6 mg/kg) induce cardiac hypertrophy accompanied by fibrosis and necrosis of the tissue [20-29].

Medium-dose ISO models

Two-days lasting administration of increased ISO dose (40 mg/kg body weight) led to a significant but temporary reduction in systolic and diastolic blood pressure, however, prolonged administration of ISO did not affect blood pressure [30-31]. The medium doses of ISO (10–85 mg/kg) induced structural changes of mitochondria that are characterized by swelling, by decreased amount of cristae and increased presence of the homogenized matrix in mitochondrial population [32-34].

High-dose ISO models

It was shown that high doses of ISO, within the 85–300 mg/kg range, induced diffuse myocardial necrosis and ultimately lead to progressive left ventricular dilatation and myocardial hypertrophy [35-39]. The high dose of ISO induced in rat heart similar myocardial damage as acute myocardial infarction. This finding suggested that high dose of ISO could be used as a model of heart failure induced by acute myocardial infarction [40]. Table 2 shows list of various drugs used for cardioprotection and dose of Isoprenaline required for inducing myocardial infarction.

STUDY OF VARIOUS DRUGS AGAINST ISOPRENALINE

Table 2: Cardioprotective effect of various drugs against Isoprenaline induced Myocardial Infarction in animals

S.No.	Drugs	Effective Dose of Drugs	Isoprenaline Dose to induce MI	Investigation Parameters	Ref. no.
1	*Garcinia indica*	250mg/kg b.w., 500mg/kg b.w.	25mg/kg. b.w.	AST, ALT, LDH, CPK, and CK-MB in serum	41
2	Atorvastatin and Quercetin	10 mg/kg and 50 mg/kg for 14 days	100 mg/kg; s.c. in last 2 days	CK-MB activity as well as cTn-I, CRP, TNF-α, and IL-10 levels, ECG, histopathology	42
3	Ritonavir	10 mg/kg/day i.p. twice daily for 2 days	150 mg/kg/day, i.p for 2 consecutive days	Serum markers SGOT and CK, heart/body weight ratio, nitric oxide level , SOD,GSH, TBARS and catalase, histopathology	43
4	*Commiphora mukul*	Three doses 100, 200 and 400 mg kg-1 p.o. for 30 days	85 mg kg-1; s.c. for 2 consecutive days	Myocardial antioxidants SOD, CAT, GSH, GSHPx, marker enzymes CK-MB, LDH, histopathology	44
5	Glycyrrhizic acid	Three doses 5, 10 and 20 mg/kg BW i.p. for 14 days	85 mg kg-1; s.c. for 2 consecutive days	SOD and GSH levels, LPO and IP levels, histopathology	45
6	*Curcuma longa*	50, 100 and 200 mg/kg p.o. for 30 days	85 mg kg-1; s.c. for 2 consecutive days	Biochemical SOD, CAT, TBARS, GSHPx, CPK, hemodynamic MAP, LVEDP and histopathology	46
7	*Tylophora indica*	100 mg/kg, 200 mg/kg for 30 days orally	150 mg kg-1; s.c. for 2 consecutive days	Enzymes LDH, CK-MB ,antioxidants such as SOD and catalase, histopathology	47
8	Abana	75 mg / 100 g	20 mg / 100 g subcutaneously twice at an interval of 24 hrs	Serum enzymes CPK, GOT, GPT and γ-GT	48
9	Magnesium chloride	40 mg/kg body weight i.v.	2 mg/kg body weight	Etermination of cardiac enzyme CK and histopathological changes	49
10	Garlic	250 and 500 mg kg-1 once daily for 3 weeks	100 mg kg-1; s.c. for 2 consecutive days	Etermination of cardiac enzyme CK and histopathological changes	50
11	*Urtica parviflora*	350 mg/kg and 500 mg/kg, p.o for 15 days	200 mg kg-1; s.c. for 2 consecutive days	ALT, AST, ALP, LDL, TC, CAT, GSH, body weight	51
12	*Syzygium*	250 mg/kg and	20 mg/100 g s.c.	AST, ALT, CPK, LDH	52

	cumini	500 mg/kg			
13	Luteolin	0.3 mg/kg body weight for 30 days	85 mg kg-1; s.c. for 2 consecutive days	Free radical scavenging, mitochondrial lipids, antioxidants and mitochondrial enzymes, histopathology	53
14	Caes*alpinia crista*	400 mg/kg body wt., administered orally for 30 days	85 mg kg/1; s.c. for 2 consecutive days	Marker enzymes LDH, CK-MB, SGOT, SGPT, lipid peroxide, glutathione, Plasma TC, TG, HDL, VLDL, histopathology	54
15	*Cucumis trigonus*	75 and 150 mg kg□1 daily for a period of 14 days	200 mg kg/1; s.c. for 2 consecutive days	Serum marker enzymes (ALT, AST, LDH and CPK), ECG, histopathology	55
16	*Tinospora cordifolia*	350 mg/kg and 650 mg/kg body weight , orally for 28 days	85 mg kg/1; s.c. for 2 consecutive days	Cardiac enzymes such as AST, ALT, CK-MB, LD, troponin-I ,Physical parameters were gross examination of heart, heart weight/body weight ratio, histopathology examination	56
17	Glutathione	200 mg/kg body wt) orally for 30 days	100mg/kg s.c. at an interval of 24 hrs on 31st and 32nd day	Marker enzymes (AST, ALT, LDH and CKMB), plasma and lipid peroxidation, and heart antioxidant enzymes (SOD, glutathione peroxides, catalase) and reduced glutathione	57
18	*Nigella sativa*	150 mg/kg body weight intragastrically for a period of 15 days	85 mg/kg body weight	Enzymes (AST, ALT, LDH, CK), lipid profile (TG, cholesterol, free fatty acids, HDL, LDL, VLDL).	58
19	Fenugreek	250 mg/kg body weight) intragastric intubation for 15 days	85 mg/kg body weight) intraperitoneal (i.p.) for two consecutive days	Antioxidants (SOD, CAT, GPx and GSH), Histopathological studies	59
20	Curcumin	100, 200, and 400 mg/kg orally for 15 days	85 mg/kg, s.c. on 13th and 14th day	Glutathione, SOD, CK-MB, LDH, TBRAS	60
21	Folic acid and Vitamin B12	10 mg kg−1, orally and 500μg kg−1, i.m. for 4 weeks	single injection of 300 mg kg−1, s.c.	Electrocardiographic parameters, heart rate, ST segment, and blood pressure, CK and LDH levels, Histopathological studies	61
22	*Vitis vinifera (grapes seed)*	100mg/kg	85 mg/kg	Maintained levels of marker enzymes (AST, ALT, and LDH & CK) and antioxidants levels.	62

23	*Punica granatum*	100mg/kg	85mg/kg	Levels of marker enzymes (AST, ALT, and LDH & CK) and antioxidants levels, Histopathology.	63
24	*Crocus sativus*	20mg/kg	85mg/kg	Maintained levels of marker enzymes (AST, ALT, and LDH &CK) and antioxidants levels.	64
25	*Sida cordifolia*	100mg/kg	150mg/kg	Maintained levels of marker enzymes (AST, ALT, and LDH &CK) and antioxidants levels.	65
26	Metformin	150 mg/kg/24 h and 10 mg/kg	15 mg/kg/24 h for 1 week	Heart-to-body weight ratio, Histopathology	66

CONCLUSION:

This work highlights and summarizes the cardioprotective effects of Isoprenaline induced myocardial infarction. From above study we conclude that Isoprenaline acts as an effective tool for inducing myocardial infarction in experimental animal models.

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