Asian Journal of Chemistry; Vol. 30, No. 7 (2018), 1579-1584

ASIAN JOURNAL OF CHEMISTRY

https://doi.org/10.14233/ajchem.2018.21258

Isolation and Characterization of Flavonoid from Methanolic Extract of *Mirabilis jalapa* Linn. Tuber and Evaluation of its Cardioprotective Property

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Received: 30 January 2018; *Accepted*: 15 March 2018; *Published online*: 31 May 2018; AJC-18934

The aim of the study is to isolate and characterization of bioconstituent present in methanolic extract of *Mirabilis jalapa* L. tuber and also evaluate the cardioprotective property of the extract as well as isolated compound from the extract. The bioactive constituent flavonoid was isolated from methanolic extract through flash chromatography technique by using solvent system ethyl acetate and methanol. The chemical structure of compound was confirmed on the basis of spectroscopy analysis and identified as 2-(3′, 4′-dihydroxy phenyl)-3,5,7-trihydroxy chromen 4-one. Administration of *Mirabilis jalapa* extract in higher dose 200 and 400 mg/kg b.w. and isolated compound to doxorubicin-intoxicated rats demonstrated prominent reduction in serum biomarker enzymes, normalization of serum lipid profiles. Also, significant modulation of malondialdehyde (MDA), endogenous non-enzymatic (GSH) and enzymatic (SOD and CAT) antioxidant and detoxification systems compared to doxorubicin control rats. This was achieved due to presence of flavonoid in the methanolic extract of *Mirabilis jalapa* L. tuber.

Keywords: *Mirabilis jalapa* L. tuber, Flavonoid, Doxorubicin, Cardioprotective.

INTRODUCTION

Mirabilis jalapa Linn. commonly known as Marvel of Peru or four o' clock flower belonging to the family Nyctaginaceae is the herbaceous plant. Traditionally the plant parts are used as antidysenteric, antiparasitic, carminative, digestive stimulant, tonic, vermifuge, wound healer, cathartic, *etc*. [1]. The roots and tubers have aphrodisiac, diuretic and purgative properties [2]. In Ayurvedic system the plant is well known for its use in treating boils, inflammations, constipation, diabetes, urinary disorders and hypertension. Dried flowers are used as a snuff for headaches, fungal infection and root decoction to wash wounds, treat skin afflictions as leprosy [3,4].

It is reported that the root of *Mirabilis jalapa* contain phyto-constituents such as alkaloids, glycosides, phytosterol and phenolic compounds [5]. But the evidences were found in the literature on the root part of this plant which is inadequate to consider as standard. Therefore, the effort was carried out to isolate and characterize the bioactive constituent from the methanolic extract of *Mirabilis jalapa* L. tuber and also establish the cardiopotective property of the tuber, which claimed by folk people of north east India. To establish cardiopotective property, the anticancer drug doxorubicin was used as positive control to produce cardiotoxicity in rats. It can leads to congestive heart failure, increase free radical formation, increase chlolesterol, triglyceride, low-density lipoprotein (LDL), decrease activity of Na^+-K^+ adenosine triphosphate, *etc*. [6].

EXPERIMENTAL

Tubers of *Mirabilis jalapa* were collected from local area of Kamrup district, India and authenticated by Dr. P.P. Baruah, Department of Botany, Gauhati University, India. The voucher specimen of *Mirabilis jalapa* tuberous root (Acc no. 18176) were kept at the Department of Pharmacognosy, Girijananda Chowdhury Institute of Pharmaceutical Sciences, Guwahati, India for future reference. The plant material was process and subjected to washing with running water, dried in shade for 10- 15 days and stored in air tight containers until used.

Preparation of extract: The dried tubers were subjected to size reduction. About 1 kg of crude powder drug was subjected to hot percolation with petroleum ether for 72 h to take out fatty material followed by cold maceration at room temperature with different solvent by increasing polarity such as benzene, diethyl ether, chloroform, acetone, ethyl acetate and 95 % methanol for about 48 h with frequent shaking up to 6 h. Extracts were then filtered, concentrated under reduced pressure in rotary evaporator (Buchi India Pvt. Ltd.) at 40 ºC and stored

in a desiccator until further compare to all other extracts methanolic extract showed the maximum quantity *i.e.* 10 g [7]. All the extracts were subjected to quantitative chemical tests as describe in reference books to identify the presence of different metabolites [7,8].

Isolation of bioactive constituents: Sample of each extracts were chromatographed on TLC [9] and found that methanolic extract showed the presence of flavonoid when eluted with ethyl acetate:methanol (7:3). For the separation of flavonoid, about 10 g of sample of *Mirabilis jalapa* tuber was further chromatographed in flash chromagraphy by using same eluent as TLC [10]. The fractions with same R_f value on TLC were pooled together and evaporated to dryness, which was crystallized to give fine powdered crystals. The structure of the isolated compound has been elucidated by DSC, UV, mass, FTIR, CHN analysis and NMR spectra.

Experimental animal: Experimental works were carried out with the guidelines set by CPCSEA and were approved by the Institutional Animal Ethical Committee (GIPS/IAEC/Phd/2015/01). Female swiss albino mice of weighed between 20-25 g were housed and the standard conditions *viz*. 22 ± 3 ºC, 50 ± 10 % humidity, 12 h interval light and dark phase were properly maintained and standard diet were followed as per the CPCSEA guideline.

Acute toxicity study: The methanolic extract of *Mirabilis jalapa* tuber was administered at of different doses of 2000 and 5000 mg/kg bw p.o. to overnight fasted experimental mice as suggested in OECD Guidelines 425 [11]. The animal behavioural changes and mortality, abnormalities were observed next 24 h and then recorded upto 14 days.

Selection of dose: After performing the acute toxicity studies, it was found that there was no mortality at the dose of 2000 mg/kg b.w. as well as 5000 mg /kg b.w. of *Mirabilis jalapa* tuber. Therefore, dose optimization was done and 100, 200 and 400 mg/kg p.o. were selected for the experimental study. For the oral dose optimization of isolated compound of *Mirabilis jalapa* tuber, the dose were calculated based on the percentage yield of isolated compound of *Mirabilis jalapa* (2.78 % w/w) calculated in terms of the minimum effective dose of *Mirabilis jalapa* tuber (200 mg/kg, p.o.) that produced significant cardioprotective activity [12]. Therefore, in the present study, dose for the oral administration of isolated compound of *Mirabilis jalapa* was selected at (5.56 mg/kg p.o.) for the experimental study.

Doxorubicin induced cardiotixicity in rats: Thirty six number of rats were divided into 6 groups (n = 6). Group I were served as doxorubicin control (dox control) received 5 mg/kg bw i.p. Group II serve as normal control receive normal saline at 5 mL/kg orally. Group III, IV and V were given *Mirabilis jalapa* tuber and group VI was given isolated compound from *Mirabilis jalapa* at the dose 100, 200, 400 mg/kg and 5.56 mg/kg b.w./d/p.o accordingly for 21 days. Except group II doxorubicin was administered to all groups of animals in three equal injection on the 7th, 14th, 21st days for a total cumulative dose of 15 mg/kg b.w. i.p. [13].

Biochemical estimation from serum: After 24 h, the last treated blood sample was collected by cardiac puncture from all groups of rats. Serum sample were separated for the estimation of the presence of different enzymes such as lactate dehydrogenase (LDH) [14], creatine phosphokinase (CPK) [15], aspartate transaminase (AST) [16], alanine transaminase (ALT). Lipid profile like total cholesterol, total triglycerides (TGL), high density lipoproteins (HDL) and low density lipoproteins (LDL) [17]. All the analyses were performed with commercially available kits based on the references using analyzer [18].

Preparation of heart tissue for estimation of oxidative stress marker: After collection of blood all the rats were sacrificed by cervical dislocation, the heart tissues were dissected immediately and homogenized with chilled 0.05 M phosphate buffer for 10 times at pH 7.4 in Teflon glass homogenizer and the homogenate was centrifuged at 1000 rpm 4 ºC for 3 min and the supernatant divided into two portions [19], one of which was used for measurement of lipid peroxidation (LPO) [20]. The remaining supernatant was again centrifuged at 12,000 rpm at 4 ºC for 15 min and used for the measurement of superoxide dismutase (SOD) [21], catalase (CAT) [22] and glutathione (GSH) [23].

Histopathological studies: Hearts were removed from the experimental animals and kept in 10 % formalin solution. The isolated heart is then processed for section cutting is about 5 µm thickness by using microtome and stained with hematoxylin and eosin (H and E) reagents and observed by light microscope to evaluate myocardium injury [13].

Statistical analysis: Results were expressed as mean value ± SEM. Data were analyzed incorporating GraphPad Prism 5 software and all statistical comparison were made using one way analysis of variance (ANOVA) followed by Dunnet's test post hoc analysis. Results were articulated where P values < 0.05 was considered statistically significant as compared to control group.

RESULTS AND DISCUSSION

The present study was aimed to isolate and characterized the bioactive constituent from methanolic extract of *Mirabilis jalapa* tuber. It was observed that methanolic extract produces the maximum percentage yield (15 %) of the extract followed by ethyl acetate (12 %) and chloroform (9.2 %) as solvents. The results from the preliminary phytochemical screening of different extract of *Mirabilis jalapa* tuberous root showed the presence of phyto-costituents like alkaloids, carbohydrate, glycosides, steroids, saponins, phenolics and flavonoids.

Isolation and characterization of compound: The fractions no. 25-38 obtained from flash chromatography showed the same R_f value (R_f = 0.33) on TLC with yellowish orange colour on spraying of ferric chloride reagent which was then crystallized from methanol to give fine yellow powdered crystals and also showed reddish pink colour with Mg+HCl (Sinoda reagent), which identified as flavonoid compound. The yield is 2.78 % w/w, m.p. is 317 ºC, ESI-MS spectrum (positive ion mode) showed the presence of base peak at *m/z* 301.12 indicating the molecular weight of compound is 301.

Infrared spectrum of the compound confirmed the presence of -OH group (3253 cm^{-1}), conjugated carbonyl group (1661 cm^{-1}) and aromatic C-C (1605 cm^{-1}). CHNS analysis revealed that carbon found to be 52.208 %, hydrogen found to be 4.181 % and oxygen found to be 43.61 %.

The compound showed strong absorption at 365 nm in its spectrum implied the presence of phenolic aromatic rings.

TABLE-1
UV SPECTRAL DATA OF COMPOUND ISOLATED FROM METHANOLIC EXTRACT OF TUBEROUS ROOT OF *Mirabilis jalapa* Linn

Preparation	λ_{max} (nm) of compound	Spectral effect	Structural diagnosis
Methanolic solution of compound	365, 257	–	Flavone-3-ol
Methanolic solution of compound + 3 drops of sodium methoxide solution	400	35 nm shift in Band I	4'-C=O
Methanolic solution of compound + 6 drops of aluminium chloride	385,263	20 nm Shift in Band I	5-OH Free
Methanolic solution of compound + 6 drops of aluminium chloride and 3 drops of hydrochloric acid	390, 268	25 nm shift in Band I	Presence of di-OH in B ring
Methanolic solution of compound + powdered NaOAc	257	Lack of shift in Band II	Absence of free 7-hydroxyl group

Note = Structural analysis was done on basis of reference literature for flavonoid.

The methanolic solution of compound exhibited typical UV absorption characteristics after the addition of various shifting reagents. The UV spectrum showed absorption bands reagents shifts of the compound to be a 7-substituted derivative. The absence of free 7-hydroxyl group in the compound was observed in lack of shift of band II in the presence of NaOAc (Table-1).

The ^{1}H NMR spectrum (Table-2) of the compound was exhibited the singlet at 2.50 ppm indicated the presence of 1-H at C-6 position. The singlet peak at 3.53 ppm indicates the presence of 1-H at C-8 position. The presence of lone pair electron on the oxygen atom of OH ion make the OH proton deshielded as a result the OH peak appeared at down field. The double peak at 6.19 ppm is due to the C-OH proton at C-3 position (J = 1.03MHz) again two doublet at 6.42 and 6.90 ppm are due to presence of OH at C-5 and C-7 position (J = 1.06 and 1.04), respectively. The multiplet at 7.56 ppm is due to 2′ 1H of the phenyl ring substituted at 2-position of the coumarin moiety (J = 1.00 MHz). The doublet at 7.68 ppm is due to one proton present in the substituted phenyl ring at C-5′ position. Singlet at 9.32 ppm is due to the 1H at C-6′ position. The singlet peak at 10.81ppm and 12.48 ppm are due to presence of C-OH proton at C-3′ and C-4′ position.

TABLE-2
^{1}H AND ^{13}C NMR SPECTRAL DATA OF ISOLATED COMPOUND FROM METHANOLIC EXTRACT OF *Mirabilis jalapa* Linn. TUBER

Position of H/C	Department	Value of δ_H	Values of δ_C
2	C		93.33
3	C-OH	6.19	135.68
4	C=O		102.98
5	C-OH	6.90	145
6	CH	2.50	98.15
7	C-OH	6.42	146.76
8	CH	3.53	115.03
9	C		115.57
10	C		119.97
1'	C		121.94
2'	CH	7.56	147.64
3'	C-OH	10.81	163.82
4'	C-OH	12.48	175.78
5'	CH	7.68	156.11
6'	CH	9.32	160.57

^{13}C NMR spectrum (Table-2) of the compound revealed the presence of 15 carbons. Signal at δ 175.9 in the ^{13}C NMR spectrum indicates the presence of conjugated carbonyl group in the compound. The prominent peak at 93.33, 98.15 coresponse to the carbon atom at position 2 and 6, respectively. The peak at 102.98 corresponds for C-(C=O) at positon 4, the peak at 135.68, 145 and 146.76 corresponds to carbon atoms (C-OH) at position 3, 5, 7, repectively. The prominent peak at 115.03 corresponds to the carbon atom at position 8. The peaks at 115.57 and 119.97 are due to the fused carbon between the two rings of chromen nucleus at position 9 and 10. The prominent peak at 163.82 and 175.78 is due to the C atom (C-OH) at position 3′ and 4′ of substituted phenyl ring. Peak at 121.94 depict the C-atom at 1′ position. Finally, peak at 174.64, 156.11 and 160.57 is due to the carbon atom (CH) at position 2′, 5′, 6′, repectively.

On the basis of physical properties and spectroscopic data, structure of compound was identified as 2-(3′,4′-dihydroxy phenyl)-3,5,7-trihydroxy chromen 4-one.

Cardioprotective activity of methanolic extract and isolated compound of *Mirabilis jalapa* tuber: Doxorubicin produced cardiotoxicity by chronic administration [24]. In present study, it was established that the different doses of *Mirabilis jalapa* and isolated product of *Mirabilis jalapa* were revealed the significant increase in cardiac biomarker enzymes and endogenous antioxidants and heart tissue histolopathology.

Cardiac tissue is especially susceptible to free radical injury, because of the lower activities of the free radical detoxifying mechanisms, such as SOD, CAT and GSH [25]. Further, doxorubicin also has a high affinity for the phospholipid component of the mitochondrial membrane in cardiac myocytes, leading to selective accumulation of doxorubicin in the heart tissue [26,27]. The doxorubicin-induced mitochondrial injury is critical to the heart because it would presumably have extreme adverse effects on the contractile functioning of the cardiac myocytes by producing alterations in the energy metabolism [28,29].

Serum lipid levels: The result shown that animal treated with DOX (5 mg/kg b.w. i.p.) stimulated cardiotoxicity and showed the increase levels of cholesterol, triglyceride and LDL and reduce level of HDL when compared with normal control. However, it was also observed that *Mirabilis jalapa* tuber treated groups with dose at 200 and 400 mg/kg b.w. and isolated product at 5.56 mg/kg bw significantly (*$p < 0.05$) decreased of level of cholesterol, triglyceride and LDL. But only the dose of 400 mg/kg b.w. *Mirabilis jalapa* increased the level of HDL when compared with doxorubicin control group where as dose 100 mg/kg b.w. was found to be not significant compared to high doses (Table-3).

Serum enzyme biomarkers: Animals treated with doxorubicin (5 mg/kg b.w. i.p.) reported the increase in the levels of

TABLE-3
EFFECT OF *Mirabilis jalapa* TUBER AND ITS ISOLATED PRODUCT ON CHOLESTEROL, TRIGLYCERIDES, HDL AND LDL LEVEL IN DOXORUBICIN TREATED RAT (MEAN ± SEM, n = 6)

Groups	Treatment	Cholesterol (mg dL^{-1})	Cholesterol decrease (%)	Triglyceride (mg dL^{-1})	Triglyceride decrease (%)	HDL (mg dL^{-1})	HDL increase (%)	LDL (mg dL^{-1})	LDL decrease (%)
I	Doxorubicin (5 mg/kg) control	286.27 ± 34.25	–	205.69 ± 26.75	–	12.69 ± 2.93	–	185.87 ± 25.93	–
II	Normal control	88.60 ± 5.96***	69.00	69.76 ± 3.18***	66.08	46.73 ± 6.26***	268.24	52.14 ± 8.45***	71.94
III	*Mirabilis jalapa* (100 mg/kg) + Doxorubicin	244.21 ± 28.96ns	14.69	183.14 ± 21.37ns	10.96	13.80 ± 4.98ns	8.74	172.65 ± 26.54ns	7.11
IV	*Mirabilis jalapa* (200 mg/kg) + Doxorubicin	177.71 ± 22.27**	37.92	133.45 ± 17.97*	35.98	21.78 ± 5.56ns	71.63	104.64 ± 17.55*	43.70
V	*Mirabilis jalapa* (400 mg/kg) + Doxorubicin	99.72 ± 8.03***	65.16	82.76 ± 8.93***	59.76	35.53 ± 4.56*	179.98	65.70 ± 12.41***	64.65
VI	Isolated product of *Mirabilis jalapa* (5.56 mg/kg) + Doxorubicin	127.85 ± 9.16***	55.33	108.67 ± 5.94**	47.16	28.16 ± 6.31ns	121.90	88.28 ± 13.71**	52.50

All results are expressed as Mean ± SEM (n = 6 in each group). Statistical comparison was determined by one way ANOVA followed by the Dunnett's comparison tests. *p < 0.05, **p < 0.01, ***p < 0.001 significant; ns = not significant when compared with control.

creatine phosphokinase (CPK), LDH, aspartate transaminase (AST) and alanine transaminase (ALT) as compared to normal control group. After treatment with *Mirabilis jalapa* at higher dose of 200 and 400 mg/kg b.w. as well as isolated product of *Mirabilis jalapa* (5.56 mg/kg b.w.), the level of creatine phosphokinase (CPK), LDH, aspartate transaminase (AST) and alanine transaminase (ALT) significantly restored towards normal in dose dependent manner where as dose 100 mg/kg bw was found to be not significant compared to high doses (Table-4). This could be due to a protective or membrane-stabilizing effect of extract and isolated compound on the myocardium, reducing the cardiac damage, and thereby restricting the leakage of these enzymes [30,31].

Antioxidant status: Effect of doxorubicin on tissue lipid peroxidation, antioxidant and antioxidant enzymes are shown in Table-5. The malondialdehyde (MDA) level was increased whereas endogenous non-enzymatic (GSH) and enzymatic (SOD and CAT) levels were significantly decreased in doxorubicin treated group as compared to normal animals. At higher dose of *Mirabilis jalapa* at 200 and 400mg/kg b.w. and isolated compound of *Mirabilis jalapa* at 5.56 mg/kg b.w. produced significant decrease in the level of MDA and increase the level of GSH and CAT as compared with doxorubicin control group but level of SOD significant decrease with *Mirabilis jalapa* at 400 mg/kg b.w. dose when compared with doxorubicin control group.

The present study has shown that administration of extracts at high doses and isolated compound efficiently counter acted the doxorubicin induced cardiac tissue damage by significant decrease in MDA and elevated the GSH content to near-normal levels, which prevented degradation of cellular macromolecules and thus cell disruption, probably by decreasing Ca^{2+} influx [32] and also increase in GSH, SOD and CAT levels [33]. These results indicated the protective effect of *Mirabilis jalapa* and isolated compound on doxorubicin-induced cardiotoxicity by boosting the endogenous non-enzymatic and enzymatic antioxidant systems, which entailed scavenging of oxidative free radicals.

Histopathological observation: Histopathological assessment of heart tissue are represented in Fig. 1. The results illustrated that the heart tissue from normal control treated animal showed

TABLE-4
EFFECT OF *Mirabilis jalapa* TUBER AND ITS ISOLATED PRODUCT ON CPK, LDH, AST AND ALT IN DOXORUBICIN TREATED RAT (MEAN ± SEM, n = 6)

Groups	Treatment	CPK (IU L^{-1})	CPK decrease (%)	LDH (IU L^{-1})	LDH decrease (%)	AST (IU L^{-1})	AST decrease (%)	ALT (IU L^{-1})	ALT decrease (%)
I	Doxorubicin (5 mg/kg) control	356.66 ± 37.28	–	285.79 ± 28.10	–	128.87 ± 17.94	–	89.60 ± 12.20	–
II	Normal control	94.49 ± 9.12***	73.50	122.46 ± 16.83***	57.15	42.54 ± 6.25***	66.98	28.67 ± 5.69***	68.00
III	*Mirabilis jalapa* (100 mg/kg) + Doxorubicin	298.30 ± 41.58ns	16.36	201.45 ± 32.84ns	29.51	125.77 ± 19.50ns	2.40	61.71 ± 8.68 ns	31.12
IV	*Mirabilis jalapa* (200 mg/kg) + Doxorubicin	191.74 ± 35.07**	46.24	161.26 ± 23.71**	43.57	82.47 ± 8.43*	36.00	45.03 ± 6.08**	49.74
V	*Mirabilis jalapa* (400 mg/kg) + Doxorubicin	111.98 ± 14.59***	68.60	131.91 ± 14.71***	53.84	52.55 ± 6.57***	59.22	33.74 ± 3.75***	62.34
VI	Isolated product of *Mirabilis jalapa* (5.56 mg/kg) + Doxorubicin	161.74 ± 9.65***	54.65	148.46 ± 9.73***	48.05	69.53 ± 5.46**	46.04	41.57 ± 5.64 ***	53.60

All results are expressed as Mean ± SEM (n = 6 in each group). Statistical comparison was determined by one way ANOVA followed by the Dunnett's comparison tests. *p < 0.05, **p < 0.01, ***p < 0.001 significant; ns = not significant when compared with control.

TABLE-5
EFFECT OF *Mirabilis jalapa* TUBER AND ITS ISOLATED PRODUCT ON MDA, GSH, AND SOD LEVEL IN DOXORUBICIN TREATED RAT (MEAN ± SEM, n = 6)

Groups	Treatment	MDA (n mol/g of wet tissue)	MDA decrease (%)	GSH (µg/g of wet tissue)	GSH increase (%)	CAT (unit/mg of protein)	CAT increase (%)	SOD (unit/mg of protein)	SOD increase (%)
I	Doxorubicin (5 mg/kg) control	65.19 ± 8.38	–	1.23 ± 0.047	–	16.04 ± 3.42	–	11.92 ± 2.92	–
II	Normal control	11.77 ± 3.04***	81.94	3.96 ± 0.42***	221.95	48.76 ± 5.88***	203.99	32.22 ± 4.834***	170.30
III	*Mirabilis jalapa* (100 mg/kg) + Doxorubicin	60.15 ± 9.25ns	7.73	1.26 ± 0.34 ns	2.43	16.43 ± 2.23ns	2.43	12.08 ± 1.16ns	1.34
IV	*Mirabilis jalapa* (200 mg/kg) + Doxorubicin	38.79 ± 5.82*	32.82	2.52 ± 0.45 *	104.87	22.58 ± 3.43ns	40.77	17.76 ± 2.37 ns	48.99
V	*Mirabilis jalapa* (400 mg/kg) + Doxorubicin	21.77 ± 4.76***	66.60	2.94 ± 0.31 **	139.02	35.55 ± 4.60*	121.63	24.47 ± 2.56*	105.28
VI	Isolated product of *Mirabilis jalapa* (5.56 mg/kg) + Doxorubicin	28.70 ± 3.74**	55.59	2.56 ± 0.24 *	108.13	32.96 ± 5.63*	105.48	19.48 ± 4.13 ns	63.42

All results are expressed as Mean ± S.E.M (n = 6 in each group). Statistical comparison was determined by one way ANOVA followed by the Dunnett's comparison tests. *p < 0.05, **p < 0.01, ***p < 0.001 significant; ns = not significant when compared with control (Doxorubicin 5 mg/kg).

normal morphological appearances whereas doxorubicin treated animal caused disorganization of normal cellular structure of heart like loss of myofibrils, vacuolization of the cytoplasm, inflammatory cell infiltration or necrosis were observed. On the other hand, the treatment with *Mirabilis jalapa* at dose 200 and 400 mg/kg b.w. as well isolated compound *Mirabilis jalapa* (5.56 mg/kg b.w.) shown significantly reduced the histopathological changes in the cardiac tissue took place due to doxorubicin (control) admistration and recovered almost normal histological appearance of cardiac muscle similar to that of normal control. The dose at 100 mg/kg b.w. did not show any significant effect on doxorubicin treated animal.

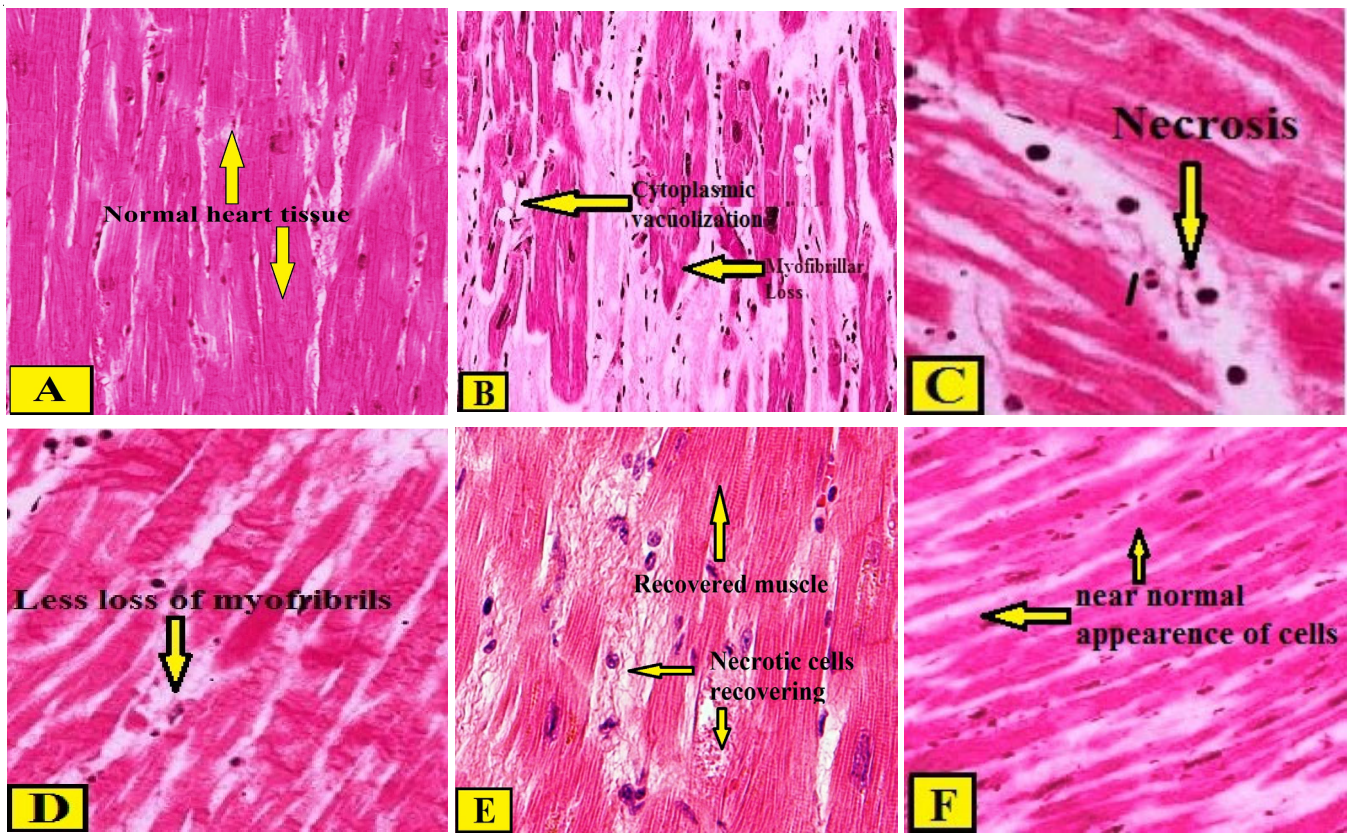

Fig. 1. Histological section [10× and 40× magnification] of the cardiac tissue. Control groups (A) presented normal architecture of cardiac tissue. Doxorubicin-treated group (B) shown degenerative changes including myofibrillar loss, cytoplasmic vacuolization, inflammatory cell infiltration. Extract dose at 100 mg/kg bw (C) shown almost similar appearance that of doxorubicin control (B). Extract treatment groups with dose 200 mg/ kg bw (D) and 400 mg/kg bw (E) and isolated compound (F) at dose 5.56 mg/kg bw shown significantly reduced doxorubicin-induced degenerative changes and recovered the cardiac cells similar to that of normal control (A)

Conclusion

Flavonoid (2-(3′,4′-dihydroxy phenyl)-3,5,7-trihydroxy chromen-4-one was isolated and characterized from methanolic extract of *Mirabilis jalapa* tuber. Therefore, from the results it can be stated that the cardioprotective activity of plant part of *Mirabilis jalapa* is due to the presence of flavonoid.

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