PHARMACOLOGY

QUERCETIN ATTENUATES OXIDATIVE STRESS, INFLAMMATION AND CARDIAC DYSFUNCTION IN ACRYLAMIDE- INDUCED CARDIOTOXICITY

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Abstract: Acrylamide (ACR) is a foodborne toxic agent, formed in food when processed at high temperature. This study aimed at evaluating the biochemical changes induced by ACR and the effect of quercetin as a treatment against ACR induced cardiotoxicity in rats. Wistar rats of either gender (n = 6) were divided into four groups; (i) normal vehicle control, (ii) ACR control group 6 mg/kg i.p. daily (iii) ACR + quercetin treatment group (25 mg/kg p.o.) and (iv) ACR + quercetin treatment group (50 mg/kg p.o.). Routinely general diagnostics characters were assessed daily. At the end of the study (after four weeks) hemodynamic parameters were evaluated and blood samples were collected for estimation of biochemical parameters. The rats were decapitated, their hearts excised, cleaned and weighed. Cardiac homogenate was used to determine antioxidants and oxidative levels. Histopathological evaluations were carried out to determine ACR induced changes. Acrylamide (ACR) treated control rats group show remarkably altered general characteristics and also had elevated markers for myocardial damage, altered hemodynamics, deranged oxidative stress level, increased expression of inflammatory cytokines and also alter histopathological remarkable changes (P < 0.05) as compared with vehicle control group. Treatment with quercetin at 25 and 50 mg/kg reversed the above changes significantly (P < 0.05), the higher dose being more effective. The present study concluded that quercetin protects against ACR-induced cardiotoxicity.

Keywords: acrylamide, quercetin, cardiotoxicity, myocardial damage

Global burden of diseases study cites that diet as major factors behind the rise in food-borne illnesses. Acrylamide formed as a result of food processing when subjected to baking, toasting and deep frying at high temperatures (1). Acrylamide (ACR), an industrially produced α, β-unsaturated reactive molecule, industrially utilized as the soil conditioner, in water waste treatment, in the cosmetic, paper and textile industries and in the laboratory as solid support for electrophoresis (2). ACR primarily discovered during occupational hazard, to later in the year 2002 found accumulating in food. It is has been classified as Group 2A carcinogen by IARC (3) and Category 2 mutagen by the European Union (4). Extensive efforts have been made to analyze the deleterious effect causing capability since its outspread.

Humans are primarily exposed through diet, the occurrence and exposures from food differ from country to country (5). It is formed in carbohydrate-rich food as an intermediate product of the Maillard reactions mainly through the reaction between the amino acid asparagine and reducing sugars when the food is baked, deep-fried, roasted, grilled at the higher temperature of 120°C (6). The reported levels of ACR intake may be at low concentration but chronic consumption of highly processed food negatively impacts the health and hence it is a threat to animals and humans which cannot be overlooked.

ACR has been shown to possess neurotoxic, genotoxic, carcinogenic, reproductive, and cardiotoxic toxicities on experimental animals. The exact mechanism of ACR induced toxicity is not entirely clear, cumulative data showed that oxidative stress and mitochondrial dysfunction played a critical role, as experimental studies show decreased antioxidant levels in various organs. Increased production of reactive oxygen species (ROS) could disturb cellular redox balance, resulting in increased

oxidative stress further declined defensive mechanism and finally cell injury or cell death (7-10). Decreased ATP production associated with mitochondrial dysfunction in the heart may lead to decreased contractility. ACR may alter the functional units responsible for contraction in the heart which may lead to conduction abnormalities such as arrhythmia (11, 12). The genotoxicity is known to be associated with its metabolite glycidamide (2, 3-epoxy1propanamide), as the parent form and metabolite can form adducts with hemoglobin, glutathione, protein, and DNA, however, glycidamide is more reactive towards DNA which explains its genotoxic potential (13). A recent study by Huang et al. reported the cardiac toxicity of acrylamide in the zebrafish (*Danio rerio*) model leading to developmental anomalies. The study suggested that the prenatal period is more vulnerable to ACR toxicity in humans. Although studies reported acrylamide has potentially toxic effects on the heart directly or indirectly (14). Therefore the in-vivo strategies to mitigate ACR-induced toxicity are required. Recently, there been a growing interest in the research on natural antioxidants because of their diverse health benefits and are directly associated with human dietary ingredients and health. Studies with plant extracts and phytochemicals have proven to show a protective effect against ACR.

Quercetin is a member of the flavonols subclass is abundant in the human diet and ubiquitously present in various vegetables as well as in tea and wine and is recognized for its beneficial health effects (15). Some epidemiological studies have shown that increasing daily intake of quercetin can reduce the risk of cardiovascular disease, neurological disorders, some metabolic disorders, and other age-related chronic diseases. The protective effect of quercetin owns to its ability as a potent antioxidant possess free radical scavenging properties, regulating intracellular signaling promoting cell survival, maintaining the integrity of cell membrane and reverse altered energy metabolism in rat heart (16). It also exerts properties such as anti-obesity, anti-inflammatory, anti-allergy, antithrombotic, anticancer and effective in mood disorders (17, 18). In light of the aforementioned pleiotropic bioactivities of quercetin, this study was conducted to investigate the possible protective effect of quercetin against ACR induced oxidative stress and damage in the heart.

MATERIAL AND METHODS

Ethical considerations

The study is a prospective case-control study. Ethical clearance and prior permission were sought from the IAEC before conducting the study (ASCB/IAEC/10/17/122).

Animals

Wistar albino rats of either gender, with an average body weight of 180-200 g were used in this study. Animals were housed in propylene cages with dust-free rice husks in a controlled environment of temperature $23 \pm 2^{\circ}C$, humidity 40-60% and 12 : 12 h light-dark cycle as per the guidelines of CPCSEA norms.

Experimental design and study protocol

One week after acclimatization to laboratory conditions, rats were randomly divided into four groups (n=6/group). Study Group I: Control (rats received normal vehicle); Group II: ACR, (rats received acrylamide (ACR) 6 mg/kg i.p. (route daily once for 4 weeks; Group III: ACR + Q (25 mg/kg) and Group IV: ACR + Q (50 mg/kg), rats received daily ACR 6 mg/kg i.p. route and Quercetin at dose 25 and 50 mg/kg by oral gavage for 4 weeks with respect of study groups, during study period food and water were available *ad libitum*.

Bodyweight, feed intake and water intake were recorded daily. At the end of the experimental period (4th week), animals were overnight fasted since 28th day (to next 16 h) on 29th days animals were utilized to performed hemodynamic parameters. At the end of the experiments, animals were sacrificed and blood was collected from the carotid artery for the assessment of the various biochemical analysis. After that, hearts were excised, cleaned and collected to estimate isolate heart weight. Heart homogenized in an appropriate buffer (pH = 7.4) and centrifuged. The resultant supernatant was used for biochemical analysis. Afterward, heart tissue was utilized for histopathological studies.

Biomarkers of cardiotoxicity

The blood sample was obtained from the overnight fasted rats after the administration of the last dose. The blood was collected by puncturing retro-orbital plexus using heparinized glass capillary tubes under chloroform anesthesia and collected in Eppendorf tubes and then centrifuged at 3000 rpm for 10 min, serum separated for analysis of hematological parameters such as Lactate dehydrogenase (LDH), Creatine kinase-MB (CK-MB) and Alkaline phosphatase (ALP). All analysis was performed with commercially available kits using autoanalyzer.

Hemodynamic evaluation

Rats were anesthetized with 25% urethane (1.5 g/kg, i.p.). Throughout the experimental protocol

body, the temp of the animals was maintained at 37°C. The neck was opened with a ventral midline incision to perform a tracheostomy. The left carotid artery was cannulated with a polyethylene tube (internal diameter 0.30 mm; outer diameter 0.40 mm) attached to a three-way cannula. The cannula was heparinized (Heparin 300 IU/mL) and connected to POWER LAB 4/30 (AD Instruments, NSW, Australia) system using a pressure transducer for the measurement of Arterial pressures (AP), Systolic arterial pressure (SAP), Diastolic arterial pressure (DAP), Mean arterial pressure (MAP) and Heart rate (HR).

Determination of glutathione (GSH) activity in tissue supernatant

Measurement of the reduced glutathione (GSH) was done by the Ellman's reagent (5, 5'-dithiobis-2-nitrobenzoic acid (DNTB) method of Ellman [19]. The homogenate was added with an equal volume of 20% trichloroacetic acid (TCA) containing 1 mM EDTA to precipitate the tissue proteins. The mixture was allowed to stand for 5 min prior to centrifugation for 10 min at 3000 rpm. The supernatant 0.2 mL was then transferred to a new set of test tubes and added 2.3 mL of potassium phosphate buffer (0.2 M, pH 7.6). To the tissue homogenate, 0.5 mL (DTNB) (0.001 M) in a buffer was added. The absorbance of the reaction product in the cuvette was read after 5 min at 412 nm using UV/ Visible double beam spectrophotometer.

Determination of malondialdehyde (MDA) content in tissue supernatant

The reaction mixture contained 0.1 mL of tissue homogenate, 0.2 mL of 8.1% sodium dodecyl sulfate (SDS), 1.5 mL of 20% acetic acid and 1.5 mL of 0.8% aqueous solution of Thiobarbituric acid (TBA). The pH of 20% acetic acid was adjusted with 1 Í NaOH to 3.5. The mixture was finally made up to 4.0 mL with distilled water and heated at 95°C for 60 min on an oil bath. After cooling under tap water, 1.0 mL of distilled water and 5.0 mL of a mixture of n-butanol and pyridine (15 : 1 by volume) was added and shaken vigorously on a vortex mixer. After centrifugation at 3000 rpm for 5 min, the absorbance of the organic layer (upper layer) was measured immediately at 532 nm using appropriate controls in UV/Visible spectrophotometer (Shimadzu 1700, Singapore), [20].

Determination of Superoxide dismutase (SOD) activity in tissue supernatant

To estimate the SOD activity in the colon tissue the method described by Kono [21] was followed. The reaction mixture contained 1.3 mL sodium carbonate buffer (50 mmol/ L), 500 µL nitroblue tetrazolium (NBT) (96 µmol/L), 100 µL Triton X-100 (0.6%) and 100 µL hydroxylamine-HCl (20 mmol/ L). After 2 min, 70 µL supernatant was added and the absorbance at 560 nm was read against a blank (reaction mixture without sample). The increase in absorbance at 540 nm was recorded to
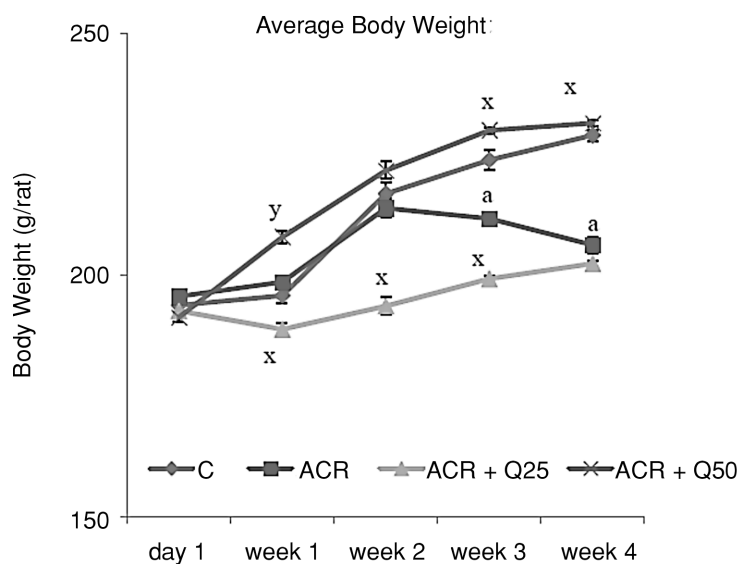

Figure 1. Effect of quercetin on body weight.
Values are mean ± SEM for n = 6 rats in each group.^{a}(p < 0.001) *vs* normal control group; ^{x}(p < 0.001), ^{y}(p < 0.01) vs acrylamide group.

calculate the percentage inhibition of NBT reduction.

Percentage inhibition of NBT reduction (x) =

The sample volume that produced 50% inhibition was considered as 1 unit of enzyme activity and SOD activity was expressed as units/ mg.

Cytokines analysis

The cytokines; Interleukin 6 (IL-6), Tumor necrosis factor-alpha (TNF-α) were evaluated using commercially available ELISA kits from Krishgen, Mumbai as per the manufacturer's instructions. The optical density of each well was read at 450 nm, values are expressed in pg/mL.

Histopathological examination

Heart samples were sent to diagnostic commercial lab histopathological slides were prepared and stained with hematoxylin and eosin (H and E) dyes and the pictures were taken from the prepared slides with the help of photomicroscope and changes in histology results were observed.

Drugs and chemicals

Acrylamide (ACR) was obtained from Sisco Research Laboratories Pvt Ltd (SRL) Delhi, quercetin was purchased from Hi-Media, Mumbai, LDH, ALP, and CK-MB kits were purchased from Reckon Diagnostics Private Limited Vadodara and ELISA kits were procured from Krishgen Biosystems (Mumbai, Maharashtra, India).

Statistical analysis

The data were expressed as Mean ± SEM were analyzed by one-way ANOVA followed by Tukey multiple comparison tests using Graph Pad Prism software package. A value of $p < 0.05$ was considered to be significant.

RESULTS

General observations

ACR treated showed no discomfort or behavioral anomalies during the experimentation period. Bodyweight of ACR treated rats significantly ($p < 0.001$) declines at the onset of the 3rd week and further declines at 4th week with respect to the normal control group. Treatment with quercetin low dose (25 mg/kg; p.o.) show a gradual increase in body weight over the duration of 4 weeks however a significant ($p < 0.001$) difference observed in body weight gain when compared to ACR treated group. Quercetin high dose (50 mg/kg; p.o.) show a significant ($p < 0.01$; $p < 0.001$) progressive increase in body weight on 1st, 2nd weeks and 3rd and a 4th week respectively when compared to ACR treated group (Fig. 1). A non-significant ($p > 0.25$) decrease in the absolute and relative weight of the heart was observed in ACR treated group (Table 1). The difference in food consumption was observed in treat-
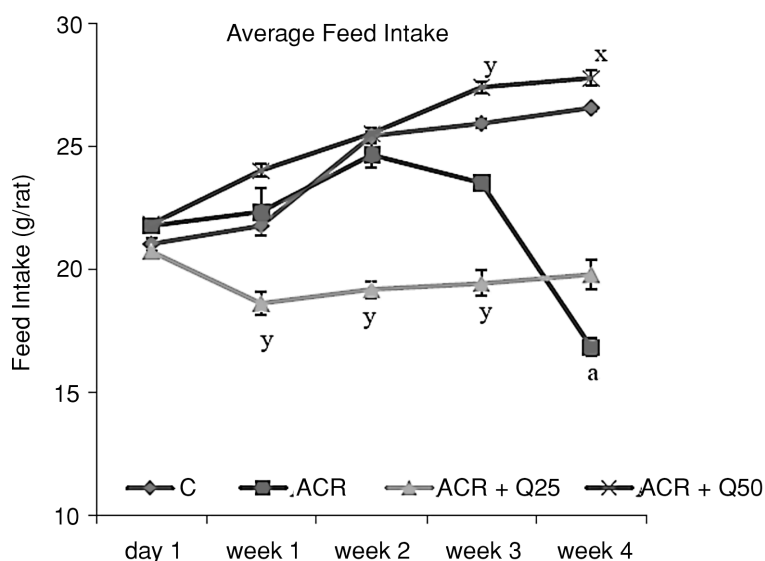

Figure 2. Effect of quercetin on feed intake.
Values are mean ± SEM for n = 6 rats in each group. a($p < 0.001$) vs normal control group; x($p < 0.001$), y($p < 0.01$) vs acrylamide group.
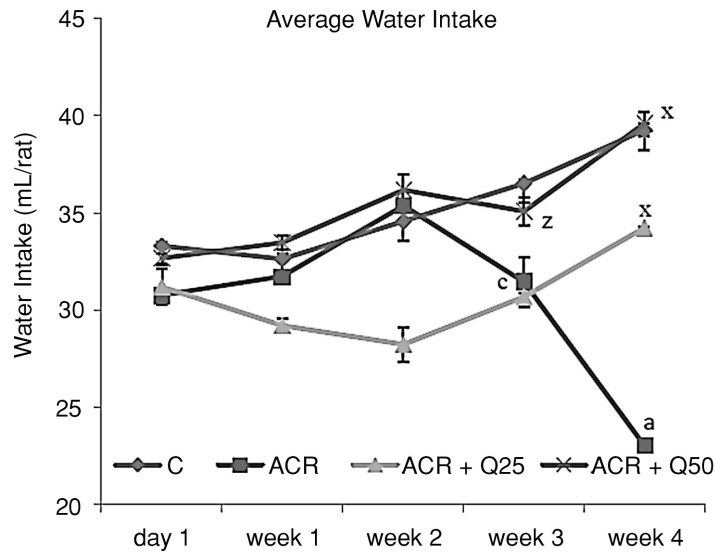
Figure 3. Effect of quercetin on water intake.
Values are mean ± SEM for n = 6 rats in each group. ^{a}(p < 0.001), ^{c}(p < 0.05) vs normal control group; ^{x}(p < 0.001), ^{z}(p < 0.05) vs acrylamide group.

Table 1. Effect of Quercetin on heart weight and relative heart weight.

Study groups	Absolute heart weight (g)	Relative heart weight* (10^{-3}) g
Control	0.72 ± 10.73	3.27 ± 0.115
ACR (6 mg/kg)	0.705 ± 20.95	3.01 ± 0.105
ACR+Q (25 mg/kg)	0.693 ± 22.72	3.19 ± 0.080
ACR+Q (50 mg/kg)	0.705 ± 12.87	2.97 ± 0.194

Values are mean ± SEM for n = 5-6 in each group.

Table 2. Effect of quercetin on serum biomarkers.

Study groups	LDH (IU/L)	CK-MB (IU/L)	ALP (IU/L)
Control	931.5 ± 2.872	91 ± 1.095	647.3 ± 10.60
ACR (6 mg/kg)	2321 ± 6.906^a	233.5 ± 0.918^a	821 ± 1.095^a
ACR+Q (25 mg/kg)	1733 ± 4.757^x	142.3 ± 4.773^x	460.5 ± 2.212^x
ACR+Q (50 mg/kg)	927.3 ± 1.862^x	69.83 ± 6.030^x	383.8 ± 1.815^x

Values are expressed as Mean ± SEM (n = 6). ^{a}(p < 0.001) *vs* normal control group; ^{x}(p < 0.001) vs acrylamide group. Abbreviations: Lactate dehydrogenase (LDH), Creatine kinase-MB (CK-MB) and Alkaline phosphatase (ALP).

ed groups. Significant (p < 0.001) reduction in food intake in ACR treated rats towards the onset of the 3rd and 4th week was observed when compared with the normal control group. Quercetin at low dose shows a gradual increase in feed consumption over the duration of 4 weeks however there is a significant (p < 0.01) difference in feed consumption when compared to ACR treated group. Quercetin at high dose shows a significant (p < 0.01, p < 0.001) progressive increase in feed intake on 3rd and a 4th week respectively when compared to ACR treated group (Fig. 2). Water consumption was significantly (p < 0.05, p < 0.001) reduced in ACR treated group when compared to control by the end of the 3rd and 4th

week respectively. Significantly (p < 0.001) increased water consumption was noted in quercetin treated groups in respect of ACR treated groups (Fig. 3).

Cardiac damage markers

Serum lactate dehydrogenase (LDH), Creatine kinase-MB (CK-MB) and alkaline phosphatase (ALP) activity show on treatment groups. A significant (p < 0.001) increase in the activities of these enzymes was observed when compared to the control group after ACR administration. Quercetin low and high dose treatment group significantly (p < 0.001) restore the normal level of cardiotoxicity elevated markers enzymes when compared to ACR treated group (Table 2).

Hemodynamic parameters

Acrylamide treated rats showed the significant (p < 0.001) reduction in the Hemodynamic parameters; Arterial pressures (AP), Systolic arterial pressure (SAP), Diastolic arterial pressure (DAP), Mean arterial pressure (MAP) and Heart rate (HR) when compared to the normal control group. Administration of quercetin (25 mg/kg) showed significant (p < 0.05. p < 0.01, p < 0.001) increase in AP, MAP, SAP and HR but non-significant (p > 0.5) increase in DAP when compared to ACR treated group. Quercetin (50 mg/kg) treated group showed a highly significant (p < 0.001) increase in AP, SAP, DAP, MAP, and HR when compared to ACR treated group (Table 3).

Biochemical analysis

The activities of antioxidant enzyme and lipid peroxidation products are presented in (Table 4). Acrylamide administration resulted in significant (p < 0.001) decrease in the Glutathione (GSH), Superoxide dismutase (SOD) and significant (p < 0.001) increase in Malondialdehyde (MDA) level in rats heart when compared with the normal control group. Quercetin 25 mg and 50 mg/kg significantly (p < 0.001) restored the altered levels of GSH and

Table 3. Effect of quercetin on hemodynamic parameters.

Study groups	AP (mmHg)	SAP (mmHg)	DAP (mmHg)	MAP (mmHg)	HR (bpm)
Control	113.4 ± 0.840	127.7 ± 0.775	93.54 ± 1.036	104.9 ± 0.850	363.3 ± 1.104
ACR (6 mg/kg)	93.06 ± 1.328[a]	99.80 ± 1.450[a]	75.68 ± 0.819[a]	83.65 ± 1.079[a]	305.58 ± 2.294[a]
ACR+Q (25 mg/kg)	98.17 ± 0.686[z]	110.5 ± 1.232[x]	78.48 ± 0.507	89.17 ± 0.682[y]	320.6 ± 2.536[x]
ACR+Q (50 mg/kg)	108.7 ± 1.292[x]	119.5 ± 1.757[x]	88.34 ± 1.967[x]	98.61 ± 1.146[x]	350.7 ± 1.548[x]

Values are expressed as mean ± SEM (n = 6). [a] (p<0.001) *vs* normal control group; [x](p < 0.001), [y](p < 0.01),[z](p < 0.05) vs acrylamide group. Abbreviations: Arterial pressures (AP), Systolic arterial pressure (SAP), Diastolic arterial pressure (DAP), Mean arterial pressure (MAP) and Heart rate (HR)].
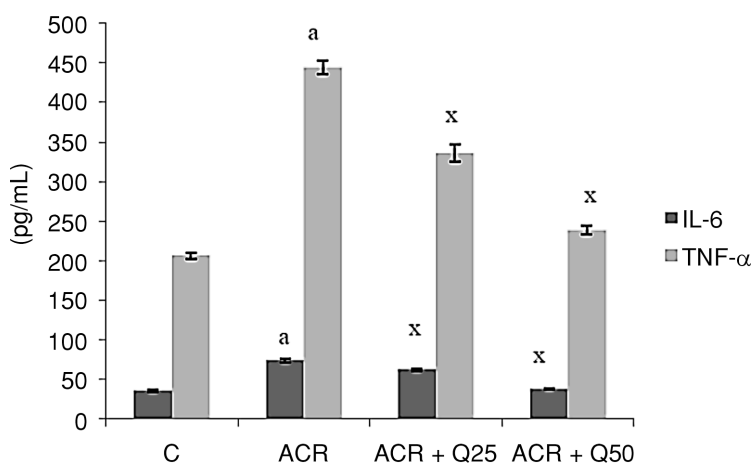

Figure 4. Effect of quercetin on serum cytokines levels.
Values are mean ± SEM for n = 6 rats in each group. [a] (p < 0.001), *vs* normal control group; [x](p < 0.001) vs acrylamide group. Abbreviations: Interleukin 6 (IL-6), Tumor necrosis factor-alpha (TNF-α)].

Table 4. Effect of quercetin on cardiac glutathione (GSH), superoxide dismutase (SOD) and malondialdehyde (MDA) level.

Study groups	GSH (řmol/mL)	MDA (nmol/mL)	SOD (U/g) tissue
Control	16.82 ± 0.376	5.43 ± 0.0802	13.49 ± 0.123
ACR (6 mg/kg)	6.075 ± 0.349[a]	13.52 ± 0.529[a]	11.40 ± 0.145[a]
ACR+Q (25 mg/kg)	11.02 ± 0.235[x]	9.39 ± 0.214[x]	16.0 ± 0.217[x]
ACR+Q (50 mg/kg)	24.58 ± 0.205[x]	6.86 ± 0.092[x]	19.73 ± 0.080[x]

Values are expressed as mean ± SEM (n = 6). [a]($p < 0.001$) *vs* normal control group; [x]($p < 0.001$) vs acrylamide group.

SOD and MDA, however, the effect was more pronounced with quercetin 50 mg/kg.

Inflammatory cytokines

Serum levels of TNF-α and IL-6 were significantly increased ($p < 0.001$) in ACR treated rats when compared to the control group. The Quercetin treated groups show significant ($p < 0.01$, $p < 0.001$) reduction in elevated cytokines levels and the reduction was more prominent in quercetin 50 mg/kg treated rats (Fig. 4).

Histopathological findings

Histopathological findings are presented in (Fig. 5). Acrylamide treated rats show inflammatory cell infiltration, myocardial loss and interstitial spaces, loss of nuclei and striations and vacuolization. Quercetin 50 mg/kg treatment recouped all the changes and was more pronounced than 25 mg/kg.

DISCUSSION

Extensive research has been carried out showcasing the toxic potential of ACR, primarily discovered during occupational hazard to later found accumulating in food. Humans are exposed to ACR through various routes of exposure but prominently from the diet at very low concentration chronically owing to the increased consumption of highly processed food rich in carbohydrates and hence a threat to human and animal health (22). However, the mechanism by which ACR exposure causes cellular dysfunction in humans and animals is not entirely clear but oxidative stress, mitochondrial dysfunction mediated cellular damage and erythrocyte deformability may play a critical role and can act as a precursor to the onset of cardiovascular disease. In this study, we demonstrate that the exposure of ACR may induce biochemical, physiological changes in the heart as our result suggests that ACR poses a threat to cardiac health and treatment with quercetin quells these changes via cardioprotective action.

During the study period, clinical conditions such as diarrhea, growth near neck and abdomen region were observed in the ACR (6 mg/kg; i.p.) treated group and low dose quercetin (25 mg/kg; p.o.) treated group. However, the frequency differed in both groups also the growth subsided with ACR withdrawal. In the current study, body weight, feed intake and water intake were significantly decreased in rats treated with ACR (6 mg/kg; i.p.) by the end of the 3rd week and at the end of the 4th- week body-weight decreased further in respect to normal maintained body weight as compared to control group. Reduction in body weight may be due to decreased feed intake, it may also be related to the significant decrease in Glutathione (GSH) levels as GSH is essential for the function and structural integrity of gut (23). Acrylamide (ACR) may be associated with alteration in thirst and hunger regulation centers in the hypothalamus. A gradual increase in weight gain, feed intake, and water intake was observed in quercetin (25 mg/kg; p.o.) in respect of normal weight gain when compared to ACR treated group. On treatment with quercetin (50 mg/kg; p.o.) significant progressive increase in body weight, feed intake, and water intake was noted as compared to ACR treated rats. The observed reduction in body weight, feed intake and water intake of ACR treated rats are in accordance with previous reports (24, 25). Cardiac markers Lactate dehydrogenase (LDH), Creatine kinase-MB (CK-MB) and Alkaline Phosphatase (ALP) are endogenous substances released in the bloodstream when the heart is damaged or stressed. Activities in the heart reflect the leaky membrane and the degree of myocardial damage. Numbers of studies consistently report the elevated activities of these enzymes in the serum clearly indicating a certain type of cardiovascular disease (26, 27) and studies also report elevated levels of these enzymes in ACR induced toxicity and our results in accordance with these reports (24). Intraperitoneal administration of ACR remarkably (6 mg/kg) elevates the levels of LDH, CK-MB; ALP as compared to control predicting the severity of

ACR-induced damage to the myocardium. On treatment with quercetin (25 mg/kg) significantly reduce in all enzyme levels when compared to ACR control group, however on treatment with quercetin (50 mg/kg) more emphasizes reduce these enzymes levels of CK-MB (69.83 IU/L) and ALP (383.8 IU/L) thus confirming the maintenance of normal structural, architectural integrity and preventing the leakage of these enzymes into the bloodstream. This was further confirmed by histopathological changes in the heart including inflammation, myocardial loss, and vacuolization.

Treatment with Acrylamide (ACR) not only resulted in biochemical changes also an alteration in cardiac physiology. In the present study, ACR is reported to be an inhibitor of both systolic and diastolic cardiac function; intraperitoneal (i.p.) administration showed a significant decrease in AP, SAP, DAP, MAP, and HR when compared to the normal control group. On treatment with quercetin, the levels of AP, SAP, DAP and HR revert back to normal when compared with the ACR treated group. A however non-significant increase in DAP and the significant increase in AP, SAP, MAP, and HR in
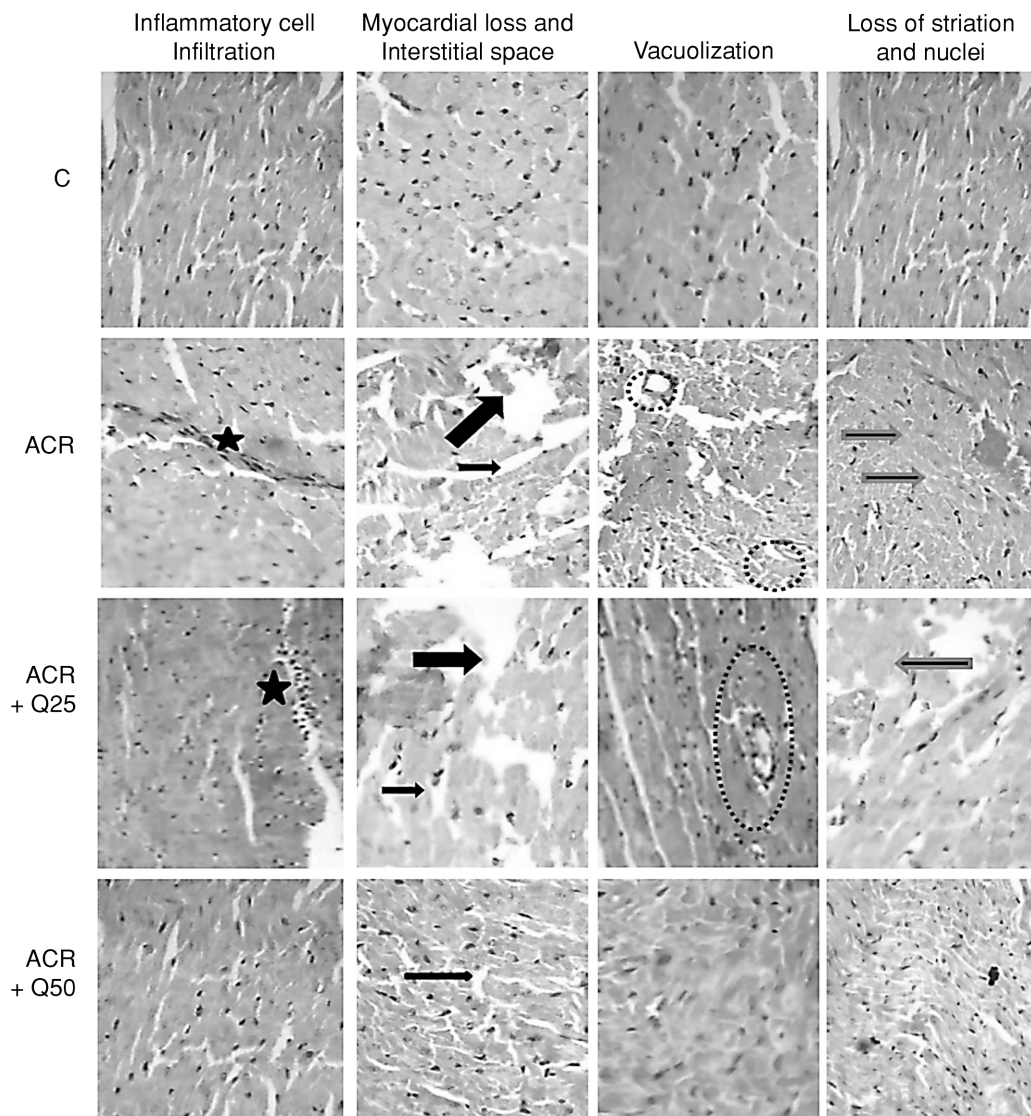

Figure 5. Effects of quercetin on histopathology changes in various study groups of heart tissues.
Star marks indicate in figure inflammatory cell infiltration, myocardial loss (thick arrows) and interstitial spaces (thin arrows), vacuolization (circles), Loss of striations and nuclei (lightly shaded arrows).

quercetin (25 mg/kg) was observed. Quercetin (50 mg/kg) clearly ameliorates these hemodynamic changes. However, the mechanism by which ACR causes alteration of cardiac physiology is not clear. Possible causes of decreased contractility of the heart due to underproduction of ATP by mitochondria may be associated, as ACR induced mitochondrial dysfunction has been reported in experimental studies (12). Acrylamide can increase blood viscosity impacting the blood fluidity which increases the occurrence of cardiovascular disease (28). Thus maintenance of the normal cardiac physiology and normalize in the hemodynamic parameters results supported that quercetin protects the heart and myocardial deformability by reducing oxidative stress.

In the current study, Acrylamide exposure was found to markedly decrease the antioxidant enzyme activity in the heart, indicating that antioxidant activity of heart tissues was impaired and increased lipid peroxidation in heart tissue when compared to the normal control group. An antioxidant defense system, mainly involved in the scavenging reactive oxygen species prevents oxidative stress. GSH (glutathione), SOD (Superoxide dismutase) which serve as scavengers of ROS (reactive oxygen species) were found to be decreased. The reduction could be due to the Acrylamide effect on the cellular redox chain and generates oxygen species and imbalance occurs between antioxidants and oxidation function (7). With the increase in Acrylamide, oxidative stress increases and it has been shown to stimulate apoptosis (14), which might explain the myocardial loss and inflammation observed in heart tissue. Quercetin as a potent antioxidant (18) showed a remarkable increase in antioxidant enzymes and reducing oxidative stress when compared with ACR group which accounts for its protective action against ACR toxicity. Reactive oxygen species induced lipid peroxidation can create deleterious effects throughout the body, such as cardiovascular and neurodegenerative diseases, quercetin can attenuate lipid peroxidation via interaction with radicals (29). In the present study, Acrylamide treated groups showed increased lipid peroxidation in heart tissue when compared to the normal control group. On treatment with quercetin (25 and 50 mg/kg), a significant decrease in lipid peroxidation and enhanced levels of antioxidants were noted. This improvement was more pronounced in rats treated with quercetin (50 mg/kg). Thus, our study indicates that quercetin ameliorates Acrylamide induced oxidative stress via its protective action. Besides,

reactive oxygen species are able to activate nuclear factor kappa B (NF-kB) and its controlled cytokine (30). ACR induced production of inflammatory cytokines Tumor necrosis factor (TNF-α) and interleukin-6 (IL-6) has been reported in experimental studies and our results are in accordance with these reports (31). In our study, ACR elevates the expression of these inflammatory cytokines and treatment with quercetin modulates the elevated levels and the following findings can be confirmed by histopathological changes. Reduced level of inflammatory cytokines confirms quercetin anti-inflammatory activity.

Biochemical findings were further confirmed by Histopathological examination. Altered cardiac histoarchitecture in ACR treated rats, such as inflammation, myocardial loss, and interstitial space, vacuolization, loss of striation and nuclei were found in our study. Quercetin treatment recouped all the above induced cardiac alterations. The improvement was more pronounced in quercetin (50 mg/kg) treated group.

CONCLUSION

The present study suggests the cardioprotective effect of quercetin on ACR induced Cardiotoxicity in rats. The protective effect of quercetin may be due to its ability to scavenge free radicals, regulate immunity as reduced inflammation was observed in treated groups and protect against oxidant stress induced by ACR. Our conducted study reports the improved level of antioxidants, heart enzymes, hemodynamic and histological alterations in quercetin treated rats. Overall these results signify the importance of quercetin as a dietary flavonoid and in the management of ACR cardiotoxicity moreover, the use of potent flavonoids such as quercetin itself may pose to be a more vivid and prominent in treating toxicity due to food-based toxins such as ACR.

Competing of interest statement

The authors declare that there is no conflict of interest.

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