In-vitro phytopharmacological effect and cardio protective activity of Rauvolfia tetraphylla L.

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Manuscript details
Abstract

The present study aimed to evaluate the cardiac protective activity in Rauvolfia tetraphylla L. The rats are divided into five groups of six in each and myocardial infarction was induced in experimental rats by intraperitoneal injection of isoproterenol hydrochloride. Each group of rats were treated with combination of isoproterenol and R. tetraphylla leaves and also treated separately. At the end of the experimental period, the blood was collected Rat from groups and examined biochemical estimation. The results provided in the paper endorse that leaf extract of R. tetraphylla has cardio protective potential.

Keywords:
Myocardial infarction, Rauvolfia tetraphylla L. and Rat

1. Introduction

Globally, cardiovascular diseases (CVD) constitute a leading cause of mortality. In developing countries like India, the incidence of CAD is increasing alarmingly and also struggling to manage the impact of CVD along with the growing burden of obesity, Type II diabetes and hypertension (El-Sayed et al., 2011). Heart disease in India occurs 10 to 15 years gushing factor between normal subjects and those with earlier than in the West. One fifth of the deaths in India are from coronary heart disease (CHD). By the year 2020, it will account for one third of the deaths. Current projections suggest that by the year 2020, India will have the largest CVD burden in the world (Devasagayam et al., 2004) and is on the verge of a cardiovascular epidemic. By the year 2015, cardiovascular mortality is likely to rise to the order of 103% in males and 90% in females. The circulatory system disorders are going to be the greatest killer in India by the end of the year 2015 (Sabari Das et al., 2002).

Myocardial Infarction (MI) is one of the main causes of death from cardiovascular diseases and associated with ischemic necrosis of cardiac muscle due to compromised supply of blood to a portion of myocardium for proper physiological function (Anversa et al., 1991). MI is commonly known as a heart attack that occurs when the blood supply to a part of the heart is interrupted, causing death of heart tissue. It is the leading cause of death for both men and women all over the world (WHO 2002).

Catecholamines at low concentrations are considered to be beneficial in regulating heart function by exerting a positive inotropic effect. Catecholamines administration at high doses or excess release of it, from the endogenous stores may deplete the energy reserve of cardiomyocytes and thus may result in biochemical and structural changes which are responsible for the development of irreversible damage. The rat model of isoproterenol (ISO) induced myocardial necrosis serves as a well accepted standardized model to evaluate several cardiac dysfunctions (Wexler 1978) and to study the efficacy of various natural and synthetic cardio protective agents (Rathore et al., 1998). ISO induced myocardial infarction is widely used experimental model for several reasons. The model is characterized by extraordinary technical simplicity, an excellent reproducibility as well as an acceptable low mortality. MI induced by ISO has been reported to show many metabolic and mor-
phologic aberrations in the heart tissue of the experimental animals similar to those observed in human myocardial infarction (Nirmala and Puvanakrishnan 1996). ISO induced necrosis is maximal in the sub-endocardial region of the left ventricle and in the interventricular septum. Continuous infusion of ISO in rat elicits typical cardiac gene expression similar to that observed in cardiac hypertrophy caused by pressure overload (Nirmala and Puvanakrishnan 1996).

2. Materials and methods

The MI was induced in experimental rat by intraperitoneal injection of isoprotrenal hydrochloride 20 mg/100 g body weight, dissolved in physiological saline, for two consecutive days (Prabhu et al., 2005).

2.1. Experimental design

Body weights of animals were recorded and they were divided into five groups of six rats each as follows.

**Group I:** Normal animals received with standard fed and water toallowed ad libitum throughout the experimental period.

**Group II:** Rats were orally fed 0.9% normal saline once daily for 21 days and in addition to received isoprotrenal (20 mg/100 g body weight) on the 22nd and 23rd day at an interval of 24 h.

**Group III:** Rats were pre-treated with *R. tetraphylla* leaf extract (500 mg/kg body weight) alone for a period of 21 days.

**Group IV:** Rats were pre-treated with *R. tetraphylla* leaf extract (500 mg/kg body weight) for a period of 21 days and in addition to received isoprotrenal (20 mg/100 g body weight) on the 22nd and 23rd day at an interval of 24 h.

**Group V:** Rats were pre-treated with Simvastatin (60 mg/kg body weight) for a period of 21 days and in addition to received isoprotrenal (20 mg/100 g body weight) on the 22nd and 23rd day at an interval of 24 h.

2.2. Collection of blood and preparation of plasma sample

At the end of the experimental period, the animals were anaesthetized using chloroform vapour prior to dissection. Blood was collected by cardiac puncture into ETDA containing tubes. The blood was allowed to standing at room temperature for 30 min and then refrigerated for another 30 min. The resultant clear part was centrifuged at 3000 rpm for 10 min, and then the plasma was isolated and stored at refrigerated until required for analysis.

2.3. Tissue homogenate

Immediately after blood collecting, the animals were sacrificed by cervical dislocation and the heart was dissected out, washed with ice-cold physiological saline. The required amount was weighed and homogenized using a Teflon homogenizer. Tissue homogenate was prepared in 0.1 M Tris Hcl buffer (pH 7.4) and used for the estimation of various biochemical parameters.

2.4. Histological studies

The heart tissue was fixed in 10% normal saline for 72 h after which the tissues were sliced to a thickness of 2.1 mm each. These were dehydrated using alcohol of graded concentration. They were further treated with paraffin wax and cast into blocks; sections of the tissues were cut on a microtome to 5 µm. These were later attached to a slide and dried. The samples slides were viewed on a photographic microscope to find out histological changes.

2.5. Biochemical examinations

2.5.1. Determination of reduced glutathione (GSH)

Reduced glutathione was estimated by method of Moron et al (1979). Briefly, 0.5 mL of serum sample was precipitated with 1 mL of 10% TCA and the precipitate was removed by centrifugation. To 0.5 mL of the supernatant, 1 mL of DTNB was added and the total volume was made up to 3 mL with phosphate buffer. The absorbance was read at 412 nm.

2.5.2. Estimation of malondialdehyde (MDA/LPO)

Malondialdehyde was estimated by the thiobarbituric acid assay method of Beuge and Aust (1978). Briefly, the serum sample was mixed thoroughly with 2 mL of TCA-TBA-HCl reagent and heated for 15 min in a boiling water bath. After that the flocculants were centrifuged at 1000 rpm for 10 min. The absorbance of the sample was read at 535 nm against a blank without sample.

2.5.3. Assay of superoxide dismutase (SOD)

Superoxide dismutase activity was determined by the procedure of Kakkar et al (1984). Briefly, 0.5 mL of serum sample was diluted to 1 mL with water. Then 2.5 mL of ethanol and 1.5 mL chloroform (all reagents chilled) were added and mixed for one minute at 40°C and then centrifuged. The enzyme activity in the supernatant was determined. Briefly, the assay mixture contained 1.2 mL of sodium pyrophosphate buffer, 0.01 mL of phenazinemethosulphate, 0.3 mL of nitroblue tetrazolium, 0.2 mL of NADH, appropriately diluted enzyme preparation, 0.01 mL of KCN and water in a total volume of 3 mL. Reaction was initiated by the addition of NADH. After incubation at 30°C for 90 s, the reaction was stopped by the addition of 1 mL glacial acetic acid. The reaction mixture was stirred vigorously and shaken with 4 mL of n-butanol. The intensity of the chromogen in the n-buta nol layer was measured at 560 nm against butanol as blank. A system devoid of enzyme served as control.

2.5.4. Assay of Catalase

The activity of catalase was assayed by the method of Beers and Sizer (1952). Briefly, 1.9 mL of distilled water and 1 mL of the hydrogen peroxide reagent, as substrate was added and incubated for 4-5 min. After that 0.1 mL of serum sample was added and recorded the decrease in absorbance for 2-3 min at 240 nm.

2.5.5. Assay of glutathione peroxidase

The activity of mitochondrial glutathione peroxidase
was assayed by the method of Rotruck et al (1973). Briefly, the reaction mixture consisted of 0.2 mL each of EDTA, sodium azide, H$_2$O$_2$, 0.4 mL of phosphate buffer; 0.1 mL of serum sample was incubated at 37°C. The reaction was arrested by the addition of 0.5 mL of TCA and the tubes were centrifuged at 2000 rpm. To 0.5 mL of supernatant, 4 mL of disodium hydrogen phosphate and 0.5 mL DTNB were added and the colour developed was read at 420 nm immediately.

2.5.6. Estimation of Ascorbic acid (Vitamin C)

The level of ascorbic acid was estimated by the method of Omaye et al (1979). Briefly, 0.5 mL of serum sample, 0.5 mL of water and 1 mL of TCA were added, mixed thoroughly and centrifuged at 8000 rpm for 10 min. To 1 ml of the supernatant, 0.2 mL of DTC reagent was added and incubated at 37°C for 3 h. Then 1.5 mL of sulphuric acid was added, mixed well and the solutions were allowed to stand at room temperature for another 30 min. The colour developed was read at 520 nm.

2.5.7. Estimation of α-tocopherol (Vitamin E)

Alpha-tocopherol was estimated by the method of Baker et al (1980). Briefly, 0.5 mL of serum sample, 1.5 mL of ethanol was added, mixed and centrifuged at 8000 rpm for 10 min. To 1 ml of the supernatant, 0.2 mL of DTC reagent was added and incubated at 37°C for 3 h. Then 1.5 mL of sulphuric acid was added, mixed well and the solutions were allowed to stand at room temperature for another 30 min. The colour developed was read at 520 nm.

2.5.8. Activity of SGOT (AST)

The serum GOT was estimated by the method of Reitman and Frankel (1957). Briefly, 0.1 mL of serum was mixed with 0.5 mL of substrate reagent and incubated for 60 min at 37°C. After that, 0.5 mL of colour reagent was added and further incubation for 20 min at 37°C. After the incubation, 3 mL of alkaline reagent was added to stop the reaction and the colour intensity was read at 505 nm.

2.5.9. Activity of SGPT (ALT)

The serum GPT was estimated by the method of Reitman and Frankel (1957). Briefly, 0.1 mL of serum was mixed with 0.5 mL of substrate reagent and incubated for 60 min at 37°C. To that, 0.5 mL of colour reagent was added and further incubated for 20 min at 37°C. After the incubation, 3 mL of alkaline reagent was added to stop the reaction and the colour intensity was read at 505 nm.

2.5.10. Estimation of Troponin-T

Troponin-T estimated by the method of Bhaskar and Rao (2002). Briefly, 0.2 mL of serum was diluted to 1.8 mL with distilled water and vortexed for 2 min. The tubes were allowed to stand for 5 min and centrifuged at 3000 rpm for 10 min. The supernatant was discarded and the precipitate was washed thrice with distilled water. Further, 4.5 mL of alkaline capper reagent was added to precipitate and wait for 10 min. Finally 0.5 mL of Folin-phonol reagent was added and read 620 nm after 20 min.

2.5.11. Determination of Lactate dehydrogenase (LDH) activity

The activity of serum lactate dehydrogenase was measured by the method of King (1965). Briefly, 0.02 mL of serum was mixed with 1 mL of buffer substrate and placed in a water bath at 37°C. To that, 0.2 mL of NAD solution was added. After incubation at 15 min, 1 mL of 2,4 DPNH was added and kept in the water bath for 15 min. Finally, 10 mL of 0.4N, NAOH was added and measured the absorbance at 440 nm.

2.5.12. Determination of serum CPK activity


2.6. Statistical Analysis

Values were expressed as mean ± SD for six rats in the each group and statistical significant differences between mean values were determined by one way analysis of variance (ANOVA) followed by the Tukey’s test for multiple comparisons. The results were statistically analyzed by GraphpadInStat Software (Graphpad Software, San Diego, CA, USA) version 3 was used and p< 0.05 were considered to be significant.

3. Results

3.1. Effect of ethanolic extract of R. tetraphylla leaf on biochemical markers

Pre-treatment of ISO alone (positive control group) developed significant heart injury as evidence by a significant elevation in the biochemical markers like MDA, Troponin T, ALT, AST, LDH and CPK and depletion of GSH levels were observed when compared with negative control (group I). Oral administration of R. tetraphylla leaf extract exhibited significant reduction in the ISO induced increase in the biochemical levels and prevented the fall of GSH levels. R. tetraphylla leaf alone treated group non significant changes were observed. Obviously the simvastatin 60 mg/kg has restored all the biochemical parameter levels significantly to near normal levels. All the results were statistically significant. The results are summarized in Table 1.

3.2. Effect of ethanolic extract of R. tetraphylla leaf on tissue ALT, AST, LDH and CPK activity

Pre-treatment ISO alone (positive control group) developed significant heart injury as evidence by a significant decline in the biochemical markers like ALT, AST, LDH and CPK were observed when compared with negative control (group I). Oral administration of R. tetraphylla leaf extract exhibited significant increased in the ISO induced increase in the cardiac enzymes. R. tetraphylla leaf alone treated group non significant changes were observed. Obviously the simvastatin 60 mg/ kg has restored all the biochemical parameter levels significantly to near normal levels. All the results were statistically significant. The results are summarized in Table 2.
Table 1. Cardiac markers in plasma of normal

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group I</th>
<th>Group II</th>
<th>Group III</th>
<th>Group IV</th>
<th>Group V</th>
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<tbody>
<tr>
<td>Lipid peroxide (MDA)</td>
<td>2.82±0.19</td>
<td>6.35±0.43*</td>
<td>2.35±0.17b</td>
<td>2.52±0.17b</td>
<td>2.37±0.16b</td>
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<tr>
<td>Reduced glutathione</td>
<td>13.18±0.84</td>
<td>8.54±0.59a</td>
<td>14.02±0.96b</td>
<td>13.25±0.91b</td>
<td>12.98±0.91b</td>
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<tr>
<td>ALT</td>
<td>39.16±2.66</td>
<td>88.64±6.08a</td>
<td>34.16±2.37b</td>
<td>39.12±2.66b</td>
<td>32.76±2.15b</td>
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<td>AST</td>
<td>37.68±2.57</td>
<td>66.45±4.79a</td>
<td>34.92±2.40b</td>
<td>36.45±2.57b</td>
<td>35.29±2.38b</td>
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<tr>
<td>LDH</td>
<td>77.21±5.33</td>
<td>188.18±12.7a</td>
<td>64.47±4.59b</td>
<td>75.74±5.32b</td>
<td>81.74±5.65b</td>
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<td>CPK</td>
<td>42.46±2.96</td>
<td>76.23±5.15a</td>
<td>46.71±3.27b</td>
<td>41.28±3.02b</td>
<td>47.46±3.38b</td>
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<td>Troponin T</td>
<td>5.25±0.36</td>
<td>12.47±0.98a</td>
<td>5.82±0.48b</td>
<td>5.63±0.39b</td>
<td>6.18±0.45b</td>
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</tbody>
</table>

3.3. Effect of ethanolic extract of *R. tetraphylla* leaf on tissue GSH, lipid peroxidation SOD, CAT and GPx activity.

There was a marked depletion of GSH, SOD and CAT levels in ISO treated group. Treatment with ethanolic extract of *R. tetraphylla* leaf prevented fall in SOD, CAT and GPx activity to a near normal level. The test extract was found to be statistically significant and normalizing tissue SOD, CAT and GPx activity.

Table 2. Cardiac markers in cardiac tissues of normal and experimental groups

<table>
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<tr>
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<tr>
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<td>0.89±0.07</td>
<td>2.25±0.14a</td>
<td>0.93±0.06b</td>
<td>0.88±0.05b</td>
<td>0.97±0.06b</td>
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<tr>
<td>Reduced glutathione</td>
<td>7.30±0.50</td>
<td>4.35±0.29a</td>
<td>7.86±0.53b</td>
<td>7.65±0.53b</td>
<td>6.94±0.48b</td>
</tr>
<tr>
<td>SOD</td>
<td>5.82±0.47</td>
<td>3.11±0.21a</td>
<td>6.72±0.47b</td>
<td>6.67±0.44b</td>
<td>6.31±0.46b</td>
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<tr>
<td>Catalase</td>
<td>9.51±0.66</td>
<td>5.42±0.38a</td>
<td>8.97±0.62b</td>
<td>9.29±0.64b</td>
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<tr>
<td>GPx</td>
<td>4.84±0.33</td>
<td>2.19±0.15a</td>
<td>4.68±0.32b</td>
<td>5.02±0.35b</td>
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<td>ALT</td>
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<td>17.12±1.19a</td>
<td>38.80±2.71b</td>
<td>37.31±2.59b</td>
<td>34.17±2.39b</td>
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<td>LDH</td>
<td>45.64±3.40</td>
<td>36.71±2.42a</td>
<td>49.78±3.54b</td>
<td>50.65±3.43b</td>
<td>51.91±3.70b</td>
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<td>CPK</td>
<td>238±16.52</td>
<td>170±11.76a</td>
<td>238±16.38b</td>
<td>240.56±16.73b</td>
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<td>176±12.08</td>
<td>120±8.19a</td>
<td>171±10.76b</td>
<td>175.91±11.05b</td>
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3.4. Histopathological studies

In figure 1A revealed the myocardium intact integrity of myocardial cell membrane, myofibrillar structure with striations and continuity with adjacent myofibrils. The interstitial space appeared intact. The vascular spaces amidst these cardiac muscle fibers appeared. In Fig.1B, some of the cardiac muscle fibers show loss of integrity of myocardial cell membrane, myofibrillar structure with loss of striations and loss of continuity with adjacent myofibrils. The interstitial space at many areas appeared to be increased. The vascular spaces appeared unremarkable amidst these cardiac muscle fibers. Administration of Rauvolfia tetraphylla extract (Fig.1C) to ISO treated rat shows normal myocardium integrity of myocardial cell membrane, intact myofibrillar structure with striations and continuity with adjacent myofibrils. The extract of *R. tetraphylla* (500 mg/kg) (Fig.1D) section studied from most of the cardiac muscle fibers show intact of integrity of myocardial cell membrane, myofibrillar structure with striations and continuity with adjacent myofibrils. The interstitial space at focal areas appeared to decreased. Standard drug simvastain (60 mg/kg) treated heart (Fig.1E) section studied from most of the cardiac muscle fibers show intact of integrity of myocardial cell membrane, myofibrillar structure with mild loss of striations and loss of continuity with adjacent myofibrils. The interstitial space at focal areas appeared to decreased.

Treatment with 60 mg/kg simvastain, the standard drug, prevented the depletion of SOD, CAT and GPx activity. The levels of lipid peroxidation were restored to near normal levels by pretreatment with ethanolic extract of *R. tetraphylla* as compared to positive control group. *R. tetraphylla* leaf alone treated group non significant changes were observed. All the results were statistically (p<0.05). The results are summarized in Table 2.

3.4. Histopathological studies

In figure 1A revealed the myocardium intact integrity of myocardial cell membrane, myofibrillar structure with striations and continuity with adjacent myofibrils. The interstitial space appeared intact. The rat shows normal myocardium integrity of myocardial cell membrane, intact myofibrillar structure with striations and continuity with adjacent myofibrils. The extract of *R. tetraphylla* (500 mg/kg) (Fig.1D) section studied from most of the cardiac muscle fibers show intact of integrity of myocardial cell membrane, myofibrillar structure with striations and continuity with adjacent myofibrils. The interstitial space at focal areas appeared to decreased. Standard drug simvastain (60 mg/kg) treated heart (Fig.1E) section studied from most of the cardiac muscle fibers show intact of integrity of myocardial cell membrane, myofibrillar structure with mild loss of striations and loss of continuity with adjacent myofibrils. The interstitial space at focal areas appeared to decreased.
The present study demonstrated both important curative and preventive modes of cardioprotective activity. It explained the cardioprotective potential of leaf extract of *R. tetraphylla* widely used isoproterenol (ISO) induced model of MI in rats. The MI is produced due to its action on the cardiac β1-receptors. ISO-induced myocardial necrosis showed membrane permeability alterations, which bring about the loss of function and integrity of myocardial membrane. A number of studies are available that suggest the crucial role of free radicals in pathogenesis of ISO-induced myocardial damage (Ojha et al., 2011). ISO-induced MI serves as a well-standardized model to study the beneficial effects of many drugs and cardiac function since it mimics the clinical conditions of MI due to ischemia in humans. Hence ISO induced MI model was used in this study.

The diagnosis of MI is based on clinical symptoms, electrocardiographic (ECG) changes and characteristic pattern of changes in some serum enzymes such as creatine kinase (CK), lactate dehydrogenase (LD) and cardiac specific proteins like troponins (Alpert et al., 2000). The serum markers of myocardial injury are used to help in establishing the diagnosis of MI. ALT, AST, LDH and CPK were present in cardiac muscle, injury to these tissues results in the release of the enzyme into the bloodstream. Increased levels are found in MI. Increased levels of ALT, AST, LDH and CPK in serum and decreased level in heart tissues were due to the leakage of these enzymes from heart tissues to blood as a result of necrosis induced by ISP in rats (Manjula et al., 1992). The myocardial cell necrosis can be due to increase in lipid peroxidation. These findings are in agreement with earlier reports (Karthikeyan et al., 2007). Treatment of *Rauvolfia tetraphylla* extract (500 mg/Kg body weight) significantly blocked the ISO-induced secretion of all cardiac diagnostic marker enzymes (CK, LDH, AST, ALT). The decline in enzymes levels could be due to potential of extract for repairing and maintenance of the membrane due to antioxidant polyphenols, thereby preventing the secretion of enzymes due to membrane stabilizing property. Our results are concur with the earlier work done by Gomathi et al (2014) studies.

Lipid peroxidation, a type of oxidative deterioration of polyunsaturated fatty acids has been linked with altered membrane structure and enzyme inactivation. Increased lipid peroxidation products in isoproterenol induced cardio toxic rats appear to be the initial stage to the tissue making it more susceptible to oxidative damage. Increased free radical production may be responsible for the observed membrane damage as evidenced by the elevated lipid peroxidationin terms of thiobarbituric acid reactive substances and lipid hydroperoxides. Plant pretreatment decreased the levels of lipid peroxides in the plasma and heart of isoproterenol induced cardio toxic rats. This shows the anti-lipoperoxidative effect of *R. tetraphylla*. Present finding is in agreement with Gomathi et al (2014) studies.

**Fig. 1A.** Normal architecture of cardiac muscle

**Fig. 1B.** ISO treated cardiac muscle of rat.

**Fig. 1C.** *R. tetraphylla* leaf alone treated cardiac muscle of rat.

**Fig. 1D.** ISO and *R. tetraphylla* treated cardiac muscle of rat

**Fig. 1E.** ISO and standard treated cardiac muscle of rat.
Free radical scavenging enzymes such as catalase, superoxide dismutase and glutathione peroxidase are the first line cellular defense against oxidative injury, decomposing $O_2$ and $H_2O_2$ before interacting to form the more reactive hydroxyl radical (OH\textsuperscript{–}). The equilibrium between these enzymes is an important process for the effective removal of oxygen stress in intracellular organelles. Glutathione plays an important role in the regulation of variety of cell function and in cell protection from oxidative injury. In the present study, significant reduction in the activities of glutathione-dependent antioxidant enzyme (GPs) and antiperoxidative enzymes (SOD and CAT) with aconcomitant decline in the level of reduced glutathionewas observed in the heart tissue of group III myocardial infarcted rats as compared to group I normal control animals, reflecting an increased oxidative stress in ISO induced myocardial infarction. This is in accordance with previous investigations, which indicated that the tissue antioxidant status was being operated at diminished level in ISO induced MI condition (Jay Rabadia et al., 2014).

Depletion of GSH results in enhanced protective mechanism towards oxidative stress in MI may consume reduced glutathione and depress reduced glutathione levels. The observed decrease in reduced glutathione levels might be due to increased utilization in protecting thiol containing proteins from lipid peroxides and from other reactive oxygen species. Lowered activities of these antioxidant enzymes may lead to the formation of $O_2$ and $H_2O_2$, which in turn can form hydroxyl radical (OH\textsuperscript{–}) and bring about a number of reactions harmful to the cellular and sub-cellular membranes in the heart tissue. Reduction noticed in the activities of the antiperoxidative enzymes in ISO -induced myocardial infarction might be due to the increase degeneration of reactive oxygen radicals such as superoxide and hydrogen peroxide, which in turn lead to the inactivation of these enzyme activities (Shiny et al., 2005; Gomathi et al., 2014). Our result agrees with the earlier report (StanelyMainzen Prince et al., 2009).

Data from human studies have suggested that an inverse correlation exists between plasma levels of vitamin E and mortality from IHD (Roy et al., 1994). Ascorbic acid present in aqueous environment has multiple antioxidant properties including the ability to regenerate alpha tocopherol radicals present at the surface of the membrane (Karthikeyan and Rani 2003). Due to the above properties of the two antioxidants, vitamins E and C have been utilized more for the neutralization of ISO mediated free radicals and lipid peroxidation process and hence a decreased level of heart vitamins E and C was observed in ISO administered rats. It is of interest that the supplementation of Rauwolfia tetraphylla produced a marked increase in Vitamin C and Vitamin E levels in ISO administered rats. Prabhu et al (2007) have reported that administration of mangiferin to ISO rats improves the antioxidant vitamins thereby reducing free radical mediated lipid peroxidation which contributes to the protective effect against MI.

5. Conclusion

In conclusion, the present results endorse our hypothesis that leaf extract of R. tetraphylla has cardio protective potential. R. tetraphylla leaf extract pretreatment improved cardiac functions, the effect which can be attributed to its ability of maintaining redox status which is disturbed by ISO challenge, via restoration of endogenous antioxidants, controlling lipid peroxide formation and preserving cardiac marker enzyme activities of CK, LDH, AST and ALT. Preservation of histo architecture of myocyte by R. tetraphylla pretreatment reconfirms these effects. The possible mechanisms action of R. tetraphylla due to the presence of phytochemicals as phenolic groups that might directly or indirectly involved in its cardioprotective effects.

Conflict of interest statement

We declare that we have no conflict of interest.

References