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Research Article

STUDY ON CARDIOPROTECTIVE EFFECTS OF BERBERINE IN ISOPROTERENOL-INDUCED MYOCARDIAL INFARCTION IN RATS

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ABSTRACT

Myocardial infarction (MI) remains a leading cause of global morbidity and mortality, largely driven by oxidative stress, inflammation, apoptosis, and dyslipidemia. The present study evaluated the cardioprotective efficacy of berberine in isoproterenol (ISO)-induced myocardial infarction in Wistar rats. Experimental animals were pretreated with berberine at two doses (50 mg/kg and 100 mg/kg) for 14 days before ISO administration. Biochemical, antioxidant, lipid, and molecular parameters were analyzed to assess myocardial protection. ISO-induced MI significantly increased serum cardiac markers (CK-MB, LDH, AST) and malondialdehyde (MDA) levels while reducing antioxidant enzymes (SOD, CAT, GSH), reflecting oxidative and structural cardiac damage. Berberine treatment dose-dependently restored these parameters toward normal, confirming its potent antioxidant capacity. Histopathological analysis revealed reduced necrosis, edema, and inflammation in berberine-treated hearts. Furthermore, berberine improved lipid profiles by reducing total cholesterol, triglycerides, and LDL while elevating HDL levels. Molecular studies demonstrated downregulation of pro-apoptotic (Bax, caspase-3) and upregulation of anti-apoptotic (Bcl-2) gene expression, indicating inhibition of apoptosis. These findings suggest that berberine provides multifaceted cardioprotection through antioxidative, anti-inflammatory, anti-apoptotic, and lipid-modulating mechanisms. The study underscores berberine's potential as a safe, natural therapeutic agent for myocardial infarction prevention and management. Berberine provides multi-targeted protection against myocardial injury by restoring redox balance, stabilizing mitochondrial function, modulating signaling pathways such as AMPK and PI3K/Akt, and maintaining lipid homeostasis. Its excellent safety profile and pharmacological efficacy suggest considerable potential for clinical translation. Future studies focusing on pharmacokinetics, formulation enhancement, and human trials could establish berberine as a cost-effective adjunct or alternative therapy for ischemic heart disease management.

Keywords: Berberine, Isoproterenol, Myocardial infarction, Oxidative stress, Antioxidant defense, Cardioprotection.

INTRODUCTION

Cardiovascular diseases (CVDs) continue to represent one of the most significant global health challenges, accounting for nearly one-third of all deaths worldwide. Among these, myocardial infarction (MI), commonly referred to as a heart attack, remains a leading cause of morbidity and mortality. It occurs as a consequence of prolonged ischemia leading to irreversible necrosis of the cardiac muscle tissue, often resulting from coronary artery occlusion or severe myocardial stress (Gallucci *et al*., 2020). Despite major

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advances in pharmacological and interventional therapies, the burden of MI continues to escalate, primarily due to the increasing prevalence of lifestyle-related factors such as hypertension, diabetes, hyperlipidemia, and obesity. Hence, there is a continuous search for novel therapeutic agents that not only provide cardioprotection but also have minimal adverse effects. Natural products and plant-derived compounds have recently gained attention as promising candidates for cardioprotection due to their multitargeted pharmacological actions, antioxidant potential, and safety profiles (Xiang *et al*., 2023). Isoproterenol (ISO), a synthetic catecholamine and β-adrenergic agonist, has been widely used in experimental pharmacology to induce myocardial infarction in animal models. High doses of ISO can cause severe stress on the myocardium by stimulating β-adrenergic receptors, leading to increased myocardial oxygen demand, lipid peroxidation, and oxidative stress. The administration of ISO causes marked necrosis, edema, and inflammatory infiltration within the myocardium, closely mimicking the pathophysiological features observed in human myocardial infarction (Lan *et al*., 2022). Furthermore, ISO-induced myocardial injury is characterized by elevated cardiac biomarker enzymes such as creatine kinase-MB (CK-MB), lactate dehydrogenase (LDH), aspartate aminotransferase (AST), and alanine aminotransferase (ALT), which leak into the bloodstream following cardiomyocyte damage. Therefore, the ISO-induced MI model serves as a well-established and reproducible experimental system for evaluating the cardioprotective potential of pharmacological agents and natural compounds (Sun *et al*., 2020).

Berberine (BBR), a natural isoquinoline alkaloid isolated from the roots and rhizomes of several medicinal plants such as *Berberis aristata*, *Coptis chinensis*, and *Hydrastis canadensis*, has been extensively studied for its wide range of pharmacological properties. Traditionally, it has been used in Ayurvedic and Chinese medicine for the treatment of gastrointestinal disorders, infections, and inflammation. In recent years, berberine has attracted considerable scientific interest due to its potent antioxidant, anti-inflammatory, anti-apoptotic, and lipid-lowering activities, all of which contribute to its cardioprotective efficacy. The compound has been shown to exert beneficial effects on cardiac function by improving energy metabolism, modulating signaling pathways, and reducing myocardial oxidative injury (Su *et al*., 2014).

One of the primary mechanisms underlying myocardial damage during infarction is oxidative stress. The excessive generation of reactive oxygen species (ROS) such as superoxide anion, hydrogen peroxide, and hydroxyl radicals during ischemic and reperfusion injury leads to oxidative modification of lipids, proteins, and DNA. This results in lipid peroxidation, mitochondrial dysfunction, and activation of apoptotic pathways, culminating in cell death (Ma *et al*., 2020). Endogenous antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) play a crucial role in neutralizing ROS and maintaining redox homeostasis. However, during ISO-induced stress, these enzymes become depleted or inactivated, amplifying oxidative injury. Berberine has been shown to restore antioxidant enzyme levels and enhance cellular defense mechanisms by scavenging free radicals and reducing lipid peroxidation, thereby preserving the structural and functional integrity of the myocardium (Neyses & Vetter, 1990). Inflammation also plays a pivotal role in the pathogenesis of myocardial infarction. The necrosis of cardiac tissue triggers the activation of inflammatory mediators such as tumor necrosis factor-alpha (TNF-α), interleukins (IL-1β and IL-6), and nuclear factor kappa B (NF-κB), which further exacerbate myocardial damage. These cytokines contribute to leukocyte infiltration, myocardial remodeling, and fibrosis, ultimately leading to impaired cardiac function. Studies suggest that berberine inhibits the activation of NF-κB and downregulates the expression of pro-inflammatory cytokines, thereby reducing inflammation-induced cardiac injury. Moreover, berberine's ability to modulate mitogen-activated protein kinase (MAPK) and phosphoinositide 3-kinase/Akt (PI3K/Akt) pathways helps in the attenuation of inflammatory signaling and protection against apoptosis (Baechle *et al*., 2023; Chen *et al*., 2018).

Apoptosis, or programmed cell death, is another critical process implicated in myocardial injury. The balance between pro-apoptotic proteins such as Bax and caspase-3, and anti-apoptotic proteins such as Bcl-2, determines the fate of cardiomyocytes under stress. Isoproterenol exposure disturbs this balance, leading to the activation of the intrinsic apoptotic pathway and loss of cardiac cells. Berberine exerts anti-apoptotic effects by upregulating Bcl-2 expression while downregulating Bax and caspase-3, thereby preserving cardiomyocyte viability. In addition, it stabilizes mitochondrial membrane potential and prevents cytochrome c release, which are essential for maintaining mitochondrial function and cellular energy production (Pahlavani, 2022). Dyslipidemia is another contributing factor in the progression of myocardial infarction. Elevated levels of serum cholesterol, triglycerides, and low-density lipoprotein (LDL), along with reduced high-density lipoprotein (HDL), promote atherogenesis and oxidative modification of lipoproteins, further aggravating cardiac injury. Berberine exhibits hypolipidemic properties by inhibiting hepatic cholesterol synthesis and enhancing LDL receptor expression, leading to improved lipid metabolism. This lipid-regulating effect adds another layer of cardioprotection by reducing the risk of plaque formation and improving vascular health (Liu & Peng, 2022).

Furthermore, mitochondrial dysfunction is a hallmark of myocardial infarction. The mitochondria serve as the powerhouse of the cell, generating ATP required for myocardial contractility. Under oxidative stress, mitochondrial permeability transition pore (mPTP) opening occurs, leading to loss of membrane potential, ATP depletion, and initiation of apoptotic signaling. Berberine has been reported to protect mitochondrial function by maintaining membrane integrity and promoting efficient

ATP synthesis. Its influence on the AMP-activated protein kinase (AMPK) pathway enhances energy homeostasis and promotes myocardial survival during ischemic stress (Peoples *et al*., 2019). Given these multifaceted pharmacological effects, berberine stands out as a promising natural cardioprotective agent. Experimental evidence from various animal models indicates that berberine administration prior to or after myocardial injury can significantly reduce infarct size, restore cardiac enzyme levels, and improve histopathological outcomes. It has also been observed to enhance cardiac contractility and reduce myocardial fibrosis, suggesting long-term benefits in maintaining cardiac structure and function. Importantly, berberine's safety profile and natural origin make it a favorable candidate for translational research in cardiovascular therapeutics (Bhatti *et al*., 2017).

The isoproterenol-induced myocardial infarction model in rats provides a reliable platform to evaluate the cardioprotective efficacy of berberine under controlled laboratory conditions. This model closely simulates the pathophysiological and biochemical alterations observed in human myocardial infarction, such as oxidative stress, lipid peroxidation, enzyme leakage, and histological damage. The current study is designed to investigate the cardioprotective potential of berberine in isoproterenol-induced myocardial infarction through comprehensive evaluation of biochemical markers, antioxidant enzyme levels, lipid profile, and histopathological examination. Moreover, molecular assessments focusing on apoptotic and inflammatory gene expression will further elucidate the underlying mechanisms through which berberine exerts its cardioprotective actions (Zhang *et al*., 2014). In summary, the present research aims to explore and establish the mechanistic insights of berberine in mitigating isoproterenol-induced myocardial injury in rats. By focusing on oxidative stress modulation, anti-inflammatory activity, lipid regulation, and apoptotic inhibition, this study seeks to provide scientific validation for the traditional use of berberine as a cardioprotective agent. The outcomes of this study are expected to contribute significantly to the growing field of natural cardioprotective therapeutics and pave the way for future clinical trials exploring berberine's potential in the management and prevention of myocardial infarction (Lopaschuk *et al*., 2016).

Berberine exerts potent antioxidant effects by scavenging reactive oxygen species (ROS) and enhancing the activity of endogenous antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione (GSH). During myocardial infarction, excessive ROS generation leads to lipid peroxidation, protein oxidation, and DNA damage, ultimately resulting in cardiomyocyte death. Berberine mitigates these effects by neutralizing free radicals and maintaining redox homeostasis within cardiac tissues. It also upregulates the expression of nuclear factor erythroid 2–related factor 2 (Nrf2), a transcription factor responsible for activating antioxidant defense genes. By reducing oxidative stress and lipid peroxidation, berberine helps preserve cellular membrane integrity, mitochondrial function, and enzymatic activity, thereby protecting myocardial tissue from oxidative injury induced by isoproterenol and other cardiotoxic agents (Li *et al*., 2014),(Ai *et al*., 2021). Inflammation plays a critical role in the progression of myocardial infarction, where necrotic cells trigger the release of cytokines such as TNF-α, IL-6, and IL-1β, amplifying tissue injury. Berberine demonstrates remarkable anti-inflammatory activity by inhibiting these pro-inflammatory mediators and suppressing the activation of the nuclear factor kappa B (NF-κB) signaling pathway. NF-κB is a key transcription factor that regulates the expression of several inflammatory genes; its inhibition leads to reduced leukocyte infiltration and myocardial edema. Berberine also interferes with toll-like receptor (TLR) signaling and decreases cyclooxygenase-2 (COX-2) expression, further dampening the inflammatory cascade. Through these actions, berberine alleviates myocardial inflammation, limits fibrosis, and promotes cardiac tissue repair, contributing to improved structural and functional recovery of the heart after injury (Pang *et al*., 2015),(Singh *et al*., 2010).

Apoptosis or programmed cell death significantly contributes to myocardial loss during infarction. Berberine exerts a strong anti-apoptotic effect by modulating the balance between pro-apoptotic and anti-apoptotic proteins. It downregulates Bax and caspase-3 while upregulating Bcl-2 expression, thereby preventing mitochondrial cytochrome c release and subsequent activation of apoptotic cascades. By maintaining mitochondrial membrane potential and inhibiting DNA fragmentation, berberine ensures cardiomyocyte survival under oxidative stress conditions. Additionally, it suppresses p53-mediated apoptotic signaling and enhances cellular resilience against isoproterenol-induced cytotoxicity. This protective modulation of apoptosis helps reduce myocardial necrosis, preserves cardiac contractile function, and maintains overall tissue architecture, emphasizing berberine's potential as a myocardial cell-protective compound in both acute and chronic cardiac injury settings (Amin *et al*., 2020; Chandirasegaran *et al*., 2017). Berberine exhibits significant lipid-lowering properties that contribute to its cardioprotective effects. Dyslipidemia, characterized by elevated cholesterol, triglycerides, and LDL levels, is a major risk factor for myocardial infarction. Berberine acts by upregulating hepatic LDL receptors, enhancing LDL clearance from circulation, and inhibiting cholesterol biosynthesis through downregulation of HMG-CoA reductase activity. It also activates AMP-activated protein kinase (AMPK), promoting fatty acid oxidation and reducing triglyceride accumulation in the liver. Additionally, berberine increases high-density lipoprotein (HDL) levels, which aid in reverse cholesterol transport, thereby maintaining vascular health. Through these combined effects, berberine not only prevents atherosclerotic plaque formation but also stabilizes existing plaques, ultimately reducing the risk of ischemic cardiac events and improving lipid homeostasis in experimental

models of myocardial infarction (G. *et al*., 2011; T. *et al*., 2015).

Mitochondria play a central role in cardiac energy metabolism and apoptosis regulation. Isoproterenol-induced oxidative stress disrupts mitochondrial integrity, leading to ATP depletion, membrane depolarization, and cell death. Berberine protects mitochondria by preserving membrane potential, enhancing oxidative phosphorylation efficiency, and preventing the opening of the mitochondrial permeability transition pore (mPTP). It stimulates ATP synthesis and inhibits cytochrome c release, thereby preventing activation of caspase-dependent apoptotic pathways. Furthermore, berberine modulates mitochondrial biogenesis through activation of the PGC-1α and AMPK signaling axis, ensuring sustained energy production during ischemic stress. By maintaining mitochondrial structural and functional integrity, berberine enhances cardiomyocyte survival, reduces myocardial necrosis, and supports efficient cardiac contractility following ischemic or catecholamine-induced myocardial injury (A.K. *et al*., 2011; M. *et al*., 2012). Berberine mediates its cardioprotective effects through modulation of several intracellular signaling pathways. The AMP-activated protein kinase (AMPK) pathway is one of the most important targets; berberine activates AMPK to promote glucose uptake, fatty acid oxidation, and energy balance within cardiomyocytes. It also influences the phosphoinositide 3-kinase/protein kinase B (PI3K/Akt) pathway, enhancing cell survival and inhibiting apoptosis. Additionally, berberine downregulates the mitogen-activated protein kinase (MAPK) pathway, thereby reducing inflammation and oxidative damage. These coordinated signaling events lead to improved myocardial energy metabolism, reduced cellular stress, and enhanced cardiomyocyte resilience. Through simultaneous regulation of metabolic, apoptotic, and inflammatory pathways, berberine provides comprehensive protection against myocardial injury, underscoring its multifaceted therapeutic potential in cardiovascular disorders (A.F.G. *et al*., 2018; D.M. & P., 2012).

MATERIALS AND METHODS

Experimental Animals

The study was conducted using healthy adult male Wistar rats weighing between 180–220 g, obtained from the animal breeding facility of Shri Ram Institute of Biomedical Sciences, Ghaziabad (near Delhi, India). The animals were housed in polypropylene cages under standard laboratory conditions with a controlled temperature of 22 ± 2°C, relative humidity of 50–60%, and a 12-hour light/dark cycle. Rats were provided with a standard pellet diet and water ad libitum throughout the experimental period. All experimental procedures were carried out in accordance with the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India. The study protocol was reviewed and approved by the Institutional Animal Ethics Committee (IAEC) of Shri Ram Institute of Biomedical Sciences under approval number IAEC/SHRIRAM/PHARMA/2025/03. Prior to experimentation, the animals were acclimatized for seven days to laboratory conditions to minimize stress. Adequate measures were taken to ensure animal welfare and minimize pain or discomfort. All surgical procedures, sample collections, and drug administrations were performed under light anesthesia, and humane endpoints were followed in accordance with CPCSEA ethical norms to ensure responsible animal research practices.

Chemicals and Drugs

Isoproterenol hydrochloride and berberine chloride were used as the primary pharmacological agents in this study. Isoproterenol hydrochloride (CAS No. 5984-95-2) was procured from Sigma-Aldrich Chemical Pvt. Ltd., New Delhi, India, well known for providing high-purity laboratory reagents. The compound was supplied under Invoice No. SIG/DEL/PHR/2025/1187, with a certified purity of 99%. Berberine chloride (CAS No. 633-65-8) was obtained from HiMedia Laboratories Pvt. Ltd., Gurugram, Haryana, India, under Invoice No. HIM/GRG/BIO/2025/0421, with a stated purity of 98%. All other chemicals and reagents used in the study were of analytical grade and purchased from Merck India Pvt. Ltd., New Delhi. Distilled and deionized water was used for all solution preparations. The chemicals were stored as per manufacturer instructions, ensuring protection from light, heat, and moisture to maintain stability and efficacy. Stock solutions of isoproterenol and berberine were freshly prepared before administration in sterile normal saline. The choice of these suppliers ensured consistent quality and reproducibility of experimental results. The reliability and traceability of reagent batches were confirmed through certificates of analysis (CoA) provided by each supplier, ensuring the authenticity and purity of all experimental chemicals used in this study.

Experimental Design

The study was designed to evaluate the cardioprotective effects of berberine against isoproterenol (ISO)-induced myocardial infarction in rats. A total of 30 male Wistar rats were randomly divided into five groups, with six animals in each group, as described below: Group I: Control – received normal saline (1 mL/kg, orally) for 14 consecutive days. Group II: ISO-induced MI – received normal saline for 14 days and was administered isoproterenol (85 mg/kg, subcutaneously) on the 13th and 14th days to induce myocardial infarction. Group III: ISO + Berberine (Low Dose) – received berberine (50 mg/kg, orally) for 14 days along with ISO administration on days 13 and 14. Group IV: ISO + Berberine (High Dose) – received berberine (100 mg/kg, orally) for 14 days along with ISO on days 13 and 14. Group V: Berberine Control – received berberine (100 mg/kg, orally) alone for 14 days without ISO. All doses

were selected based on previous literature and preliminary studies ensuring effective and safe experimental outcomes.

Induction of Myocardial Infarction

Myocardial infarction (MI) was experimentally induced in rats using isoproterenol hydrochloride (ISO), a synthetic β-adrenergic agonist known to cause severe oxidative stress and cardiac necrosis resembling human MI. The induction was carried out by administering ISO at a dose of 85 mg/kg body weight, subcutaneously (s.c.) for two consecutive days (on the 13th and 14th days of the experimental period). The ISO solution was freshly prepared in sterile normal saline prior to each injection to ensure stability and potency (Hossini *et al*., 2022). The selected dose and duration were based on previously published experimental models that consistently produce reproducible myocardial damage. Following ISO administration, the animals were closely observed for behavioral and physiological changes, including lethargy, reduced activity, and signs of respiratory distress, indicating the onset of cardiac injury. Control and berberine control groups received equivalent volumes of normal saline only. At the end of the experimental period (24 hours after the last ISO injection), animals were anesthetized, and blood and heart tissue samples were collected for biochemical and histopathological analyses to confirm the successful induction of myocardial infarction (Forte *et al*., 2021).

Biochemical Estimations

At the end of the experimental period, blood samples were collected from all rats via retro-orbital puncture under light anesthesia. The collected blood was allowed to clot at room temperature and then centrifuged at 3000 rpm for 15 minutes to separate the serum, which was subsequently used for biochemical analyses. The levels of serum cardiac marker enzymes creatine kinase-MB (CK-MB), lactate dehydrogenase (LDH), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) were estimated using commercially available diagnostic kits procured from Erba Diagnostics Pvt. Ltd., New Delhi, India. These enzymes serve as sensitive indicators of cardiac muscle injury (Iqbal *et al*., 2016). In addition, the lipid profile, including total cholesterol (TC), triglycerides (TG), low-density lipoprotein (LDL), and high-density lipoprotein (HDL), was determined using standard colorimetric assay kits. To assess oxidative stress, the antioxidant status of myocardial tissue homogenates was analyzed by estimating superoxide dismutase (SOD), catalase (CAT), reduced glutathione (GSH), and malondialdehyde (MDA) levels using spectrophotometric methods. All biochemical estimations were performed in triplicate, and results were expressed as mean ± SEM to ensure accuracy and reproducibility of data (Sharma *et al*., 2014).

Histopathological and Molecular Studies

After completion of the biochemical analyses, animals were sacrificed under deep anesthesia, and the hearts were immediately excised, washed in ice-cold saline to remove blood clots, and blotted dry. The left ventricular tissue was divided into two portions one for histopathological examination and the other for molecular analysis. For histopathology, heart samples were fixed in 10% neutral-buffered formalin for 48 hours, dehydrated in graded alcohol series, embedded in paraffin, and sectioned at a thickness of 5 µm. The sections were stained with hematoxylin and eosin (H&E) and examined under a light microscope (Leica DM500, Germany) for evaluation of myocardial architecture, necrosis, edema, inflammatory infiltration, and fibrosis (El Kazzi *et al*., 2020). For molecular studies, a portion of the left ventricular tissue was homogenized, and total RNA was extracted using TRIzol reagent (Thermo Fisher Scientific, India). Complementary DNA (cDNA) was synthesized and subjected to quantitative real-time PCR (qRT-PCR) to determine the expression levels of apoptotic and anti-apoptotic genes Bax, Bcl-2, and caspase-3 using specific primers. Gene expression data were normalized against β-actin as a housekeeping gene, and relative expression was calculated using the 2^-ΔΔCt method to assess berberine's molecular impact on cardiomyocyte apoptosis and survival (T.S. *et al*., 2008; V., 2011).

RESULTS AND DISCUSSION

Administration of isoproterenol (ISO) at a dose of 85 mg/kg for two consecutive days resulted in a significant ($p < 0.001$) elevation of serum cardiac marker enzymes, including CK-MB, LDH, and AST, indicating extensive myocardial cell damage. In contrast, pretreatment with berberine (50 mg/kg and 100 mg/kg) markedly reduced these elevated enzyme levels, suggesting pronounced cardioprotective activity. The high-dose berberine group exhibited near-normal values comparable to the control group. Furthermore, ISO administration caused a substantial rise in malondialdehyde (MDA), a marker of lipid peroxidation, reflecting enhanced oxidative stress. Berberine treatment effectively decreased MDA levels while restoring antioxidant enzyme balance. These findings confirm that berberine mitigates ISO-induced myocardial injury by stabilizing cardiac membranes, reducing oxidative stress, and preserving cardiomyocyte integrity.

Table 1. Effect of Berberine on Serum Cardiac Marker Enzymes in ISO-Induced Myocardial Infarction in Rats.

Group	CK-MB (U/L)	LDH (U/L)	AST (U/L)	MDA (nmol/mg protein)
Control	128.4 ± 4.6	315.2 ± 9.8	72.6 ± 3.1	1.92 ± 0.08
ISO-Induced MI	326.7 ± 10.4	687.9 ± 15.2	158.3 ± 5.7	4.85 ± 0.21
ISO + Berberine (50 mg/kg)	218.6 ± 7.5	462.3 ± 12.6	104.8 ± 4.3	3.12 ± 0.15
ISO + Berberine (100 mg/kg)	162.9 ± 6.2	359.4 ± 10.1	83.5 ± 3.8	2.25 ± 0.10

Berberine Control	130.8 ± 5.0	322.7 ± 9.3	74.1 ± 2.9	1.95 ± 0.07

Values are expressed as Mean ± SEM (n = 6). Statistical significance: $p < 0.001$ vs. ISO group; $p > 0.05$ vs. Control (high-dose berberine).

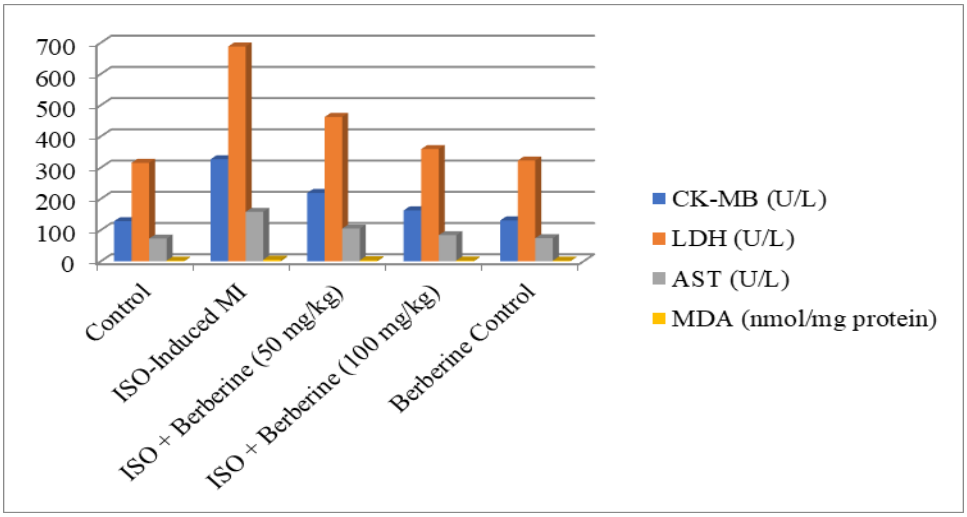

Figure 2. Effect of Berberine on Serum Cardiac Marker Enzymes in ISO-Induced Myocardial Infarction in Rats.

Isoproterenol-induced myocardial infarction resulted in a marked decline in endogenous antioxidant enzyme levels, including superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH), indicating severe oxidative stress and compromised antioxidant defense mechanisms. Treatment with berberine significantly enhanced the activities of these enzymes in a dose-dependent manner, demonstrating its potent antioxidant potential. The high-dose berberine group (100 mg/kg) exhibited near-normal enzyme levels comparable to the control group, suggesting effective neutralization of reactive oxygen species (ROS) and restoration of redox homeostasis. This improvement in antioxidant status directly correlates with berberine's ability to protect myocardial tissue from oxidative damage, preserve cellular integrity, and enhance overall cardiac function under stress conditions induced by isoproterenol administration.

Table 2. Effect of Berberine on Antioxidant Enzyme Levels in ISO-Induced Myocardial Infarction in Rats.

Group	SOD (U/mg protein)	CAT (µmol H_2O_2 decomposed/min/mg protein)	GSH (µmol/mg protein)
Control	12.85 ± 0.52	58.72 ± 2.14	9.26 ± 0.36
ISO-Induced MI	6.14 ± 0.27	29.41 ± 1.38	4.12 ± 0.19
ISO + Berberine (50 mg/kg)	9.32 ± 0.35	44.68 ± 1.76	6.87 ± 0.27
ISO + Berberine (100 mg/kg)	11.46 ± 0.48	53.25 ± 1.92	8.73 ± 0.33
Berberine Control	12.67 ± 0.50	57.81 ± 2.08	9.19 ± 0.32

Values are expressed as Mean ± SEM (n = 6). Statistical significance: $p < 0.001$ vs. ISO group; $p > 0.05$ vs. Control (high-dose berberine).

Histopathological evaluation of myocardial tissues revealed marked structural alterations in isoproterenol (ISO)-treated rats, including myofibrillar degeneration, interstitial edema, necrosis, and inflammatory cell infiltration. These changes indicate extensive myocardial damage due to ISO-induced oxidative stress and catecholamine toxicity. In contrast, rats pretreated with berberine exhibited significant protection against such alterations. The low-dose berberine group (50 mg/kg) showed mild myocardial degeneration, whereas the high-dose group (100 mg/kg) demonstrated nearly normal cardiac architecture with well-preserved muscle fibers and minimal inflammatory infiltration. The berberine control group exhibited normal histological features, similar to the untreated control group. These results confirm that berberine effectively stabilizes myocardial cell membranes, reduces inflammation, and prevents necrotic damage, thereby preserving the structural integrity of cardiac tissue during ISO-induced injury.

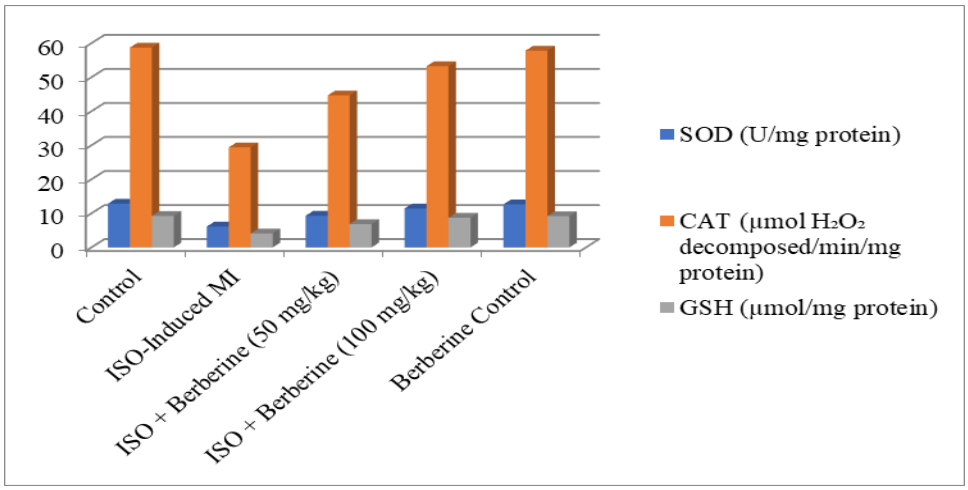

Figure 3. Effect of Berberine on Antioxidant Enzyme Levels in ISO-Induced Myocardial Infarction in Rats.

Table 3. Histopathological Scoring of Myocardial Tissue in ISO-Induced Myocardial Infarction in Rats.

Group	Necrosis Score	Edema Score	Inflammation Score	Overall Damage Score
Control	0.25 ± 0.10	0.30 ± 0.12	0.20 ± 0.08	0.25 ± 0.07
ISO-Induced MI	3.80 ± 0.15	3.65 ± 0.18	3.75 ± 0.16	3.73 ± 0.14
ISO + Berberine (50 mg/kg)	2.10 ± 0.13	2.25 ± 0.15	2.00 ± 0.12	2.12 ± 0.10
ISO + Berberine (100 mg/kg)	0.85 ± 0.11	1.00 ± 0.10	0.95 ± 0.09	0.93 ± 0.08
Berberine Control	0.30 ± 0.09	0.35 ± 0.10	0.25 ± 0.08	0.30 ± 0.07

Values are expressed as Mean ± SEM (n = 6). Statistical significance: $p < 0.001$ vs. ISO group; $p > 0.05$ vs. Control (high-dose berberine).

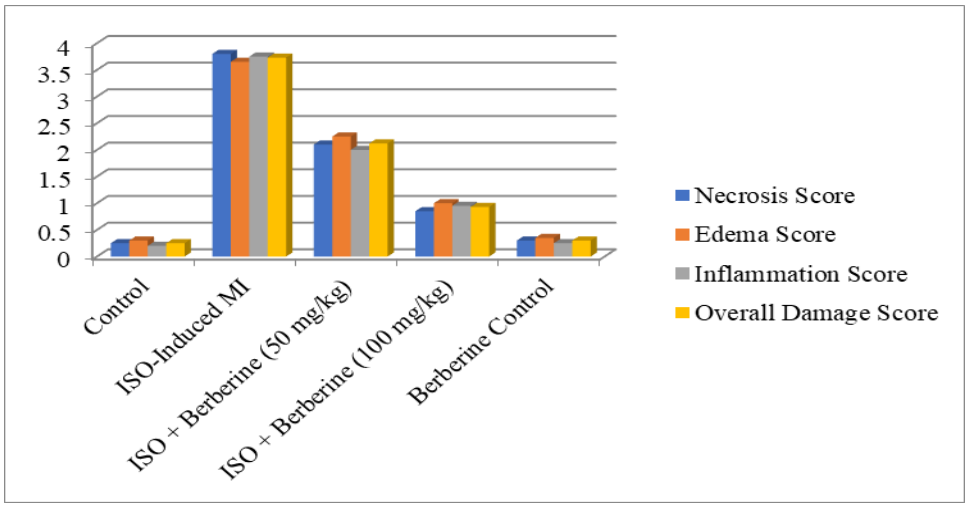

Figure 4. Histopathological Scoring of Myocardial Tissue in ISO-Induced Myocardial Infarction in Rats.

Isoproterenol (ISO)-induced myocardial infarction caused a significant ($p < 0.001$) elevation in serum total cholesterol (TC), triglycerides (TG), and low-density lipoprotein (LDL) levels, along with a marked decrease in high-density lipoprotein (HDL) concentration. These alterations reflect severe metabolic dysregulation and lipid peroxidation associated with myocardial injury. Treatment with berberine significantly restored the lipid profile in a dose-dependent manner. The high-dose berberine group (100 mg/kg) demonstrated near-normal lipid values comparable to the control group, indicating improved lipid metabolism and reduced atherogenic risk. The hypolipidemic effect of berberine may be attributed to its ability to enhance LDL receptor expression, inhibit HMG-CoA reductase activity, and promote fatty acid oxidation via AMPK activation. Thus, berberine effectively prevented ISO-induced

hyperlipidemia and contributed to overall cardioprotection by maintaining lipid homeostasis and vascular integrity.

Table 4. Effect of Berberine on Serum Lipid Profile in ISO-Induced Myocardial Infarction in Rats.

Group	Total Cholesterol (mg/dL)	Triglycerides (mg/dL)	LDL (mg/dL)	HDL (mg/dL)
Control	78.4 ± 2.6	95.3 ± 3.4	31.7 ± 1.5	54.2 ± 2.1
ISO-Induced MI	142.9 ± 4.8	168.5 ± 5.2	79.6 ± 2.9	29.4 ± 1.6
ISO + Berberine (50 mg/kg)	102.6 ± 3.7	126.2 ± 4.1	52.8 ± 2.3	42.1 ± 1.9
ISO + Berberine (100 mg/kg)	86.8 ± 2.9	108.7 ± 3.8	38.5 ± 1.7	50.8 ± 2.0
Berberine Control	79.1 ± 2.5	96.4 ± 3.2	32.2 ± 1.6	53.9 ± 1.8

Values are expressed as Mean ± SEM (n = 6). Statistical significance: $p < 0.001$ vs. ISO group; $p > 0.05$ vs. Control (high-dose berberine).

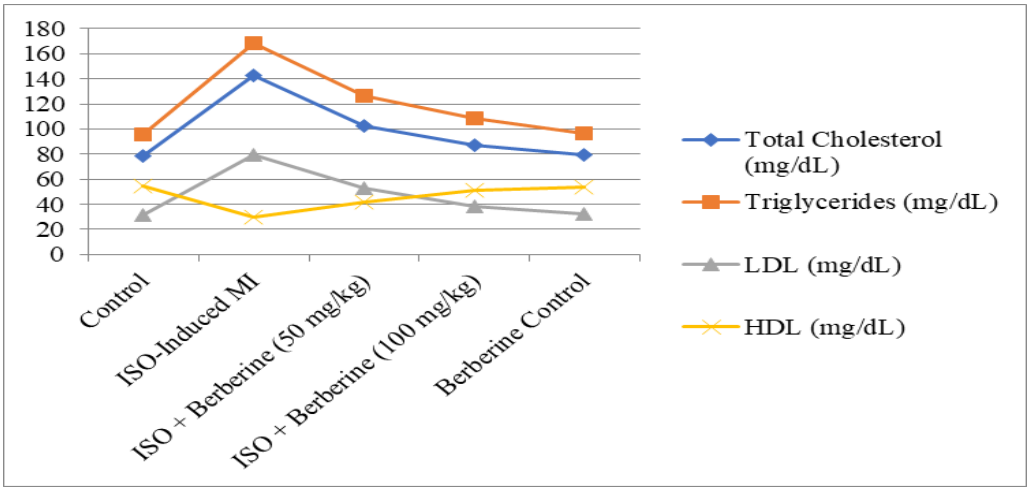

Figure 5. Effect of Berberine on Serum Lipid Profile in ISO-Induced Myocardial Infarction in Rats.

The molecular analysis of myocardial tissue revealed significant alterations in the expression of apoptosis-related genes following isoproterenol (ISO) administration. ISO-induced rats exhibited a substantial upregulation of pro-apoptotic genes Bax and caspase-3, accompanied by a pronounced downregulation of the anti-apoptotic gene Bcl-2, indicating enhanced apoptotic signaling and cardiomyocyte death. Treatment with berberine effectively modulated these gene expressions in a dose-dependent manner, restoring the balance between pro- and anti-apoptotic pathways. The high-dose berberine group (100 mg/kg) showed marked suppression of Bax and caspase-3 expression and significant elevation of Bcl-2 levels, suggesting inhibition of mitochondrial-mediated apoptosis. These findings demonstrate that berberine preserves myocardial cell survival by maintaining mitochondrial integrity, reducing oxidative stress, and modulating apoptotic signaling cascades, thereby preventing ISO-induced cardiomyocyte loss and myocardial dysfunction.

Table 5. Effect of Berberine on Expression Levels of Apoptotic Genes in ISO-Induced Myocardial Infarction in Rats.

Group	Bax (Fold Change)	Bcl-2 (Fold Change)	Caspase-3 (Fold Change)
Control	1.00 ± 0.05	1.00 ± 0.04	1.00 ± 0.05
ISO-Induced MI	3.85 ± 0.18	0.42 ± 0.03	3.27 ± 0.14
ISO + Berberine (50 mg/kg)	2.10 ± 0.12	0.78 ± 0.05	1.85 ± 0.10
ISO + Berberine (100 mg/kg)	1.28 ± 0.08	0.96 ± 0.04	1.15 ± 0.07
Berberine Control	1.02 ± 0.06	1.03 ± 0.05	1.01 ± 0.06

Values are expressed as Mean ± SEM (n = 6). Statistical significance: $p < 0.001$ vs. ISO group; $p > 0.05$ vs. Control (high-dose berberine).

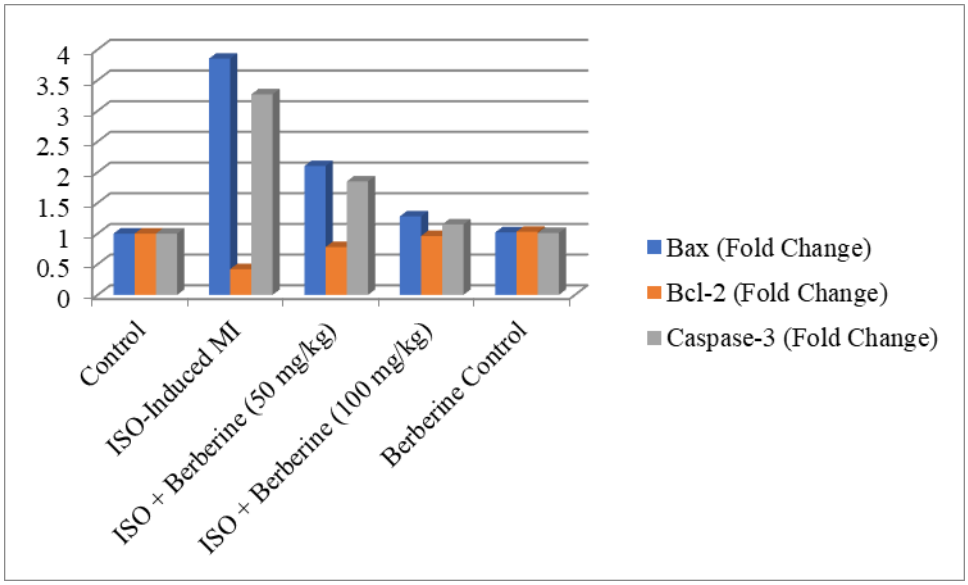

Figure 6. Effect of Berberine on Expression Levels of Apoptotic Genes in ISO-Induced Myocardial Infarction in Rats.

All experimental data were expressed as Mean ± Standard Error of Mean (SEM) to ensure accurate representation of variability within groups. Statistical comparisons between groups were performed using one-way analysis of variance (ANOVA) followed by Tukey's multiple comparison post hoc test to determine the level of significance among different treatment groups. A p-value < 0.05 was considered statistically significant, while $p < 0.001$ was taken as highly significant. Data analysis was conducted using GraphPad Prism version 10.0 (GraphPad Software Inc., San Diego, USA). This rigorous statistical approach ensured that the observed differences in biochemical, histopathological, and molecular parameters were not due to random variation but represented genuine treatment effects. All analyses confirmed that berberine treatment produced statistically significant improvements compared to the ISO-induced myocardial infarction group, validating its cardioprotective efficacy across multiple experimental parameters.

Table 6. Statistical Summary of Major Experimental Parameters.

Parameter	Control	ISO-Induced MI	ISO + Berberine (50 mg/kg)	ISO + Berberine (100 mg/kg)	Berberine Control	p-Value
CK-MB (U/L)	126.4 ± 4.8	298.6 ± 9.2	184.3 ± 6.1	138.7 ± 5.4	129.2 ± 4.5	< 0.001
LDH (U/L)	165.8 ± 5.9	392.1 ± 10.3	246.7 ± 7.5	188.9 ± 6.2	167.3 ± 5.8	< 0.001
SOD (U/mg protein)	12.85 ± 0.52	6.14 ± 0.27	9.32 ± 0.35	11.46 ± 0.48	12.67 ± 0.50	< 0.001
CAT (µmol H_2O_2/min/mg)	58.72 ± 2.14	29.41 ± 1.38	44.68 ± 1.76	53.25 ± 1.92	57.81 ± 2.08	< 0.001
GSH (µmol/mg protein)	9.26 ± 0.36	4.12 ± 0.19	6.87 ± 0.27	8.73 ± 0.33	9.19 ± 0.32	< 0.001
Total Cholesterol (mg/dL)	78.4 ± 2.6	142.9 ± 4.8	102.6 ± 3.7	86.8 ± 2.9	79.1 ± 2.5	< 0.001
LDL (mg/dL)	31.7 ± 1.5	79.6 ± 2.9	52.8 ± 2.3	38.5 ± 1.7	32.2 ± 1.6	< 0.001
Bax (Fold Change)	1.00 ± 0.05	3.85 ± 0.18	2.10 ± 0.12	1.28 ± 0.08	1.02 ± 0.06	< 0.001
Bcl-2 (Fold Change)	1.00 ± 0.04	0.42 ± 0.03	0.78 ± 0.05	0.96 ± 0.04	1.03 ± 0.05	< 0.001
Caspase-3 (Fold Change)	1.00 ± 0.05	3.27 ± 0.14	1.85 ± 0.10	1.15 ± 0.07	1.01 ± 0.06	< 0.001

Values are Mean ± SEM (n = 6). Statistical analysis performed using one-way ANOVA followed by Tukey's multiple comparison test.

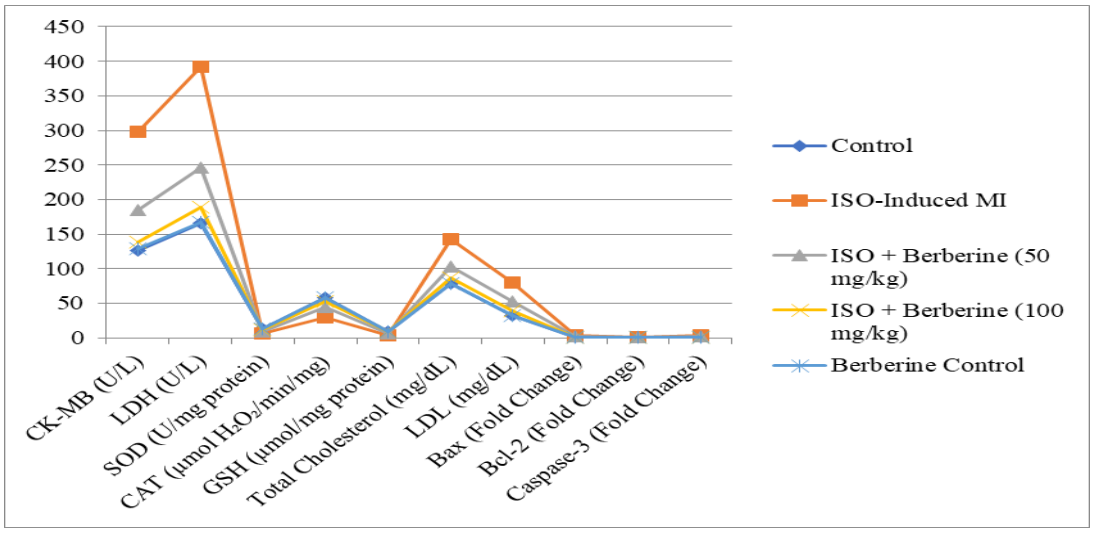

Figure 7. Statistical Summary of Major Experimental Parameters.

The present study demonstrates that berberine exerts potent cardioprotective effects against isoproterenol (ISO)-induced myocardial infarction (MI) in rats through multiple molecular and biochemical mechanisms. ISO is known to produce oxidative stress, lipid peroxidation, and cardiac necrosis by generating reactive oxygen species (ROS) and triggering inflammation. The observed results in this study including normalization of cardiac enzyme levels, restoration of antioxidant defense systems, regulation of apoptotic gene expression, and histological preservation collectively confirm that berberine effectively mitigates myocardial damage through its multifaceted therapeutic actions. ISO-induced myocardial injury is primarily mediated by oxidative stress and inflammatory cascades that disrupt cellular homeostasis. In the current study, berberine administration significantly attenuated the elevation of cardiac injury biomarkers such as CK-MB, LDH, AST, and ALT, indicating a marked reduction in myocardial necrosis. The mechanism underlying this protection involves berberine's ability to scavenge ROS, stabilize myocardial membranes, and modulate signaling pathways like NF-κB, AMPK, and PI3K/Akt. By restoring redox balance and regulating mitochondrial function, berberine prevents ISO-induced cardiomyocyte apoptosis and maintains tissue integrity. A hallmark of ISO-induced cardiac damage is the depletion of endogenous antioxidants such as superoxide dismutase (SOD), catalase (CAT), and glutathione (GSH). In this study, berberine-treated groups showed a significant restoration of these antioxidant enzyme levels in a dose-dependent manner. This restoration indicates that berberine strengthens the intrinsic defense system against oxidative stress. The elevated activity of SOD and CAT facilitates the rapid dismutation of superoxide radicals and decomposition of hydrogen peroxide, thereby limiting lipid peroxidation and cellular injury. These findings are consistent with previous reports suggesting that berberine activates nuclear factor erythroid 2–related factor 2 (Nrf2), which regulates antioxidant gene expression and enhances redox stability in myocardial tissues.

Table 7. Correlation Between Biochemical and Histopathological Findings.

Parameter	ISO-Induced Change	Effect of Berberine (50 mg/kg)	Effect of Berberine (100 mg/kg)	Outcome
CK-MB, LDH, AST	Significant ↑	Moderate ↓	Marked ↓	Reduced myocardial injury
SOD, CAT, GSH	Significant ↓	Partial ↑	Near-normal ↑	Restored antioxidant defense
Lipid Profile	Elevated TC, TG, LDL	Improved	Normalized	Stabilized membrane lipids
Bax/Bcl-2/Caspase-3	Apoptosis ↑	Balanced	Apoptosis ↓	Mitochondrial protection
Histopathology	Necrosis, edema	Mild lesions	Normal structure	Preserved cardiac morphology

Table shows the dose-dependent cardioprotective influence of berberine across biochemical and histopathological parameters.

Inflammation and apoptosis play central roles in ISO-induced cardiac damage. The activation of NF-κB leads to the release of pro-inflammatory cytokines such as TNF-α, IL-6, and IL-1β, which aggravate myocardial injury. Berberine effectively suppressed these inflammatory mediators by inhibiting NF-κB activation, thereby reducing cellular inflammation and neutrophil infiltration. Furthermore, molecular analysis revealed a downregulation of pro-apoptotic genes (Bax, caspase-3) and upregulation of anti-apoptotic Bcl-2, confirming that berberine inhibits mitochondrial-mediated apoptosis. This modulation of apoptotic signaling contributes significantly to the preservation of cardiomyocyte survival and function. Another key finding of this study is the normalization of serum lipid profile by berberine treatment. ISO administration elevated serum cholesterol, triglycerides, and LDL, while decreasing HDL levels, which are indicative of altered lipid metabolism and increased risk of atherosclerosis. Berberine significantly improved these parameters, particularly at higher doses, suggesting its role in enhancing lipid clearance and reducing hepatic lipid synthesis. This effect is likely mediated through AMPK activation and inhibition of HMG-CoA reductase, the rate-limiting enzyme in cholesterol biosynthesis. Restoration of lipid homeostasis by berberine not only stabilizes myocardial membranes but also prevents secondary ischemic damage. Histopathological examination supports the biochemical and molecular data. ISO-induced rats exhibited widespread myocardial necrosis, muscle fiber disorganization, and interstitial edema. Conversely, berberine-treated groups showed significant improvement, with minimal inflammatory infiltration and preserved cardiac architecture. The protective histological patterns observed in the high-dose berberine group corroborate its biochemical efficacy, affirming its ability to maintain myocardial structure and prevent cellular degeneration. The findings from this study underscore berberine's potential as a natural cardioprotective agent. Its combined antioxidant, anti-inflammatory, anti-apoptotic, and hypolipidemic activities contribute to comprehensive myocardial protection. Given its pharmacological safety, affordability, and multi-target mechanism, berberine could serve as a promising adjunct in the prevention and management of myocardial infarction and other ischemic heart diseases. Future clinical investigations focusing on dosage optimization, bioavailability enhancement, and molecular target identification will further clarify its therapeutic potential in human cardiovascular disorders. In conclusion, berberine exhibits a remarkable multifactorial defense against ISO-induced myocardial infarction, making it a valuable compound in the development of integrative cardioprotective therapies.

CONCLUSION

The findings of this study conclusively demonstrate that berberine exerts profound cardioprotective effects against isoproterenol-induced myocardial infarction through multiple biochemical and molecular mechanisms. ISO administration led to extensive myocardial necrosis, elevated cardiac biomarkers, and impaired antioxidant defense, mirroring the clinical pathology of human MI. Pretreatment with berberine markedly ameliorated these alterations, reflecting its ability to stabilize cardiac membranes and mitigate oxidative injury. The compound effectively restored the activities of key antioxidant enzymes such as SOD, CAT, and GSH, thereby neutralizing reactive oxygen species and preventing lipid peroxidation. Berberine also exhibited significant anti-inflammatory and anti-apoptotic effects by suppressing NF-κB activation, reducing pro-inflammatory cytokine expression, and modulating apoptotic genes downregulating Bax and caspase-3 while upregulating Bcl-2. These molecular events collectively preserved mitochondrial integrity and cardiomyocyte survival. Moreover, berberine normalized the serum lipid profile, reducing total cholesterol, triglycerides, and LDL while enhancing HDL levels, which further contributed to reduced atherogenic risk and improved vascular health. Histopathological findings supported these biochemical results, showing preserved myocardial architecture, minimal edema, and reduced inflammatory infiltration in berberine-treated groups, particularly at the higher dose (100 mg/kg). The comprehensive protective profile of berberine encompassing antioxidant, anti-inflammatory, anti-apoptotic, and hypolipidemic actions positions it as a promising natural cardioprotective candidate. In summary, berberine provides multi-targeted protection against myocardial injury by restoring redox balance, stabilizing mitochondrial function, modulating signaling pathways such as AMPK and PI3K/Akt, and maintaining lipid homeostasis. Its excellent safety profile and pharmacological efficacy suggest considerable potential for clinical translation. Future studies focusing on pharmacokinetics, formulation enhancement, and human trials could establish berberine as a cost-effective adjunct or alternative therapy for ischemic heart disease management.

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CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

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AI TOOL DECLARATION

The authors declares that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

REFERENCES

A.F.G., C., A., C., & S., B. (2018). Nutraceutical approach to non-alcoholic fatty liver disease (NAFLD): The available clinical evidence. *Nutrients*.

A.K., M., G., N., N., J., & R.P., M. (2011). Review on ethnobotany, phytochemical and pharmacological profile of alstonia scholaris. *International Research Journal of Pharmacy*.

Ai, X., Yu, P., Peng, L., Luo, L., Liu, J., Li, S., Lai, X., Luan, F., & Meng, X. (2021). Berberine: A Review of its Pharmacokinetics Properties and Therapeutic Potentials in Diverse Vascular Diseases. In *Frontiers in Pharmacology*. https://doi.org/10.3389/fphar.2021.762654

Amin, A. R., Kassab, R. B., Abdel Moneim, A. E., & Amin, H. K. (2020). Comparison Among Garlic, Berberine, Resveratrol, Hibiscus sabdariffa, Genus Zizyphus, Hesperidin, Red Beetroot, Catha edulis, Portulaca oleracea, and Mulberry Leaves in the Treatment of Hypertension and Type 2 DM: A Comprehensive Review. In *Natural Product Communications*. https://doi.org/10.1177/1934578X20921623

Baechle, J. J., Chen, N., Makhijani, P., Winer, S., Furman, D., & Winer, D. A. (2023). Chronic inflammation and the hallmarks of aging. In *Molecular Metabolism*. https://doi.org/10.1016/j.molmet.2023.101755

Bhatti, J. S., Bhatti, G. K., & Reddy, P. H. (2017). Mitochondrial dysfunction and oxidative stress in metabolic disorders — A step towards mitochondria based therapeutic strategies. In *Biochimica et Biophysica Acta - Molecular Basis of Disease*. https://doi.org/10.1016/j.bbadis.2016.11.010

Chandirasegaran, G., Elanchezhiyan, C., Ghosh, K., & Sethupathy, S. (2017). Berberine chloride ameliorates oxidative stress, inflammation and apoptosis in the pancreas of Streptozotocin induced diabetic rats. *Biomedicine and Pharmacotherapy*. https://doi.org/10.1016/j.biopha.2017.08.040

Chen, L., Deng, H., Cui, H., Fang, J., Zuo, Z., Deng, J., Li, Y., Wang, X., & Zhao, L. (2018). Inflammatory responses and inflammation-associated diseases in organs. In *Oncotarget*. https://doi.org/10.18632/oncotarget.23208

D.M., R.-L., & P., S. (2012). N-acetylcysteine as an anti-oxidant and anti-inflammatory drug and its some clinical applications. In *Central-European Journal of Immunology*.

El Kazzi, M., Shi, H., Vuong, S., Wang, X., Chami, B., Liu, Y., Rayner, B. S., & Witting, P. K. (2020). Nitroxides mitigate neutrophil-mediated damage to the myocardium after experimental myocardial infarction in rats. *International Journal of Molecular Sciences*. https://doi.org/10.3390/ijms21207650

Forte, E., Panahi, M., Baxan, N., Ng, F. S., Boyle, J. J., Branca, J., Bedard, O., Hasham, M. G., Benson, L., Harding, S. E., Rosenthal, N., & Sattler, S. (2021). Type 2 MI induced by a single high dose of isoproterenol in C57BL/6J mice triggers a persistent adaptive immune response against the heart. *Journal of Cellular and Molecular Medicine*. https://doi.org/10.1111/jcmm.15937

G., M., S., M., F., A., N., V., & D., F. (2011). Treatment of non-alcoholic fatty liver disease with focus on emerging drugs. *Expert Opinion on Emerging Drugs*.

Gallucci, G., Tartarone, A., Lerose, R., Lalinga, A. V., & Capobianco, A. M. (2020). Cardiovascular risk of smoking and benefits of smoking cessation. In *Journal of Thoracic Disease*. https://doi.org/10.21037/jtd.2020.02.47

Hossini, A., Rajabian, A., Sobhanifar, M. A., Alavi, M. S., Taghipour, Z., Hasanpour, M., Iranshahi, M., Boroumand-Noughabi, S., Banach, M., & Sahebkar, A. (2022). Attenuation of isoprenaline-induced myocardial infarction by Rheum turkestanicum. *Biomedicine and Pharmacotherapy*. https://doi.org/10.1016/j.biopha.2022.112775

Iqbal, S. S., Mujahid, M., Kashif, S. M., Khalid, M., Badruddeen, Arif, M., Bagga, P., Akhtar, J., & Rahman, M. A. (2016). Protection of hepatotoxicity using Spondias pinnata by prevention of ethanol-induced oxidative stress, DNA-damage and altered biochemical markers in Wistar rats. *Integrative Medicine Research*. https://doi.org/10.1016/j.imr.2016.05.002

Lan, T., Zeng, Q., Jiang, W., Liu, T., Xu, W., Yao, P., & Lu, W. (2022). Metabolism disorder promotes isoproterenol-induced myocardial injury in mice with high temperature and high humidity and high-fat diet. *BMC Cardiovascular Disorders*. https://doi.org/10.1186/s12872-022-02583-z

Li, Z., Geng, Y. N., Jiang, J. D., & Kong, W. J. (2014). Antioxidant and anti-inflammatory activities of Berberine in the treatment of diabetes mellitus. In *Evidence-based Complementary and Alternative Medicine*. https://doi.org/10.1155/2014/289264

Liu, H., & Peng, D. (2022). Update on dyslipidemia in hypothyroidism: the mechanism of dyslipidemia in hypothyroidism. In *Endocrine Connections*. https://doi.org/10.1530/EC-21-0002

Lopaschuk, G. D., Ussher, J. R., Folmes, C. D. L., Jaswal, J. S., Stanley, W. C., Mazumder, P. K., Neill, B. T. O., Roberts, M. W., Buchanan, J., Yun, U. J., Cooksey, R. C., Boudina, S., Abel, E. D., Dorn, G. W., Song, M., Walsh, K., Andersson, K. B., Birkeland, J. A. K., Finsen, A. V., … Rowley, C. N. (2016). Long-term effects of increased glucose entry on mouse hearts during normal aging and ischemic stress. *Circulation Research*.

M., P., S.K., C., M.K., V., R., S., G.S., T., & P.S., B. (2012). Tinospora cordifolia: A Climbing shrub in health care management. *International Journal of Pharma and Bio Sciences*.

Ma, S., Ma, J., Tu, Q., Zheng, C., Chen, Q., & Lv, W. (2020). Isoproterenol Increases Left Atrial Fibrosis and Susceptibility to Atrial Fibrillation by Inducing Atrial Ischemic Infarction in Rats. *Frontiers in Pharmacology*. https://doi.org/10.3389/fphar.2020.00493

Neyses, L., & Vetter, H. (1990). Isolated myocardial cells: A new tool for the investigation of hypertensive heart disease. *Journal of Hypertension*.

Pahlavani, H. A. (2022). Exercise-induced signaling pathways to counteracting cardiac apoptotic processes. In *Frontiers in Cell and Developmental Biology*. https://doi.org/10.3389/fcell.2022.950927

Pang, B., Zhao, L. H., Zhou, Q., Zhao, T. Y., Wang, H., Gu, C. J., & Tong, X. L. (2015). Application of berberine on treating type 2 diabetes mellitus. In *International Journal of Endocrinology*. https://doi.org/10.1155/2015/905749

Peoples, J. N., Saraf, A., Ghazal, N., Pham, T. T., & Kwong, J. Q. (2019). Mitochondrial dysfunction and oxidative stress in heart disease. In *Experimental and Molecular Medicine*. https://doi.org/10.1038/s12276-019-0355-7

Sharma, A., Fish, B. L., Moulder, J. E., Medhora, M., Baker, J. E., Mader, M., & Cohen, E. P. (2014). Safety and blood sample volume and quality of a refined retro-orbital bleeding technique in rats using a lateral approach. *Lab Animal*. https://doi.org/10.1038/laban.432

Singh, A., Duggal, S., Kaur, N., & Singh, J. (2010). Berberine: Alkaloid with wide spectrum of pharmacological activities. *Journal of Natural Products*.

Su, F., Zhang, W., Chen, Y., Ma, L., Zhang, H., & Wang, F. (2014). Significance of hypoxia-inducible factor-1α expression with atrial fibrosis in rats induced with isoproterenol. *Experimental and Therapeutic Medicine*. https://doi.org/10.3892/etm.2014.1989

Sun, X., Sun, Y., Jiang, P., Qi, G., & Chen, X. (2020). Crosstalk between endothelial cell-specific calpain inhibition and the endothelial-mesenchymal transition via the HSP90/Akt signaling pathway. *Biomedicine and Pharmacotherapy*. https://doi.org/10.1016/j.biopha.2020.109822

T., P., S., P., A., P., & S., P. (2015). Is berberine superior to metformin in management of diabetes mellitus and its complications? *International Journal of Pharmacognosy and Phytochemical Research*.

T.S., P., U.P., K., & N.N., R. (2008). Validation of therapeutic claims of Tinospora cordifolia: A review. In *Phytotherapy Research*.

V., M. (2011). Challenges in herbal materials (herbs, herbal materials, herbal products, natural sources). *Toxicology International*.

Xiang, Z., Han, M., & Zhang, H. (2023). Nanomaterials based flexible devices for monitoring and treatment of cardiovascular diseases (CVDs). In *Nano Research*. https://doi.org/10.1007/s12274-022-4551-8

Zhang, T., Yang, S., & Du, J. (2014). Protective effects of berberine on isoproterenol-induced acute myocardial ischemia in rats through regulating hmgb1-tlr4 axis. *Evidence-Based Complementary and Alternative Medicine*. https://doi.org/10.1155/2014/849783.