ABSTRACT: Arrhythmia is an important cause of death after myocardial infarction (MI). Different substances have been evaluated for their anti-arrhythmic effect in MI. This study was performed to evaluate the anti-arrhythmic impacts of crocin in an MI animal model (rat) by estimation of the expression of connexin 43 (Cx43). Fifty male Sprague−Dawley rats were grouped into 5 groups, each composed of 10 rats. The first group was regarded as the normal control group and the second one was considered as the MI group, which was caused by ligation of the left anterior descending artery. The other three groups received crocin 50 or 10 mg/kg/day or metoprolol 100 mg/kg/day for 1 week, following ligation of the left anterior descending artery. Evaluated outcomes were cardiac Cx43 expression, arrhythmia incidence, histological findings, and myocyte resting potential. Crocin-treated MI groups showed a significantly lower arrhythmia score than the non-treated MI group, 10 mg/kg/day (1.85 ± 0.55, p < 0.01) and 50 mg/kg/day (1.70 ± 0.33, p < 0.01). Groups that received crocin 10 mg/kg/day (66.30 ± 2.59, p < 0.01), crocin 50 mg/kg/day (68.10 ± 2.43, p < 0.01), and metoprolol 100 mg/kg/day (~63.54 ± 0.63 mV, p < 0.01) significantly prevented depolarization in comparison with the non-treated MI group. Expression of Cx43 mRNA in crocin 10 mg/kg/day (1.54 ± 0.24, p < 0.01), crocin 50 mg/kg/day (1.73 ± 0.09, p < 0.01), and metoprolol 100 mg/kg/day (1.75 ± 0.14, p < 0.01) treatment groups was significantly higher in comparison with the non-treated MI group. Crocin showed a preventive effect on the arrhythmogenic impact of MI in an experimental model of ischemic injury through an increase in expression of Cx43.

INTRODUCTION

One of the main mortality causes in the world is cardiovascular diseases (CVDs). The World Health Organization (WHO) estimated that 17.9 million people died due to CVDs in 2017, which represents 31% of all deaths worldwide, out of which 6.7 million died from stroke and 7.4 million died because of coronary heart diseases. CVDs are still a significant challenge in the age of nanotechnology, proteomics, and genomics, and researchers have attempted to develop a viable evidence-based approach for preventing CVD onset.

It has been reported that there are some compounds with biological actions that can lead to the reduction of CVDs. As an example, flavonoids can improve cardiovascular health via different mechanisms, such as antidiabetic and hypocholesterolemic activities, antioxidant effect, vasorelaxation effect, and anti-atherosclerotic characteristics. Likewise, carotenoids, like zeaxanthin, lutein, and crocin, exert biological actions that can provide various cardiovascular advantages.
One of the most well-known spices is saffron (Crocus sativus L.), which has medicinal properties. Commonly, it is utilized in European regions, including Spain, Greece, and Italy, and also in regions of Asia, like India and Iran. Traditionally, it is known to have therapeutic effects on palpitation according to Persian, Ayurvedic, and Chinese medicine.

The major constituent of saffron is crocin, which gives its red color and has various cardiovascular effects. Crocin has been reported as a suppressor of the renin-angiotensin system. Some studies have indicated that crocin has an impact on blood pressure. According to the reports, in a dose-dependent manner, crocin lessens hypertension in rat caused by deoxycorticosterone acetate (DOCA) salt. In addition, cardiovascular toxicity induced by diazinon (DZN) in rats was improved by crocin through oxidative stress reduction. Multiple studies have demonstrated that crocin has a protective effect against cardiac arrhythmias induced by reperfusion.

The development of effective and experiential diagnostic techniques for the management and treatment of cardiac arrhythmias has exceeded the speed from the last decade. The detection of main candidate genes for monogenic arrhythmia syndromes demonstrates that to bring the fundamental biology to the clinic is the potent approach. Different biological pathways are suggested to be involved in the induction of cardiac ischemic arrhythmia. For example, integrin-linked kinase induces multiple biological pathways such as glycogen synthase kinase 3β, protein kinase B, extracellular signal-regulated kinases, Rac1 pathways, and myosin light chain. It is known that deletion of the integrin-linked kinase can induce arrhythmia and cardiac death in an animal model of ischemic heart insult. A recent study supported the hypothesis that inhibition of connexin 43 (Cx43) is involved in the anti-arrhythmic effect of the integrin-linked kinase pathway. Different diagnostic modalities have been used in myocardial infarction (MI). Electrocardiography, serum markers (such as troponin, myoglobin, and creatine kinase), echocardiography, and angiography are the most popular modalities used in the diagnosis of MI. Various prognostic models are also applied for MI.

Although crocin’s cardiovascular effects have been evaluated by several research works, and anti-arrhythmic effects have been reported for it, no study has investigated the underlying mechanism. The present research aims at examining the expression of Cx43 as the possible pathway for crocin’s anti-arrhythmic effects by use of an experimental model of arrhythmia in MI infarction in rats.

**RESULTS**

**Arrhythmia Score, Interval, Frequency, and Duration.** There was no arrhythmia observed in the normal control group, which led to zero scores. The MI group showed different types of arrhythmias with an arrhythmia score of 4.2 ± 0.65 (p < 0.01 in comparison with the control group). Treated MI groups, crocin 10 mg/kg/day (1.85 ± 0.55, p < 0.01), crocin 50 mg/kg/day (1.70 ± 0.33, p < 0.01), and metoprolol 100 mg/kg/day (1.850 ± 0.74, p < 0.01), showed a significantly lower arrhythmia score than the non-treated MI group. In Bonferroni’s corrected multiple comparison test (p = 1.00), no significant differences were observed among the treated groups (Figure 1).

The RR interval was significantly longer, and the mean frequency and duration of ventricular arrhythmias were significantly lower in groups treated with crocin and metoprolol compared with the untreated MI group (p < 0.01) (Figure 2).

**Resting Membrane Potential.** The MI group showed a significantly depolarized resting membrane potential of cardiac myocytes from −83.71 ± 1.11 mV in the sham control to −66.70 ± 2.45 mV in the MI control group (p < 0.01). Groups

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**Figure 1.** Arrhythmia scores in the study groups (MI control, MI metoprolol 100 mg/kg/day, MI crocin 10 mg/kg/day, and MI crocin 50 mg/kg/day). MI: myocardial infarction.

**Figure 2.** RR intervals, mean frequency, and duration of ventricular arrhythmia in the study groups (MI control, MI metoprolol 100 mg/kg/day, MI crocin 10 mg/kg/day, and MI crocin 50 mg/kg/day). MI: myocardial infarction.
treated with crocin 10 mg/kg/day \((-76.30 \pm 2.59, p < 0.01)\), crocin 50 mg/kg/day \((-78.10 \pm 2.43, p < 0.01)\), and metoprolol 100 mg/kg/day \((-73.54 \pm 0.63 \text{ mV}, p < 0.01)\) significantly prevented this depolarization in comparison with the non-treated MI group (Figure 3).

**Histogram.** Interstitial hemorrhage and extensive edema were observed in the hearts’ anterior area in MI rats. Nevertheless, there was a significantly lower pathologic change in the rats’ hearts in the metoprolol and crocin groups.

**Western Blot.** The western blot method was used for evaluating the Cx43 protein expression. According to the results of this method, Cx43 protein was significantly lower in the infarcted zone in the MI group \((0.73 \pm 0.24, p < 0.01)\) than in the normal control \((2.12 \pm 0.09)\). In crocin 10 mg/kg/day \((1.54 \pm 0.24, p < 0.01)\), crocin 50 mg/kg/day \((1.73 \pm 0.09, p < 0.01)\), and metoprolol 100 mg/kg/day \((1.75 \pm 0.14, p < 0.01)\) groups, this reduction decrease was significantly hindered in comparison with the non-treated MI group (Figure 4).

**Real-Time PCR Analysis.** Real-time PCR (RT-PCR) analysis was used for evaluating the expression of the Cx43 mRNA, results of which indicated a lower expression of Cx43 mRNA in the infarcted zone in the MI group \((0.14 \pm 0.08, p < 0.01)\) in comparison with the normal control \((0.61 \pm 0.09)\). In crocin 10 mg/kg/day \((0.39 \pm 0.08, p < 0.01)\), crocin 50 mg/kg/day \((0.62 \pm 0.12, p < 0.01)\), and metoprolol 100 mg/kg/day \((0.49 \pm 0.11, p < 0.01)\) groups, this reduction was significantly hindered in comparison with the non-treated MI group (Figure 5).

**DISCUSSION**

As shown by the results of the experiment, there are anti-arrhythmic effects observed for crocin, similar to metoprolol, which was applied dose dependently in experimental models of MI. In this regard, reduction in Cx43 expression in MI is prevented.

As previously demonstrated, different rhythm disturbances, such as ventricular fibrillation (VF), premature ventricular contraction (PVC), and ventricular tachycardia (VT), are induced by experimental myocardial ischemia in animals. \(^{20}\) It is considered that these arrhythmias are attributed to delay following depolarizations in surviving Purkinje fibers, causing increased automaticity in the ischemic area in these fibers. \(^{21}\)

As a beta-receptor blocker, metoprolol is a member of class II anti-arrhythmic drugs. It was shown that beta-blockers are preventers of sudden cardiac death resulting from malicious ventricular arrhythmias in MI. According to available evidence, metoprolol’s preventive impact on the degradation of Cx43 serves as a significant pathway in its anti-arrhythmic effect in MI. By this pathway, gap junction communication can be recovered, which results in enhanced conduction velocity. Ultimately, these changes decrease vulnerability to ventricular arrhythmias. As supported by the current research findings, the crocin’s anti-arrhythmic impact might be via mechanisms similar to metoprolol.

There is considerable evidence supporting the crocin’s cardioprotective impact. As a result of pretreatment with crocin, protective effects are produced through the reduction of creatine phosphokinase (CPK) activities, the redox status restoration, and apoptosis suppression in cardiotoxicity caused by patulin in rats. \(^{25}\) Additionally, the protective effect was produced by crocin through the restoration of activity of CK-MB and MDA levels in the heart, as well as improvement of histopathological changes, such as hemorrhages, cardiac muscle cell necrosis, hypertrophy, and infiltration of inflammatory cells in cardiotoxicity induced by DZN. \(^{23}\) Besides, crocin caused improved activity in CPK, CK-MB, and LDH in coronary effluent, cardiac tissue oxidative stress biomarker (MDA), antioxidant enzymes (SOD and catalase), as well as total antioxidant capacity in an animal model (rat) having cardiac ischemia–reperfusion injury. \(^{24}\) Myocardial CK-MB activity was recovered and the elevated MDA level in the heart was reduced in isoproterenol-induced MI by crocin. Also, it caused improvement of changes in the heart tissue, like edema, myocardial necrosis, and leukocyte infiltration. \(^{25}\) Additionally, in an ischemia-reperfusion model of separated heart tissues, the
ST segment increase was recovered by crocin, and crocin enhanced cardiac dysfunction and decreased the infarct size. A cardioprotective effect was produced by crocin in a rat heart ischemia/reperfusion model by enhancing the mechanical function and regulating the production of nitric oxide. Moreover, it was indicated that crocin protects against cardiac arrhythmias induced by reperfusion, supposedly because of the anti-oxidant characteristics of crocin. As found in our results, in addition to these effects, reduction in Cx43 expression is a significant pathway in the crocin effect in MI.

Potential anti-arrhythmic mechanisms of crocin were introduced in our results. Cx43, a critical protein in the cardiac gap junction structure, plays a vital role in cell coupling in electrical signal conduction. There is an association between the Cx43 expression changes and different kinds of arrhythmias in MI. As shown by the experiment in our study, crocin has a preventive effect in a reduction in protein levels and Cx43 messenger RNA in MI. Thus, our findings indicate that crocin has a potential anti-arrhythmia mechanism that might be like metoprolol in preventing degradation of Cx43. Nevertheless, future surveys are required for further evaluation of crocin’s preventive mechanism in the degradation of Cx43.

**CONCLUSIONS**

In conclusion, crocin was demonstrated to be able to reduce the incidence of arrhythmias in an experimental model of MI. This effect was dose-dependent. Based on the observed results, Cx43 expression seems to be a potential pathway in crocin’s anti-arrhythmic mechanisms.

**METHODS**

**Animals.** 50 male Sprague–Dawley rats (weighed 200–250 g) were kept at 23 ± 2 °C with a humidity of 60 ± 5%, under a 12 h light/dark cycle. The care and use of animals were followed for housing animals and conducting experimental procedures. The local Animal Ethics Committee of Shandong University approved all experimental protocols and procedures. First, animals were sorted into 5 groups composed of 10 rats each. The first group was regarded as the normal control group and the second one was considered as MI group, MI being caused by ligation of the left anterior descending artery. The other three groups were treated groups, which received crocin 50 or 10 mg/kg/day or metoprolol 100 mg/kg/day for 1 week following ligation of the left anterior descending artery.

**Induction of MI.** MI was induced via ligation of the left anterior descending artery as previously explained in detail. For induction of MI, anesthesia was applied using sodium thiopental (60 mg/kg body weight, i.p.). Then, the animals experienced tracheal intubation and then were ventilated at a tidal volume of 1.5 cm³/kg and 60–70 breaths/min. This was followed by making an incision on the chest’s left fourth intercostal site. The pericardium was gradually torn, and a 0.6 silk thread was cautiously passed around the left anterior descending artery and fastened. Following 30 min of ischemia, the LAD suture was removed and the ischemic myocardium was reperfused for 120 min. Surface electrocardiography was used for ischemic injury confirmation as previously described. After an identical procedure, a sham group was made, but the actual tying of the suture was absent.

**Recording the Electrocardiogram and Scoring of Arrhythmia.** We recorded the limb lead II electrocardiogram (ECG) in rats. Assessment of the arrhythmias was done according to the previously described process. Using arrhythmia scores, the arrhythmia duration and incidence were quantified. To this end, as seen in Table 1, a grade was given to the animals. The Kubios HRV ECG analysis software was used for the analysis of duration and mean frequency ventricular arrhythmia and mean RR interval in each group.

**Table 1. Scoring of Arrhythmia**

| Score | Definition               |
|-------|--------------------------|
| 0     | no arrhythmia            |
| 1     | <10 s PVC and/or VT      |
| 2     | 11–30 s PVC and/or VT    |
| 3     | 31–90 s PVC and/or VT    |
| 4     | 90–180 s PVC and/or VT, <10 s reversible VF |
| 5     | >180 s PVC and/or VT, >10 s reversible VF |
| 6     | irreversible VF          |

“Ventricular tachycardia (VT), ventricular fibrillation (VF), and premature ventricular contraction (PVC).”

**Evaluation of the Pathologic Changes.** For pathologic investigation, three rats were selected and used from each group. To better visualize the infarcted area, we used a transjugular injection of 2,3,5-triphenyl tetrazolium chloride. In pathologic investigation, sections with full thickness from the myocardium were studied. Using formalin 10%, fixation was done with common laboratory pathologic sample processing.

**RT-PCR and Cardiac Cx43 Western Blotting.** After perfusion experiments, the heart’s ventricular tissue was separated and instantly frozen. On the same day that ventricular sarcolemma was to be prepared, the kept ventricle was homogenized in the hypotonic membrane buffer. This buffer was composed of 1 mM iodoacetamide, 1,10-phenanthroline (1 mM), 0.4 mM phenylmethylsulfonyl fluoride (PMSF), and 1 mM pepstatin A with an ultrasonic homogenizer. The cardiac Cx43 content was determined using the western blotting method.

**RT-PCR, which is an assay of Cx43, was conducted.** The reagent was used for RNA extraction from the cardiac tissue and transcribed into complementary DNA. The RT-PCR reaction solution included reverse and forward primers for an ultimate reaction, besides complementary DNA. Testing of genomic DNA was done using GAPDH as a negative control.

**Statistical Analysis.** Results of the analysis were presented as mean ± SD. Outcomes of groups were compared by one-way ANOVA test. Using the Bonferroni test, the multiple comparisons test was performed for preventing the incorrect appearance of data as statistically significant. Data with P values below 0.05 were considered statistically significant. SPSS 25.0 software was used for data analysis. Graphs were created using Graph-Pad Prism 8.
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The authors declare no competing financial interest. Ethics Statement: The animal study in the present work was reviewed and approved by the Ethics Board of Shandong University.

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REFERENCES
(1) Mensah, G. A.; Forouzanfar, M. H.; Naghavi, M.; Lozano, R.; Ezzati, M.; Moran, A.; Feigin, V.; Vos, T.; Murray, C. J. Comparable estimates of mortality and trends for cardiovascular diseases including congenital heart disease in 21 world regions in 1990 and 2010: The global burden of diseases, injuries and risk factors study. J. Am. Coll. Cardiol. 2013, 61, No. E1406. Van Minh, H.; Byass, P.; Wall, S. Mortality from cardiovascular diseases in Bavi District, Vietnam. Scand. J. Public Health 2003, 31, 26–31.
(2) Gyawali, B.; Mishra, S. R.; Ghimire, S.; Hansen, M. R. H.; Shah, K. J.; Subedee, K. C.; Soti, P. B.; Neupane, D.; Kallestrup, P. The burden and correlates of multiple cardiometabolic risk factors in a semi-urban population of Nepal: a community-based cross-sectional study. Sci. Rep. 2019, 9, 15382. Lindh, M.; Banefelt, J.; Fox, K. M.; Hallberg, S.; Tai, M.-H.; Eriksson, M.; Villa, G.; Svensson, M. K.; Qian, Y. Cardiovascular event rates in a high atherosclerotic cardiovascular disease risk population: estimates from Swedish population-based register data. Eur. Heart J. 2019, 5, 225–232.
(3) Lauer, M. S. Advancing cardiovascular research. Chest 2012, 141, S00–S05. O’Donnell, C. J.; Nabel, E. G. Genomics of cardiovascular disease. N. Engl. J. Med. 2011, 365, 2089–2109.
(4) Bourassa, M. G.; Gurné, O.; Bangdiwala, S. I.; Ghali, J. K.; Young, J. B.; Rousseau, M.; Johnstone, D. E.; Yusuf, S. Natural history and patterns of current practice in heart failure. J. Am. Coll. Cardiol. 1999, 23, A14–A19. Vinson, J. A.; Jang, J.; Dabbagh, Y. A.; Serry, M. M.; Cai, S. Plant polyphenols exhibit lipoprotein-bound antioxidant activity using an in vitro oxidation model for heart disease. J. Agric. Food Chem. 1995, 43, 2798–2799. Johnston, T.; Korlenko, T.; Piirto, M.; Sahebkar, A. Preventing cardiovascular heart disease: Promising nutraceutical and non-nutraceutical treatments for cholesterol management. Pharmacol. Res. 2017, 120, 219–225.
(5) Micek, A.; Gods, J.; Del Rio, D.; Galvano, F.; Grosso, G. Dietary Flavonoids and Cardiovascular Disease: A Comprehensive Dose-Response Meta-Analysis. Mol. Nutr. Food Res. 2021, 65, 2001019. Dhdula, P. V.; Joubert, E.; Muller, C. J. F.; Louw, J.; Johnson, R. Hyperglycemia-induced oxidative stress and heart disease-cardioprotective effects of roobios flavonoids and phenylpyruvic acid-2-O-D-glucoside. Nutr. Metab. 2017, 14, 1–18. Chen, Z.; Zhang, S.-L. The role of flavonoids in the prevention and management of cardiovascular complications: a narrative review. Ann. Palliative Med. 2021, 10, 8254–8263.
(6) Emanuelli, T.; Augusti, P. R.; Roehrs, M. Protective effects of carotenoids in cardiovascular disease and diabetes. Fruit and Vegetable Phytochemicals: Chemistry and Human Health; Wiley, 2017; pp 347–382. DOI: 10.1002/978111958042.1ch16 Yamagata, K. Carotenoids regulate endothelial functions and reduce the risk of cardiovascular disease. Carotenoids 2017, 25, 106.
(7) Deldar, N.; Monsef, M.; Salimanpour, M.; Ostovar, M.; Heydari, M. Wound Healing Potential of Crocin and Safranin, Main Saffron (Crocus sativus L.), the Active Constituents in Excision Wound Model in Rats. Galen Med. J. 2021, 10, 1900. Ebrahiim, F.; Aryaeian, N.; Pahlavani, N.; Abbasi, D.; Hosseini, A. F.; Fallah, S.; Moradi, N.; Heydari, I. The effect of saffron (Crocus sativus L.) supplementation on blood pressure, and renal and liver function in patients with type 2 diabetes mellitus: A double-blinded, randomized clinical trial. Avicenna J. Phytomed. 2019, 9, 322. Abu-Izneid, T.; Rauf, A.; Khalil, A. A.; Olatunde, A.; Khalid, A.; Alhumaydi, F. A.; Aljohani, A. S.; Sabah Uddin, M.; Heydari, M.; Khayrullin, M. Nutritional and health beneficial properties of saffron (Crocus sativus L.): a comprehensive review. Crit. Rev. Food Sci. Nutr. 2022, 62, 2683–2706.
(8) Cardone, L.; Castronuovo, D.; Perniola, M.; Cicco, N.; Candido, V. Saffron (Crocus sativus L.), the king of spices: An overview. Sci. Hortic. 2020, 272, 109560.
(9) Yousefi, M.; Shafaghí, K. Saffron in Persian traditional medicine. Saffron; Elsevier, 2020; pp 393–404. Mzabri, I.; Addi, M.; Berrichi, A. Traditional and modern uses of saffron (Crocus sativus). Cosmetics 2019, 6, 63.
(10) Abedimanesh, N.; Motlagh, B.; Abedimanesh, S.; Bathaie, S. Z.; Sepaham, A.; Ostadrahimi, A. Effects of crocin and saffron aqueous extract on gene expression of SIRT1, AMPK, LOX1, NF-κB and MCP-1 in patients with coronary artery disease: A randomized placebo-controlled clinical trial. Phytother. Res. 2020, 34, 1114–1122.
(11) Shafei, M. N.; Faramarzi, A.; Khajavi Rad, A.; Anaeigoudari, A. Crocin prevents acute angiotensin II-induced hypertension in anesthetized rats. *Avicenna J. Phytomed.* 2017, 7, 345–352. PubMed

(12) Shafei, M. N.; Faramarzi, A.; Khajavi Rad, A. K.; Anaeigoudari, A. Crocin prevents acute angiotensin II-induced hypertension in anesthetized rats. *Avicenna J. Phytomed.* 2017, 7, 345.

(13) Jahanbakhsh, Z.; Rasoulian, B.; Jafari, M.; Shekarforoush, S.; Esmailidelahj, M.; Mohammadi, M. T.; Aghai, H.; Salehi, M.; Khoshbaten, A. Protective effect of crocin against reperfusion-induced cardiac arrhythmias in anesthetized rats. *EXCLI J.* 2012, 11, 20–29. PubMed

(14) Grace, A. A.; Roden, D. M. Systems biology and cardiac arrhythmias. *Lancet* 2012, 380, 1498–1508 From NLM.

(15) Zhou, P.; Yang, X.; Yang, D.; Jiang, X.; Wang, W. E.; Yue, R.; Fang, Y. Integrin-linked kinase activation prevents ventricular arrhythmias induced by ischemia/reperfusion via inhibition of connexin 43 remodeling. *J. Cardiovasc. Transl. Res.* 2021, 14, 610–618.

(16) Mythili, S.; Malathi, N. Diagnostic markers of acute myocardial infarction. *Biomed. Rep.* 2015, 3, 743–748 From NLM. Reddy, K.; Khaqan, A.; Henning, R. J. Recent advances in the diagnosis and treatment of acute myocardial infarction. *World J. Cardiol.* 2015, 7, 243–276. From NLM Wu, Y.; Pan, N.; An, Y.; Xu, M.; Tan, L.; Zhang, L. Diagnostic and prognostic biomarkers for myocardial infarction. *Front. Cardiovasc. Med.* 2021, 7, 617277.

(17) Wang, J.; Shen, B.; Feng, X.; Zhang, Z.; Liu, J.; Wang, Y. A Review of Prognosis Model Associated With Cardiogenic Shock After Acute Myocardial Infarction. *Front. Cardiovasc. Med.* 2021, 8, 754303 From NLM. Sato, R.; Sakamoto, K.; Kaikita, K.; Tsujita, K.; Nakao, K.; Ozaki, Y.; Kimura, K.; Ako, J.; Noguchi, T.; Yasuda, S.; et al. Long-Term Prognosis of Patients with Myocardial Infarction Type 1 and Type 2 with and without Involvement of Coronary Vasospasm. *J. Clin. Med.* 2020, 9, 1686. From NLM

(18) Sedighi, M.; Nazari, A.; Faghihi, M.; Rafieiean-Kouraei, M.; Karimi, A.; Moghiamian, M.; Mozaffarpur, S. A.; Rashidipour, M.; Namdari, M.; Cheraghi, M.; Rasoulian, B. Protective effects of cinnamon bark extract against ischemia-reperfusion injury and arrhythmias in rat. *Phytother. Res.* 2018, 32, 1983–1991.

(19) Scofield, S. L.; Singh, K. Confirmation of Myocardial Ischemia and Reperfusion Injury in Mice Using Surface Pad Electrophysiology. *J. Visualized Exp.* 2016, 117 From NLM.

(20) Tribulová, N.; Knezl, V.; Okruhlíková, L.; Sležák, J. Myocardial gap junctions: targets for novel approaches in the prevention of life-threatening cardiac arrhythmias. *Physiol. Res.* 2008, 57, S1–S13.

(21) Di Diego, J. M.; Antzelevitch, C. Ischemic ventricular arrhythmias: experimental models and their clinical relevance. *Heart Rhythm* 2011, 8, 1963–1968 From NLM.

(22) Goyal, S. N.; Arora, S.; Sharma, A. K.; Joshi, S.; Ray, R.; Bhatia, J.; Kumari, S.; Arya, D. S. Preventive effect of crocin of Crocus sativus on hemodynamic, biochemical, histopathological and ultrastructural alterations in isoproterenol-induced cardiotoxicity in rats. *Phytomedicine* 2010, 17, 227–232 From NLM.

(23) Boussabbeh, M.; Ben Salem, I.; Neffati, F.; Najjar, M. F.; Bacha, H.; Abd-Essefi, S. Crocin Prevents Patulin-Induced Acute Toxicity in Cardiac Tissues via the Regulation of Oxidative Damage and Apoptosis. *J. Biochem. Mol. Toxicol.* 2015, 29, 479–488 From NLM.

(24) Dianat, M.; Esmaeili-zadeh, M.; Badavi, M.; Samarbaf-zadeh, A.; Naghizadeh, B. Protective Effects of Crocin on Ischemia-reperfusion Induced Oxidative Stress in Comparison With Vitamin E in Isolated Rat Hearts. *Jundishapur J. Nat. Pharm. Prod.* 2014, 9, No. e17187, From NLM.

(25) Razavi, B. M.; Hossein-zadeh, H.; Movassaghi, A. R.; Imenshahidi, M.; Abnous, K. Protective effect of crocin on diazinon induced cardiotoxicity in rats in subchronic exposure. *Chem.-Biol. Interact.* 2013, 203, 547–555 From NLM.

(26) Dianat, M.; Esmaeili-zadeh, M.; Badavi, M.; Samarbaf-zadeh, A.; Naghizadeh, B. Protective effects of crocin on hemodynamic parameters and infarct size in comparison with vitamin E after ischemia reperfusion in isolated rat hearts. *Planta Med.* 2014, 80, 393–398 From NLM.