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Original Article

Cardioprotective Role of Flavokawain A against Isoproterenol-Induced Acute Myocardial Infarction in Rats via Modulation of NF-KB/HO-1/NQO-1 Pathways

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ABSTRACT

Background: Myocardial Infarction (MI) is a prevalent condition of heart disease. Despite remarkable growth in the treatment of heart diseases, MI remains the foremost cause of mortality worldwide and a significant pathological concern. **Objectives:** The primary aim of this work is to analyze the therapeutic activities of flavokawain A on Isoproterenol (ISO)-induced MI in rats. Materials and Methods: The rats were administered with ISO (85 mg/kg) in order to induce MI and pretreated with flavokawain A. The heart and body weights of all rats were measured. An analysis was conducted on the levels of uric acid, CRP and total protein. The cardiac function marker enzymes and antioxidant levels were studied using kits. The Na^+/K^+, Mg^{2+} activities and Ca^{2+} ATPase, Na^+, K^+ and Ca^{2+} ions, as well as the inflammatory markers and NF-κB and HO-1/NQO-1 protein levels, were examined using corresponding kits. Heart samples were subjected to histopathological examination to identify histological alterations. **Results:** The findings demonstrated that flavokawain A treatment led to an elevation in body weight and a diminution in heart weight in rats with MI. The uric acid and CRP levels were reduced, while the total protein levels are elevated in the flavokawain A-treated rats. The serum levels of cardiac marker enzymes were significantly reduced, but these levels in the cardiac tissues were elevated in MI rats treated with flavokawain A. In addition, flavokawain A enhanced antioxidant levels, reduced inflammatory cytokines and controlled the Na^+, K^+ and Ca^{2+} ion levels. Flavokawain A treatment in MI rats considerably enhanced the Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase enzyme activities in their cardiac tissues. Flavokawain A treatment also improved the histological abnormalities in the cardiac tissues. **Conclusion:** The treatment of flavokawain A significantly enhanced the levels of antioxidants and the Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase enzymes. Additionally, it effectively diminished the inflammatory marker levels and controlled cardiac enzyme activities. Therefore, it has the potential to be an effective treatment option in the future for treating MI.

Keywords: Myocardial infarction, Flavokawain A, Inflammation, Ca^{2+} ATPase, Antioxidants.

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INTRODUCTION

Cardiovascular Disease (CVD) is the major cause of deaths around the world, surpassing other diseases such as cancer. CVD encompasses conditions affecting the heart and blood vessels and continues to be a significant contributor to global mortality. In 2021, the World Health Organisation reported that CVD was responsible for 32% of global mortality. Among these conditions, Myocardial Infarction (MI) was associated with 85% of the deaths. MI is a significant contributor to both disease and mortality worldwide among the many CVDs.[1] Blockage in the coronary artery results in inadequate blood flow to the heart, leading to the death of the heart muscle and finally producing ischemic tissue necrosis along with other pathological and anatomical alterations. The development of MI involves the occurrence of hyperlipidemia, oxidative stress and the process of peroxidation of lipids in the cell membranes. The prevalence of MI is steadily growing each year.[2]

Catecholamines play a critical role in maintaining cardiac contractility and metabolism, particularly at low concentrations. However, an excessive amount of naturally occurring or externally administered catecholamines can lead to significant stress in the heart muscle, disrupted energy metabolism and both biochemical

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and structural alterations that ultimately culminate in necrosis of cardiac tissue, resembling an infarction. Therefore, the use of the β-adrenoceptor agonist Isoproterenol (ISO) in rodents allows for a quick and non-invasive approach to replicate the clinical manifestations of human MI.[3] The etiology of ISO-induced MI is intricate and multifaceted, primarily involving the production of harmful ROS in cardiomyocytes, leading to oxidative stress. As a result, there is a gradual deterioration of the mitochondria and an increase in the buildup of Ca^{2+} inside the cells. Additionally, oxidative stress is participated in the onset of cardiac remodeling and fibrosis.[4] Moreover, MI induces an inflammatory reaction that amplifies damage to the heart muscle in the initial stage. This reaction is marked by the over release of inflammatory markers and the migration of immune cells into the area deprived of blood flow. The presence of ROS activates these activities, which in turn promote signal transmission and regulate the expression of certain transcription molecules, such as NF-κB.[5]

MI arises when blood flow to a specific area of the heart is suddenly blocked, leading to reduced oxygen supply and the eventual death of the affected heart tissues. Cardiac muscle atrophy greatly diminishes the heart's contractile force and ultimately results in heart failure.[6] Despite the significant growth in the treatment of CVD, effectively treating MI and associated CVD is still difficult. Therefore, there is a pressing need for new medicines to enhance the therapeutic efficacy of these conditions.[7] Flavokawain A is a bioactive chalcone compound found naturally in the root of the *Piper methysticum*. It has been shown that flavokawain A has various pharmacological properties such as anticancer,[8] anti-sepsis,[9] anti-inflammatory,[10] anti-fibrotic and antioxidant[11] and antiarthritic[12] properties. However, its therapeutic roles against MI have not been reported yet. Hence, the current work is aimed at addressing the therapeutic importance of flavokawain A against ISO-induced MI in rats.

MATERIALS AND METHODS

Chemicals

The major chemicals and compounds utilized in this study such as Flavokawain A, ISO and others were acquired from Sigma-Aldrich, USA. All the biochemical parameters were assayed using ELISA kits (Abcam, USA and Elabscience, USA, respectively).

Experimental rats

The current study used healthy Wistar rats, which were obtained from an institutional animal house. Prior to initiating the assays, the animals were acclimated to the lab environment for 7 days and housed in infection-free enclosures. The circumstances were maintained at temperature 26±5°C, air moisture 55±5% and a light/dark cycle of 12/12 hr.

Treatment methods

The one-week acclimated rats were alienated into four groups. Group-I consisted of control rats that were administered with 0.1% NaCl. Group-II rats received ISO (85 mg/kg; i.p.) on the 19th and 20th day to initiate MI. The group-III and -IV received oral pre- and co-treatment of 25 and 50 mg/kg of Flavokawain A for 20 days with ISO on the 19th and 20th days. Following the conclusion of the treatments, rats were sacrificed and blood was then collected to prepare the serum. The entire cardiac tissues were removed and washed with saline solution. The removed heart was weighed to ascertain any changes in the weight. The heart tissues were subjected to histological studies and remaining tissue portions were used to prepare homogenates and subsequent biochemical examinations.

Analysis of uric acid, CRP and total proteins

The CRP, uric acid and total protein levels in the serum of experimental animals were investigated using an assay kit as per the guidelines given by the manufacturer (Abcam, USA).

Analysis of cardiac marker enzymes

The ALT, AST, GGT and CK activities were assessed in both serum and cardiac tissue homogenates of the experimental rats using commercially obtained assay kits (Elabscience, USA). The experiments were done as per the manufacturer's procedure specifications.

Analysis of antioxidant levels

The concentrations of SOD, CAT and GSH in the cardiac tissue homogenates of the experimental rats were examined using assay kits (Elabscience, USA). The assays were done as per the manufacturer's guidelines.

Analysis of Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities and Na^+, K^+ and Ca^{2+} ion levels

The Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activity in the cardiac tissue homogenates of experimental rats were analyzed using the previous methods.[13-15] The concentrations of K^+, Na^+ and Ca^{2+} ions in the cardiac tissue homogenates of the experimental rats were evaluated using commercially available kits (Abcam, USA).

Analysis of inflammatory cytokines and oxidative marker protein levels

An assay kits were utilized to assess the inflammatory cytokines, such as TNF-α, IL-6 and IL-1β in both cardiac tissue homogenates and serum of the experimental rats. The manufacturer's guidelines suggestions from Abcam, USA, were followed to conduct the assays in triplicate. The concentrations of NF-κB, HO-1 and NQO-1 proteins in the heart tissue homogenates were assessed using respective assay kits. The experiments were conducted in triplicate following the manufacturer's guidelines (Abcam).

Histopathological analysis

The heart obtained from experimental animals were treated with a 10% formalin and subsequently, they were dehydrated by adding ethanol in increasing concentrations. Subsequently, the cardiac tissues are subjected to paraffinization and sliced into 6 µm diameter. The sections were then stained with eosin and hematoxylin. The stained slides were then inspected using microscope using a 40× objective lens.

Statistical analysis

The data are presented as mean±SD of triplicate assays. The data were analyzed using a commercially accessible GraphPad Prism software. The differences in the means were assessed using a one-way ANOVA and Tukey's *post hoc* assay. A significance at $p<0.05$ was fixed to compare between groups.

RESULTS

Effect of flavokawain A on heart and body weights of experimental rats

The rats exhibited a progressive decrease in the body weight while an increase in their heart weight as a consequence of ISO-induced MI. Contrastingly, the 25 and 50 mg/kg of flavokawain A treatment resulted in a significant elevation in their body weight and decrease in their heart weight (Figure 1).

Effect of flavokawain A on the uric acid, CRP and total protein levels

Figure 2 demonstrates a significant increase in uric acid and CRP while reduced the total protein levels in the serum of the MI rats, compared with control. Interestingly, the flavokawain A treatment significantly regulated these changes. Treatment of flavokawain A (25 and 50 mg/kg) successfully diminished the uric acid and CRP levels as well as increased the total protein levels in their serum.

Effect of flavokawain A on the cardiac enzymes in the heart and serum samples

Figure 3 reveals a significant increase in the CK, ALT, AST and GGT levels in the serum of the MI rats than the control. Furthermore, these marker enzymes were considerably reduced in the cardiac tissue homogenates of the MI rats. Whereas, the treatment of flavokawain A at the concentrations of the 25 and 50 mg/kg led to a considerable decrease in the ALT, CK, AST and GGT levels in their serum. The flavokawain A treatment also remarkably elevated these enzyme activities in the heart tissues (Figure 3).

Effect of Flavokawain A on antioxidants in the cardiac tissues

The occurrence of oxidative stress in the rats with MI was validated by assessing the levels of cardiac antioxidant levels. The findings of this analysis are presented in Figure 4. An ISO-induced rats exhibited a drastic decrease in the CAT, SOD and GSH levels in their cardiac tissue tissues, when compared to control. Contrastingly, the flavokawain A at a dosage of 25 and 50 mg/kg-treated rats demonstrated a stable elevation in the CAT, SOD and GSH levels, as shown in Figure 4. These findings were are evidenced the antioxidant potentials of the flavokawain A.

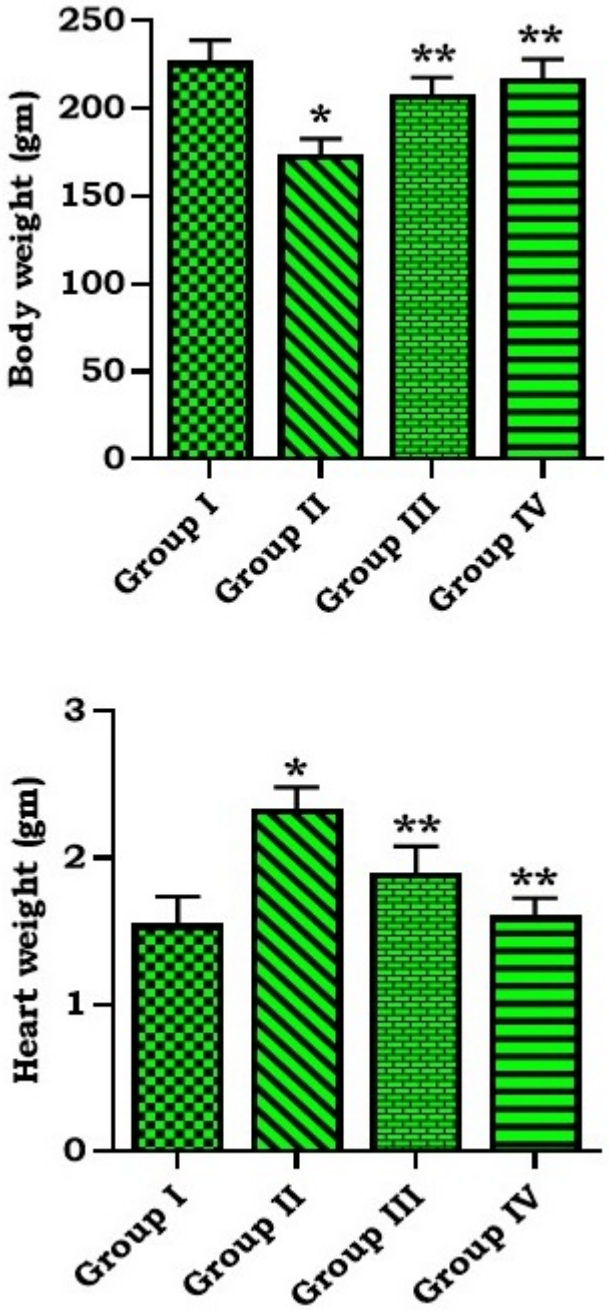

Figure 1: Effect of flavokawain A on the heart and body weights of the experimental rats. The data are presented as the mean±SD of the triplicate assays. *$p<0.01$ compared to the control group and **$p<0.05$ compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups.

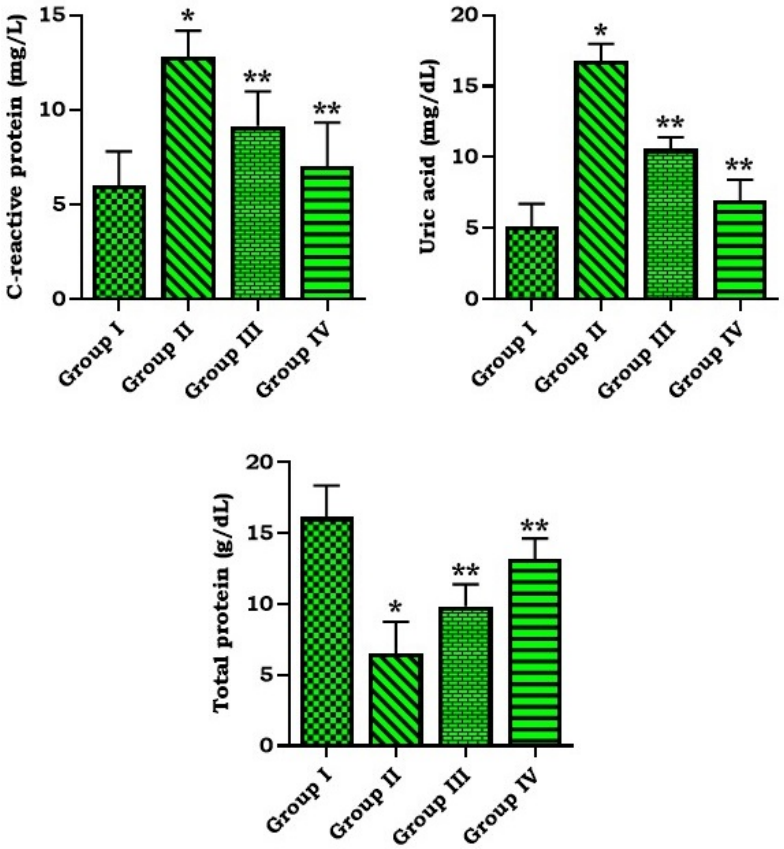

Figure 2: Effect of flavokawain A on the CRP, uric acid and total protein levels in the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups.

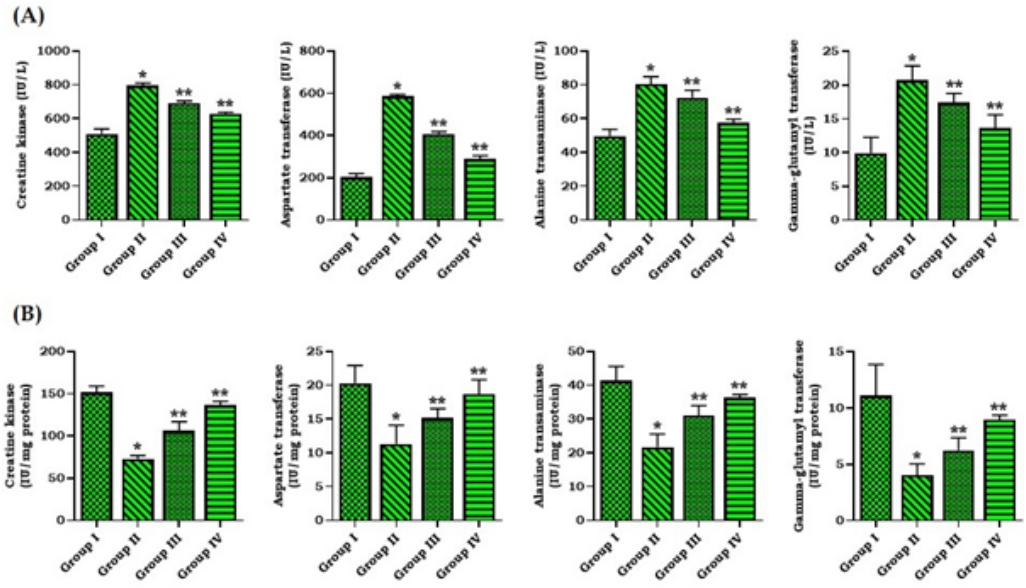

Figure 3: Effect of flavokawain A on the cardiac marker enzyme activities in the serum and cardiac tissues of the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups. (A): Cardiac marker enzymes in the serum; (B): Cardiac marker enzymes in the heart tissue homogenates.

Effect of flavokawain A on the Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities and Na^+, K^+ and Ca^{2+} ion levels in the cardiac tissues

Figure 5 illustrates that the Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities were significantly increased in the cardiac tissues of the MI rats, in contrast to the control group. In contrast, the flavokawain A treatment exhibited a remarkable increase in these ATPases activities, returning them to levels close to normal. Furthermore, a considerable elevation in the Na^+ and Ca^{2+} ion levels and a reduction in the K^+ ion levels were noted in the cardiac tissues, as compared to the control group. Whereas, the MI rats that were treated with the flavokawain A at a 25 and 50 mg/kg concentrations exhibited a noteworthy reduction in the Na^+ and Ca^{2+} ion levels and increase in the K^+ ions (Figure 5).

Effect of flavokawain A on the inflammatory cytokine levels

A significant rise in both serum cardiac tissue levels of TNF-α, IL-6 and IL-1β were observed in the MI rats. Whereas, the flavokawain A treatment significantly diminished these cytokine levels in both serum and heart samples of the MI rats (Figure 6). These outcomes highlighted the anti-inflammatory properties of the flavokawain A.

Effect of flavokawain A on the NF-κB/HO-1/NQO-1 signalling protein levels in the cardiac tissues

Figure 7 demonstrates that the rats induced with ISO had significantly higher NF-κB and NQO-1 levels while reduced HO-1 levels were observed in their cardiac tissues when compared to control group. Significantly, the flavokawain A treatment led to a remarkable diminution in the NF-κB and NQO-1 levels and increased the HO-1 expressions in the cardiac tissues of the MI rats.

Effect of flavokawain A on the heart tissue histopathology

The control rats exhibited the normal cardiac histo-architectures. In contrast, the ISO-induced rats demonstrated drastic histological changes, including ruptures in the heart muscle, degeneration of myocytes and moderate bleeding accompanied by myocyte necrosis. Interestingly, the treatment of flavokawain A at a dosage of 25 and 50 mg/kg effectively protected the cardiac tissues from the damages due to the MI. Flavokawain A significantly decreased the histological changes caused by ISO (Figure 8).

DISCUSSION

MI is a prevalent form of heart disease. Despite remarkable growth in the therapy of heart diseases, MI remains the foremost cause of mortality worldwide and a major global pathological problem. MI is caused by an increase in the requirement for nutrients and oxygen by the heart muscle and a decrease in the delivery of nutrients and oxygen through the coronary circulation. This leads to damage to the heart cells and is one of the most deadly consequences of cardiovascular disorders.[16] Oxidative injury is extensively recognized as a primary pathophysiological process that contributes to MI.[17] The oxidative stress arises due to the accumulation of oxidized lipoproteins, which in turn triggers the development of atherosclerosis.[18] In this stage, inflammatory cells

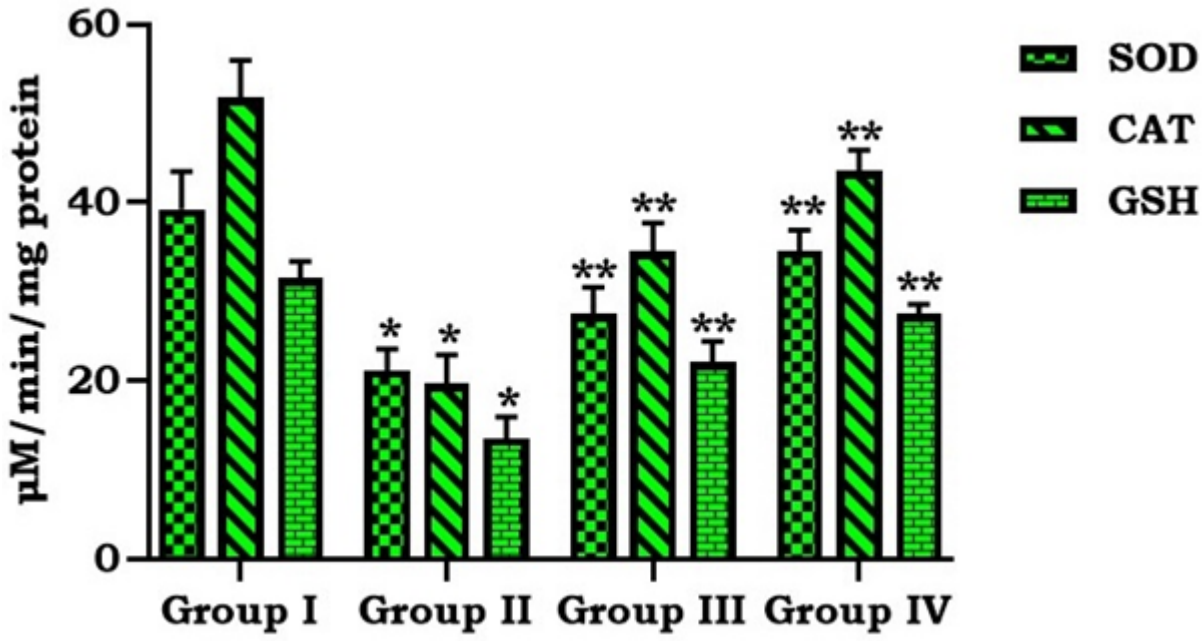

Figure 4: Effect of flavokawain A on the antioxidant levels in the cardiac tissues of the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups.

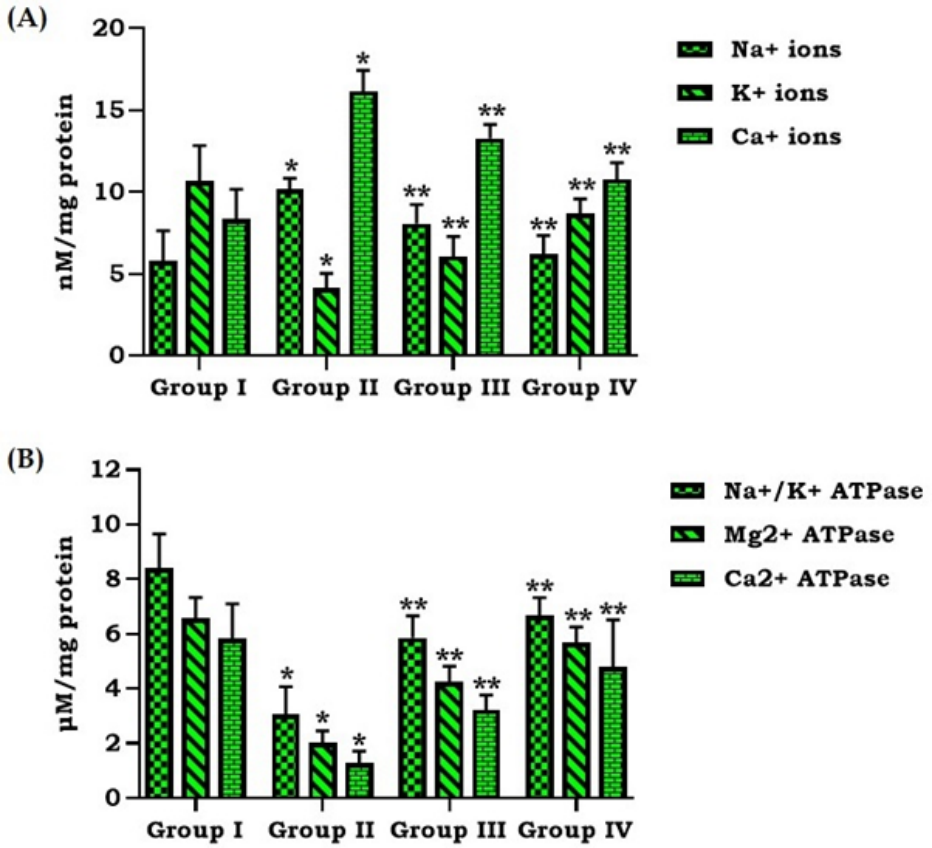

Figure 5: Effect of flavokawain A on the Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities and Na^+, K^+ and Ca^{2+} ion levels in the cardiac tissues of the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups. (A): Na^+, K^+ and Ca^{2+} ion levels; (B): Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities.

generate ILs, ROS and other chemokines via signaling pathways, which amplify the inflammation.[19] Concurrently, fibroblasts undergo proliferation and differentiation into myofibroblasts, releasing substantial levels of extracellular proteins to regulate the structural integrity of the infarction site. After MI, the cardiac tissues undergo structural remodeling at the scar site as a result of cell death in the granulation tissues.[20] Overproduction of ROS and inflammatory responses lead to metabolic stress and reduced tissue defense systems, eventually causing additional damage and death of cardiomyocytes.[21]

During heart failure, there is high ROS accumulation and subsequent reduction in antioxidant mechanisms, such as SOD, CAT and GSH and NF-κB pathway activation.[22] Anti-oxidants, such as CAT, SOD and GSH, are the initial defense mechanisms of cells against damage caused by the superoxide anion radical and H_2O_2. These antioxidants act before the production of reactive hydroxyl radicals.[23] Prior research has demonstrated that during the progression of MI, the heart had a restricted ability to work against the deleterious effects of ROS, which makes it highly vulnerable to oxidative stress. The cardiac antioxidant system plays a crucial function in neutralizing ROS.[24] Hence, it would be advantageous to provide sufficient levels of antioxidants and enhance the body's natural ability to counteract oxidative stress in order to prevent MI development.[25] Similarly, the present results show a drastic decrease in antioxidant levels. Captivatingly, the flavokawain A treatment remarkably increased the antioxidant levels in the MI rats, which highlights the potential antioxidant effects of flavokawain A.

The myocardium holds elevated levels of diagnostic biomarkers of MI; when it undergoes metabolic injury, it releases these substances into the extracellular fluids. Myocardial enzymes are the most accurate biomarkers of tissue damage among all the macromolecules that released from injured tissue due to their specificity for heart tissue and their ability to catalyze reactions.[26] Myocardial cells can be injured or destroyed when there is a lack of oxygen or glucose supply. This can cause the heart membrane to become permeable or even rupture completely, leading to the release of enzymes. The serum CK, AST and ALT activity assays are a crucial diagnostic tool due to their sensitivity and significant presence in cardiac tissue and their almost absence in other tissues. CK activity serves as a valuable indicator for promptly diagnosing MI. The release of cytosolic enzymes like CK, AST and ALT into the bloodstream can take place when cell membranes are damaged or become permeable. These enzymes are used

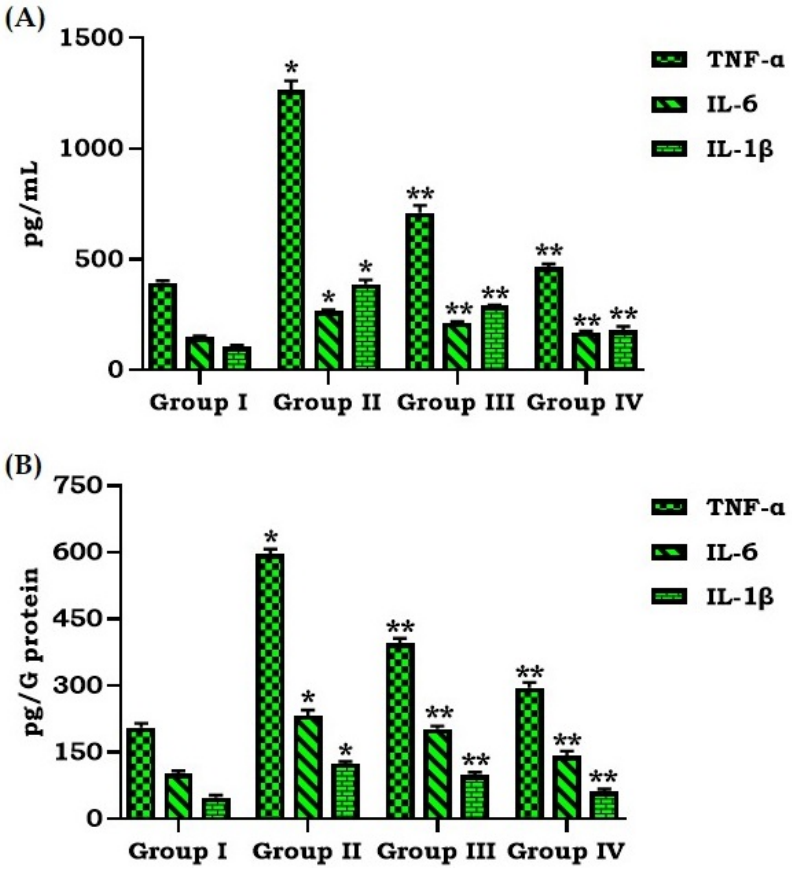

Figure 6: Effect of flavokawain A on the inflammatory cytokine levels in the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups. (A): Inflammatory cytokine levels in the serum; (B): Inflammatory cytokine levels in the heart tissues.

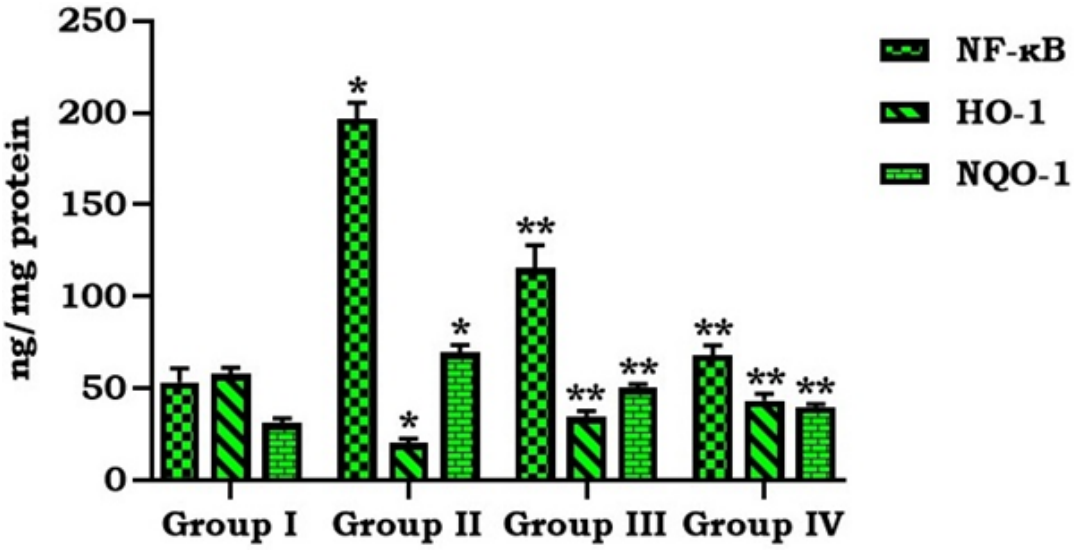

Figure 7: Effect of flavokawain A on the NF-κB/HO-1/NQO-1 signaling protein levels in the cardiac tissues of the experimental rats. The data are presented as the mean±SD of the triplicate assays. *p<0.01 compared to the control group and **p<0.05 compared to the ISO-induced MI group. The data was subjected to statistical analysis using One-Way ANOVA, followed by Tukey's *post hoc* analysis, to assess and compare the differences between the groups.

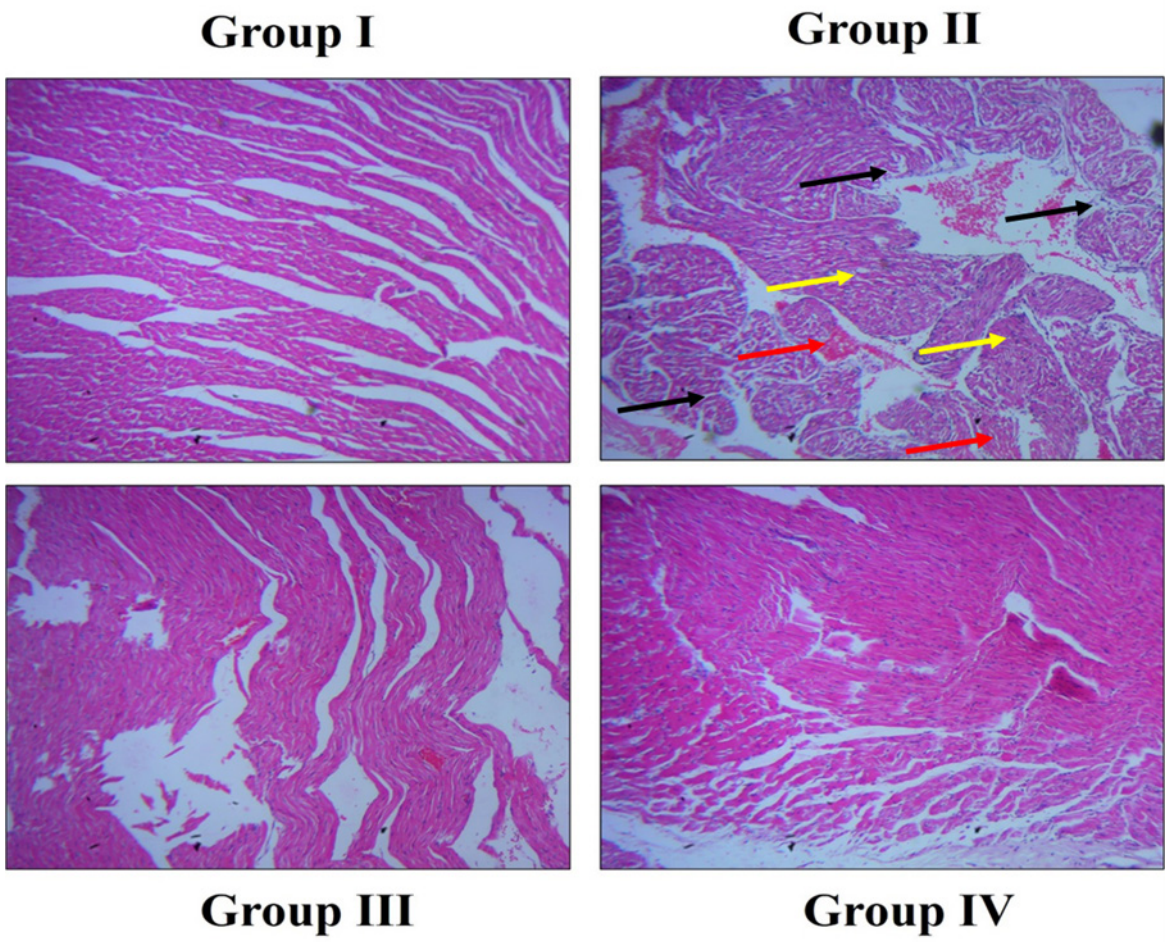

Figure 8: Effect of flavokawain A on the heart tissue histopathology of the experimental rats. Group I: Normal control group; Group II: ISO-induced MI group: Group III: ISO-induced MI+25 mg/kg of flavokawain A-treated group; Group V: ISO-induced MI+50 mg/kg of flavokawain A-treated group. Black arrows: Ruptures in the heart muscle; Yellow arrows: Degeneration of myocytes; Red arrows: Moderate bleeding accompanied by myocyte necrosis.

as diagnostic markers to identify tissue injuries. The levels of these intracellular enzymes in the blood indicate changes in the integrity of the plasma membrane.[27] Moreover, the concentration of enzymes found in the serum is said to be directly related to the quantity of dead cells, which indicates the change in the integrity of the plasma membrane.[28] The present study observed a notable increase in cardiac enzymes, such as CK, a marker for acute myocardial damage, ALT, GGT and AST, in the serum and a subsequent diminution in the heart tissues of the MI rats. This may have been caused by changes in membrane permeability and disintegration resulting from MI produced by ISO. The injury to the cardiomyocytes resulted in the release of enzymes into the bloodstream, causing an upsurge in the amount of these enzymes in the blood.[29] Notably, the flavokawain A treatment drastically diminished these enzymes in the serum while improved in the heart of the MI rats.

ATPases have a crucial function in the relaxation and contraction processes of the heart muscle by regulating the ion concentrations at normal levels within the myocytes. The ischemia state can lead to a decrease in ATPase activity, which may cause injury and necrotic alterations in the myocardial cells.[30] During ischemia conditions in cardiac cells, excessive calcium levels trigger the activation of Ca^{2+}-ATPases, leading to the depletion of high-energy phosphate reserves. This, in turn, indirectly inhibits the transport of Na^+ and K^+ ions and causes the inactivation of Na^+/K^+ ATPase. Prolonged elevation of Ca^{2+} levels can trigger cytotoxicity that leads to disruptions in cellular structure and function. Activation of phospholipase by Ca^{2+} can lead to a decline in mitochondrial activity, causing a decrease in membrane potential and the termination of ATP formation.[31] ISO binds to membrane lipids, leading to injury and inhibiting the function of membrane-associated enzymes. The decrease in ATPase activity during the ischemia condition may be accountable for inducing functional injury in the affected myocardial cells.[32] Hence, the assessment of enzyme activities associated with the membrane will reveal any changes in the membrane during pathological situations. The present results revealed that the MI rats showed decreased Na^+/K^+, Mg^{2+} and Ca^{2+} ATPase activities. Furthermore, the Na^+ and Ca^{2+} ion levels were increased and the K^+ ion levels

were reduced in the MI rats. Interestingly, the flavokawain A treatment effectively regulated these changes in the MI rats. These outcomes highlight the cardioprotective role of flavokawain A.

Cytokines, including IL-6, IL-1β and TNF-α are biomarkers of inflammation that are crucial in multiple inflammatory cascades. NF-κB is a multifactorial transcription molecule that regulates the transcription of many genes responsible for the development of MI.[33] The presence of pro-inflammatory markers and the onset of inflammation may contribute to the progression of MI. Within the cells, the presence of ROS stimulates the generation and discharge of these cytokines by cardiomyocytes. IL-6, TNF-α and IL-1β are biomarkers that can predict the early onset of organ dysfunction and trigger apoptosis in cardiomyocytes.[34] The current study demonstrated drastic elevations in these cytokine levels in the MI rats. Whereas, the flavokawain A treatment remarkably decreased these cytokine levels in the MI rats, which proves its anti-inflammatory properties.

To understand the molecular processes behind the antioxidant properties of flavokawain A, the NO-1/NQO-1 signaling proteins in the heart tissues were investigated. It has been shown that cells often counteract the harmful effects of ROS by activating the Nrf-2 gene. Conversely, poor activation of Nrf2 is linked to the progression of CVD.[35] Nrf2 regulates the expression of several genes that encode proteins with cytoprotective properties, including NQO-1 and HO-1.[36] HO-1, which is the inducible HO type, mostly provides protection to cells under inflammatory circumstances.[37] By reducing the expression of inflammatory markers, HO-1 can safeguard cells from inflammatory injury.[38] HO-1 is activated by oxidative injury and other factors generated during inflammation, most likely as a component of a cellular defense mechanism in response to stress. Furthermore, HO-1 suppressed the expression of inflammatory cytokines and provided cellular protection against inflammatory damage. Its role is to provide negative feedback to prevent excessive cell activation, thereby regulating the inflammatory response.[39] In this work, the findings exhibited that the MI rats had a decreased HO-1 level, while increased NQO-1 levels were observed. However, the flavokawain A treatment remarkably increased the HO-1 levels and reduced the NQO-1 levels in the heart tissues of the MI rats. These findings proved that flavokawain A has antioxidative effects by regulating HO-1/NQO-1 signaling in MI rats.

CONCLUSION

The present results revealed that treatment with flavokawain A can protect and mitigate the biochemical and histological damage to the cardiac tissues caused by MI. This salutary effect of flavokawain A may be due to its notable anti-inflammatory and antioxidant activities. Therefore, flavokawain A can be used as a new therapeutic intervention to treat MI. In order to enhance our understanding of the anti-inflammatory, antioxidant and cardioprotective properties of flavokawain A in countering MI, further experiments are required in the future.

ACKNOWLEDGEMENT

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ETHICAL APPROVAL

This work has approved by the institutional animal ethical committee by Xing'anmeng People's Hospital, Xing'anmeng, Inner Mongolia-137400, China.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

MI: Myocardial infarction; **ISO:** Isoproterenol; **CVD:** Cardiovascular disease; **ROS:** Reactive oxygen species; **SOD:** Superoxide dismutase; **GPx:** Glutathione peroxidase; **CAT:** Catalase.

SUMMARY

Cardiac muscle atrophy greatly diminishes the heart's contractile force and ultimately results in heart failure. Flavokawain A is a bioactive chalcone compound found naturally in the root of the *Piper methysticum*. It has been shown that flavokawain A has various pharmacological properties. flavokawain A treatment remarkably increased the HO-1 levels and reduced the NQO-1 levels in the heart tissues of the MI rats. These findings proved that flavokawain A has antioxidative effects by regulating HO-1/NQO-1 signaling in MI rats.

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