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Anti-fibrotic Actions of Roselle Extract in Rat Model of Myocardial Infarction

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Abstract

Heart failure-associated morbidity and mortality is largely attributable to extensive and unregulated cardiac remodelling. Roselle (*Hibiscus sabdariffa*) calyces are enriched with natural polyphenols known for antioxidant and anti-hypertensive effects, yet its effects on early cardiac remodelling in post myocardial infarction (MI) setting are still unclear. Thus, the aim of this study was to investigate the actions of roselle extract on cardiac remodelling in rat model of MI. Male Wistar rats (200–300 g) were randomly allotted into three groups: Control, MI, and MI + Roselle. MI was induced with isoprenaline (ISO) (85 mg/kg, s.c) for two consecutive days followed by roselle treatment (100 mg/kg, orally) for 7 days. Isoprenaline administration showed changes in heart weight to body weight (HW/BW) ratio. MI was especially evident by the elevated cardiac injury marker, troponin-T, and histological observation. Upregulation of plasma levels and cardiac gene expression levels of inflammatory cytokines such as interleukin (IL)-6 and IL-10 was seen in MI rats. A relatively high percentage of fibrosis was observed in rat heart tissues with over-expression of collagen (Col)-1 and Col-3 genes following isoprenaline-induced MI. On top of that, cardiomyocyte areas were larger in heart tissues of MI rats with upregulation of atrial natriuretic peptide (ANP) and brain natriuretic peptide (BNP) gene expression, indicating cardiac hypertrophy. Interestingly, roselle supplementation attenuated elevation of plasma troponin-T, IL-6, IL10, and gene expression level of IL-10. Furthermore, reduction of cardiac fibrosis and hypertrophy were observed. In conclusion, roselle treatment was able to limit early cardiac remodelling in MI rat model by alleviating inflammation, fibrosis, and hypertrophy; hence, the potential application of roselle in early adjunctive treatment to prevent heart failure.

Keywords Roselle · Isoprenaline · Fibrosis · Inflammation · Hypertrophy · Cardiac remodelling

Introduction

Ischemic diseases, particularly MI, remains as a major concern as it imposes great burden to public health and economy by causing the highest number of deaths globally [1]. In Malaysia, MI alone accounted for 13.2% of total death for the year of 2016 [2]. Prolonged imbalance between myocardial oxygen supply and demand could lead to MI [3]. Poor MI prognosis is often associated with heart failure and is attributable largely to extensive and unregulated cardiac remodelling [4]. Cardiac remodelling comprises of several molecular and cellular changes which then contribute to alteration in structure, size, shape, and function of the heart [5]. Furthermore, the extent of cardiac remodelling following MI would predict the mortality and morbidity of MI patients [6], besides escalates the risk of death by at least three to fourfolds [7]. Some of the important structural event after MI is left ventricular (LV) remodelling. Underlying mechanism of LV remodelling are multifactorial such as oxidative stress, inflammation, hypertrophy, and fibrosis [6].

Post infarct inflammatory response is essential for physiological repair at damage site [8]. However, exuberant inflammatory response may have resulted in deleterious effect by extending necrosis and increasing the rate of cardiomyocytes death [9]. Since cardiomyocytes are non-regenerative, thus

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post infarct immense loss of cardiomyocytes burdens myocardium. Loss of viable cardiomyocytes is replaced with fibrosis by deposition of extracellular matrix (ECM) to preserve the structural integrity of heart, avoiding cardiac rupture [8]. However, unregulated cardiac fibrosis stiffens and hardens the heart wall which in turn reduces contractility of heart and results in declining heart functions [10].

Isoprenaline (ISO)-induced MI is an established and widely used model to study cardiac remodelling. ISO, an adrenergic agonist stimulates overload of intracellular calcium, positive inotropic, and chronotropic effect. These result in increased oxygen demand in myocardium, leading to ischemia and infarction. Upon administration, ISO undergoes auto-oxidation, generating highly cytotoxic free radical accumulation which then stimulates peroxidation of phospholipid membrane, causing severe damage to myocardial membrane [11]. Concomitantly, previous studies showed that ISO-induced MI rats model exhibited characteristics of inflammation [12], cardiac fibrosis [13, 14] and hypertrophy [15] which eventually triggers LV remodelling [16]. Several lines of evidence have shown that subcutaneous injection of ISO at 85 mg/kg produces subendocardial myocardial aberrations that are similar to those seen in humans clinically [17–19]; hence, justify the suitability of this model to study MI.

Roselle (*Hibiscus sabdariffa* Linn.) is a local natural plant enriched with natural anti-oxidant compounds such as ascorbic acid, quercetin, chlorogenic acid, caffeic acid, and anthocyanin [15]. In addition, we recently identified two anthocyanin compounds; delphinidin-3-O-sambubioside and cyanidin-3-O-sambubioside [20, Supplementary Fig S1] from roselle aqueous extract that was also used in this study. In vitro and in vivo studies revealed antioxidant [21, 22] capacity of roselle, besides proven as an anti-hypertensive [15] and anti-obesity [22] agent. Roselle was also reported to protect red blood cells from hydrogen peroxide-induced membrane damage in vivo [23]. In our recent research, roselle was shown to ablate cardiac fibrosis, hypertrophy, and oxidative stress in obesity-associated MI model when given for a longer period, 28 days [20]. However, effects of roselle in limiting early cardiac remodelling when given for a shorter duration, 7 days, has not yet been sought. Therefore, this study was to investigate the effects of roselle in limiting early cardiac remodelling in a MI rat model by targeting inflammatory responses, fibrosis, and hypertrophy.

Materials and Methods

Extraction of Roselle

Dried roselle (*Hibiscus sabdariffa* Linn. UMKL-1) calyces were purchased from HERBagus Sdn Bhd in Kepala Batas, Malaysia with voucher specimen of PID050515-05 from Forest Institute of Malaysia (FRIM). Roselle aqueous extract was prepared according to methods described in [20]. Dried roselle calyces were firstly pulverized into powder with blender prior to mixing them with distilled water in 1:10 ratio for 48 h with occasional shaking. The mixture was then filtered with clean, sterile strainer. The filtrate was kept frozen at − 80 in aluminium wrapped container to allow solidification for freeze drying. The solidified filtrate was then freeze-dried (Labconco, US) and kept at 4 °C in dark bottle until use.

Animals

A total of twenty-four adult Wistar male rats (200–300 g; 7–8 weeks) were used in this study. The rats were acclimatized in the laboratory condition for 1 week with ad libitum access to water and normal rat diet before carrying out the experiment. Throughout the experiment, all rats were housed under the same laboratory conditions of ambient room temperature and lighting (12 h light dark cycle). All the procedures involving animal handling were subjected to approval from Universiti Kebangsaan Malaysia Animal Ethics Committee (FSK/2016/SATIRAH/23-NOV./812-NOV.-2016-NOV.-2017).

Study Design

Rats were randomly allotted into three groups ($n = 6$): Control, MI, and MI + R. MI and MI + R groups were induced with subcutaneous injections of isoprenaline hydrochloride (Tokyo Chemical Industry, Japan) dissolved in normal saline at the dose of 85 mg/kg [20] while the control group received subcutaneous injection of normal saline; both for two consecutive days with 24 h interval. Rats then received the following treatments for 7 days via oral forced feeding: (1) Control (distilled water), (2) MI (distilled water), and (3) MI + R (roselle aqueous extract at 100 mg/kg in distilled water). On the 10th day upon first induction, the rats were anesthetized with 1 g/kg urethane (Sigma Aldrich, US) and sacrificed by cervical dislocation. Blood samples were collected in heparin tubes and separated by centrifugation. The heart tissue was excised immediately, washed with cold phosphate buffer saline (PBS), and used for further analysis.

Biochemical Analysis of Plasma

Plasma level of troponin-T and inflammatory markers (IL-6, IL-10) were quantitatively measured using commercial ELISA kit (Elabscience Biotechnology Co., Ltd). Briefly, 100 µl of standard (IL-6 and IL-10: 1.56–100 pg/ml; troponin-T) and plasma samples (control and roselle-treated) were added into microplate wells pre-coated with specific

antibodies and incubated at 25 °C for 2 h. Excessive solution was decanted and the wells were washed three times with PBS. Then 50 µl of biotinylated specific antibodies (1:100 dilutions) were added and incubation was done at 25 °C for 2 h, followed by 1-h incubation with 100 µl avidin-conjugated horseradish peroxidase (HRP) (1:200 dilutions) at 25 °C. Excessive solution was decanted and the wells were washed three times as described formerly. Substrate solution (100 µl) was added to each well to allow enzyme-substrate reaction. Enzyme-substrate reaction was terminated with 100 µl stop solution. The microplate was read spectrophotometrically at 450 nm.

Plasma lactate dehydrogenase (LDH) activity was assayed as previously described with slight modifications [24]. Briefly, 10 µl of plasma samples (control and roselle-treated) were added into microplate wells followed by 240 µl of PBS. Then, 10 µl of nicotinamide adenine dinucleotide (NADH) solution (2.5 mg/ml) were added and the plate was incubated at 25 °C for 20 min. Next, 10 µl of sodium pyruvate solution (2.5 mg/ml) was added. Activity of LDH was measured five times at 340 nm for 5 min with 1-min intervals.

Gene Expression

Total RNA was extracted from the left ventricle tissue using QIAzol lysis reagent (Qiagen, Germany). Using random primer, 2 µg of mRNA was reverse-transcribed using Quantinova Reverse Transcription Kit (Qiagen, Germany) according to manufacturer's protocol. Reaction mixtures containing 2 µg of mRNA, 4 µl of reverse transcription mix, 1 µl of reverse transcriptase, 2 µl of gDNA removal mix, and RNase-free water were prepared. The cDNA was then used to determine gene expression of inflammatory markers (IL-6 and IL-10), fibrosis markers (Col-1 and Col-3), fibronectin (FN) and hypertrophic marker atrial natriuretic peptide (ANP), and brain natriuretic peptide (BNP) in real-time PCR (RT-qPCR) using Quantinova SYBR® Green (Qiagen, Germany). The ribosomal 18S gene was used as endogenous control. RT-qPCR reactions containing 0.02 µg cDNA, 10 µl of SYBR Green Master Mix, 1 µl of forward primer (1 µmol/l), 1 µl of reverse primer (1 µmol/l) were prepared. RT-qPCR cycles were as follows: initial denaturation at 95 °C for 20 s, 40 cycles of denaturation at 95 °C for 3 s, and annealing at 60 °C for 30 s. Quantitative analysis was performed using CFX 96 Real-Time PCR Detection System (Bio-Rad, CA, USA). Primer sequences are given in Table 1.

Cardiac Histological Analysis

Small portion of left ventricle tissue was fixed with 10% formalin then embedded into paraffin blocks. Tissues were sectioned into 4-µm thickness using rotary microtome and stained with Haematoxylin and Eosin to measure the size of cardiomyocyte area (10 cells per field, 10 fields per heart section, 400× magnification) while Picrosirius Red staining was done to demonstrate infarct area (stitched images, 25 fields, 40× magnification) and to estimate percentage of collagen deposition (10 fields per heart, 100× magnification). Quantitative measurement of the heart, including cardiomyocyte cross-sectional area was calculated by measuring circumferential length of cardiomyocytes while percentage of collagen deposition was quantified using Sirius Red Macro software. Both quantitative measurements were done using ImageJ software (Bethesda, Maryland, USA).

Statistical Analysis

All values were expressed as mean ± SEM and analyzed with One-way analysis of variance (ANOVA) using GraphPad Prism software version 6. Values of $p < .05$ were considered statistically significant.

Results

Body and Organ Weight

After 7 days post-isoprenaline induction, MI group had significantly lower body weight (BW) as compared to control

Table 1 Primer sequences

Gene	Forward (5′–3′)	Reverse (5′–3′)
18S	TTCGAGGCCCTGTAATTGGA	GCAGCAACTTTAATATAGGCTATTGG
IL6	TCTCTCCGCAAGAGACTTCCA	ATACTGGTCTGTTGTGGGTGG
IL10	CCTGCTCTTACTGGCTGGAG	TGTCCAGCTGGTCCTTCTTT
COL 1	TGCTGCCTTTTCTGTTCCTT	AAGGTGCTGGGTAGGGAAGT
COL 3	GTCCACGAGGTGACAAAGGT	CATCTTTTCCAGGAGGTCCA
FN	GAAAGGCAACCAGCAGAGTC	CTGGAGTCAAGCCAGACACA
ANP	GGAAGTCAACCCGTCTCAGA	TGGGCTCCAATCCTGTCAAT
BNP	ACAAGAGAGAGCAGGACACC	TCTGGAGACTGGCTAGGACT

Primer sequences used in gene expression study

Table 2 Body and organ weight

Group	Body Weight (BW, g) (a)	Heart Weight (HW, mg) (b)	HW/BW (mg/g) (c)
Control	266.6 ± 8.8	836 ± 20	3.2 ± 0.1
MI	238.8 ± 9.0*	883 ± 40*	3.7 ± 0.2*
MI + R	262.0 ± 3.3	891 ± 20	3.4 ± 0.1

Values are presented as mean ± SEM for $n = 6$–8 per group

*$p < .05$ in relative to Control

group. MI group also showed increased absolute heart weight (HW) as well as HW/BW ratio. However, roselle treatment had no effects on body weight, heart weight, and heart weight to body weight ratio (Table 2).

Roselle Attenuated Expression of Cardiac Injury Markers in Post MI Model

Myocardial injury was evident by relatively higher plasma troponin-T and LDH levels after 7 days in untreated rats (Fig. 1a, b). Compared to the control group, the plasma troponin-T level was twofold higher in untreated rats (Fig. 1a). Similarly, the LDH level showed upward trend in the MI group (Fig. 1b). Roselle treatment was able to lower the level of plasma troponin-T but not the level of LDH.

Roselle Reduced Expression Levels of Plasma Inflammatory Markers in Post MI Model

The protein levels of inflammation markers, IL-6 (Fig. 2a), and IL-10 (Fig. 2b) were significantly elevated in MI rat's plasma when compared to control after 7 days of MI induction. Roselle supplementation reduced the expression levels of IL-6 and IL-10 significantly in MI rats.

Roselle Reduced Cardiac Inflammatory Gene Expressions in Post MI Model

The expression levels of IL-6 (Fig. 3a) and IL-10 (Fig. 3b) genes were elevated in MI group as compared to control after 7 days of MI induction. Roselle treatment significantly attenuated the expression level of IL-10 gene but showed no positive effects on IL-6 gene expression.

Roselle Limited Cardiac Fibrosis in Post MI Model

MI rats exhibited infarcted area stained in red as demonstrated by stitched image of heart section stained with

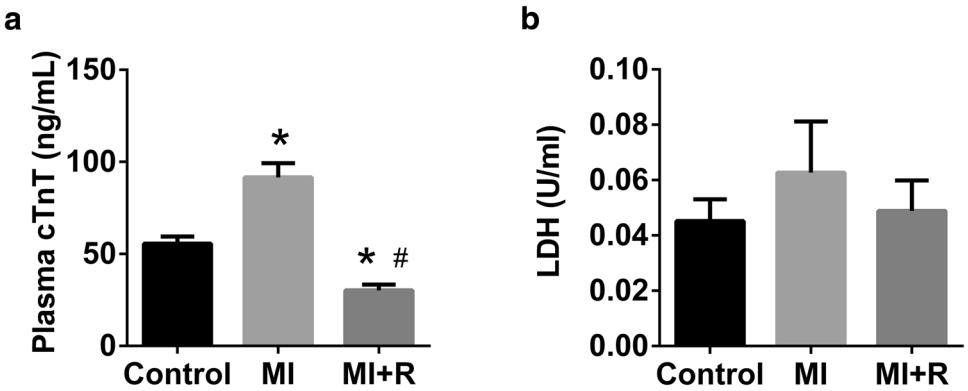

Fig. 1 Roselle supplementation attenuated expression of cardiac injury markers in post MI model. Measurement of plasma levels of troponin-T (**a**) and LDH (**b**). Plasma troponin-T and LDH levels elevated in response to MI. Roselle supplementation was able to reduce plasma level of troponin-T but not LDH. Values are presented as mean ± SEM for $n = 6$ per group. *$p < .05$ relative to Control, #$p < .05$ relative to MI

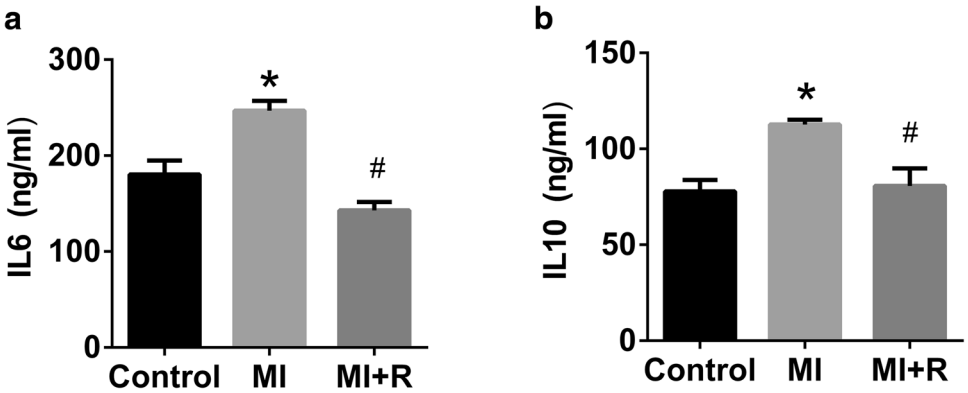

Fig. 2 Roselle lowered expression levels of plasma inflammatory markers in post MI model. Plasma levels of **a** IL-6 and **b** IL-10 significantly elevated 7-day post MI. Roselle supplementation attenuated the expression levels of the markers significantly. Values are presented as mean ± SEM for $n = 6$ per group. *$p < .05$ relative to Control, #$p < .05$ relative to MI

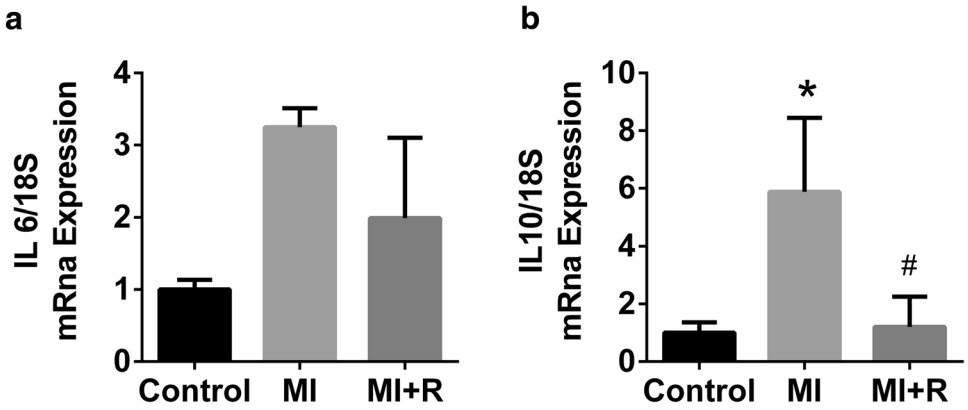

Fig. 3 Roselle reduced gene expression of inflammatory markers in post MI model. Fold changes in gene expression of **a** IL-6 and **b** IL-10 in relative to the housekeeping gene 18S. In MI group, IL-6 and IL-10 gene expression was upregulated. Roselle treatment only affected the expression of IL-10 positively. Values are presented as mean ± SEM for $n = 6$ per group. $^{*}p < .05$ relative to Control, $^{\#}p < .05$ relative to MI

Picrosirius red stain (Fig. 4a). MI induced remarkable collagen deposition as shown by Picrosirius red-stained heart section in MI group (Fig. 4b) as compared to control. Quantification of heart section showed significant higher amount of collagen in MI rats compared to control rats (Fig. 4c). Slight elevation of Col-1 gene expression was seen in MI rats (Fig. 4d). This was accompanied by a significant upregulation of Col-3 gene expression (Fig. 4e) and slight elevation of FN gene expression (Fig. 4f). Roselle supplementation significantly reduced collagen deposition in Picrosirius red stained-heart section as well as the Col-3 gene expression. Neither MI nor roselle supplementation affected the gene expression of Col-1 and FN.

Roselle Modulates Cardiomyocyte Hypertrophy in Post MI Model

In this study, MI caused significant enlargement of cardiomyocytes area after 7 days as observed through Haematoxylin and Eosin-stained heart section (Fig. 5a) and further supported by quantification of the cardiomyocytes area size (Fig. 5b). This is accompanied by significant upregulation in ANP (Fig. 5c) and BNP (Fig. 5d) gene expression. Roselle treatment managed to downregulates gene expression of ANP but failed to normalize the enlargement of cardiomyocytes area and gene expression of BNP seen in MI rats.

Discussion

Heart failure is a significant contributor to premature mortality among patients surviving MI. Post-MI cardiac remodelling is driven by unregulated tissue repair process to compensate the stress caused by tissue loss [25]. Series of well-coordinated events, initiated by inflammatory phase for 3–4 days, followed by a reparative phase with resolution of inflammation, collagen deposition, and scar formation over the next several days have been reported in MI models [12, 13, 15]. Transition of these processes requires adequate and timely physiological balance for optimal repair [26]. Consistently, using an established rat model of MI, we demonstrated that acute ISO administration initiated myocardial necrosis, inflammation, fibrosis, and hypertrophy. In the ISO-induced MI rat model, Roselle supplementation for 7 days was able to partially (i) lower myocardial injury, (ii) reduce systemic and local inflammation, (iii) limit cardiac fibrosis, and (i) down regulate cardiac hypertrophy.

Isoprenaline, a synthetic catecholamine is widely used as an inducer of myocardial infarction due to its technical simplicity and low mortality rate compared to coronary artery ligation technique. Several mechanisms have been proposed to explain isoprenaline-induced myocardial injury; for instance, (i) myocardial hyperactivity, (ii) metabolism alteration, (iii) coronary hypotension, (iv) calcium overload, (v) energy depletion, and (vi) excessive production of free radicals resulted from oxidative metabolism of catecholamines [27]. Several lines of evidences have reported that isoprenaline-induced myocardial infarction rat model experienced loss of cardiomyocytes due to myocardial necrosis that was accompanied by inflammation and cardiac fibrosis [12–14].

MI following isoprenaline administration could result in prominent increase of absolute HW with decreased BW due to upsurge of HW/BW ratio. Similar to our findings, previous studies reported that increase of HW/BW ratio [28–30] was mainly attributed by water accumulation in the intramuscular spaces [27], increased protein content [12], and cellular infiltration into damaged heart tissues [31]. As low as 1% increment in intramuscular water content will compromise 10% of myocardial functions [32]. In this study, roselle treatment given for 7 days was able to reduce the HW/BW ratio although the reduction was not statistically significant.

In addition to heart weight changes, isoprenaline intoxicated rats exhibited myocardial damages as evidenced by the histological changes. In myocardial damages, injury markers such as troponin T and LDH leak from myocardia into the circulation due to inability of maintaining cellular membrane integrity [29]. Consistent with other studies, MI was

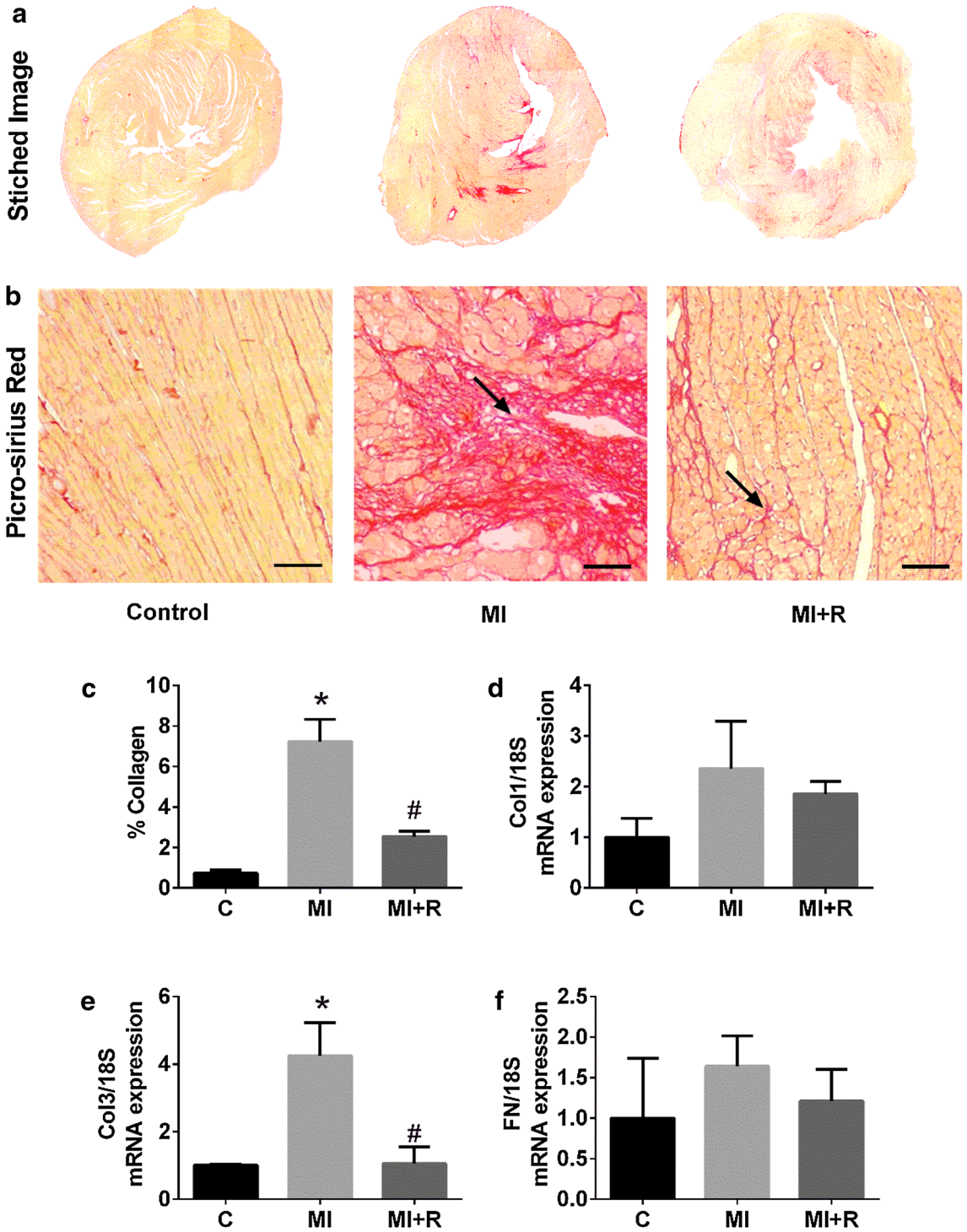

Fig. 4 Roselle supplementation limited cardiac fibrosis in post MI model. Representative images of **a** Picrosirius red-stained stitched images of heart sections; 40× magnification and **b** Picrosirius red-stained left ventricle sections; 100× magnification; scale bar 50 µm. Arrows show deposition of collagen in red stain. **c** Measurement of collagen deposition (%) in Picrosirius red-stained left ventricle sections. Fold changes in gene expression of **d** Col-1, **e** Col-3, and **f** FN in relative to the 18S gene. MI induced high deposition of collagen and roselle seems to be lowered collagen deposition. Roselle supplementation also reduced Col-3 gene expression in MI rats. Values are presented as mean ± SEM for $n = 6$ per group. $^{*}p < .05$ relative to Control, $^{\#}p < .05$ relative to MI. (Color figure online)

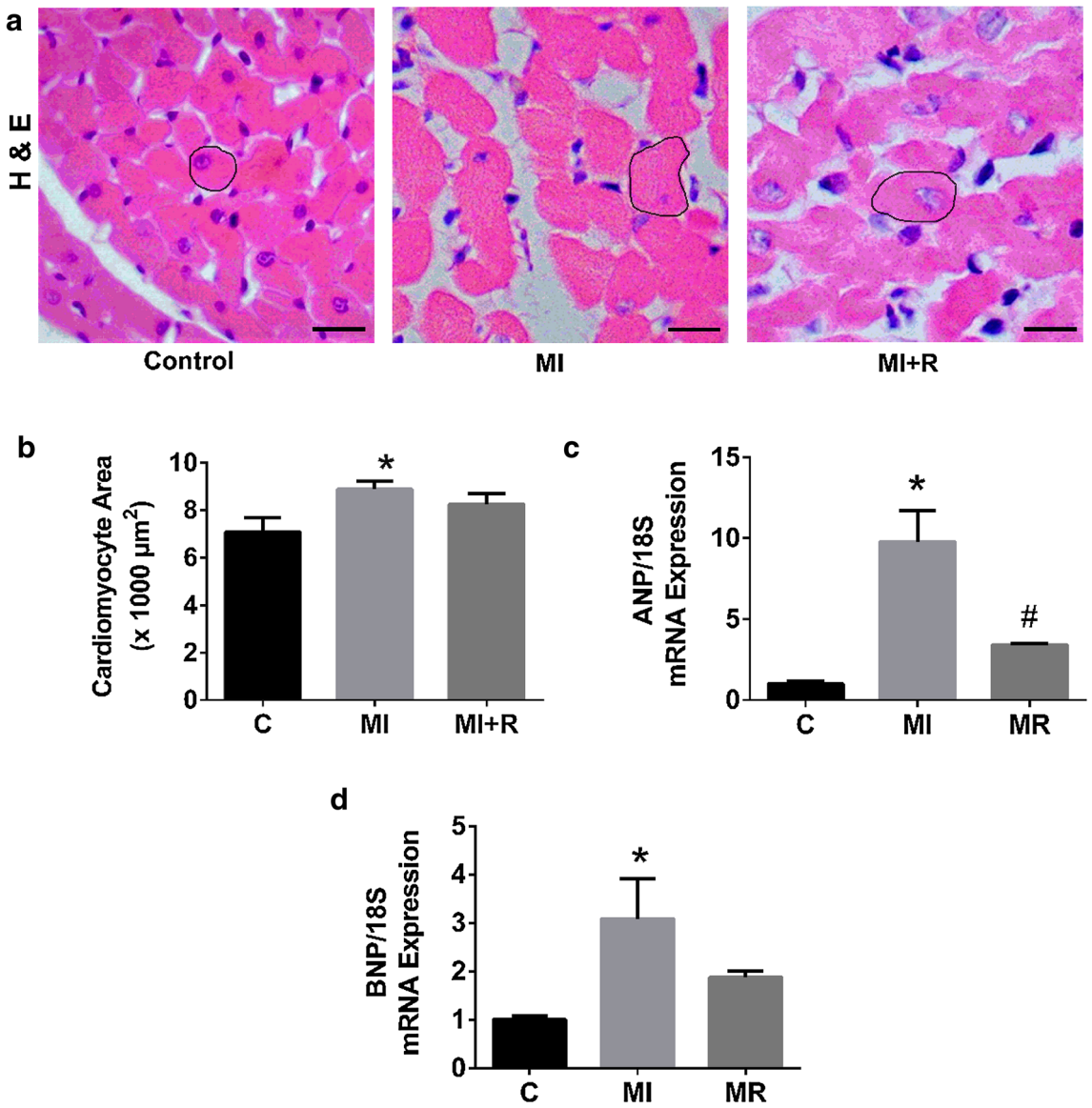

Fig. 5 Roselle modulates cardiomyocytes hypertrophy in post MI model. Representative images of **a** H&E-stained left ventricle sections; 40× magnification; scale bar 50 µm. Representative cardiomyocytes are circled in black showing cardiomyocytes area measured. **b** Measurements of cardiomyocyte cross-sectional areas in H&E-stained left ventricle sections. Fold changes in gene expression of **c** ANP, **d** BNP in relative to the 18S gene. MI induced cardiomyocytes hypertrophy evident by increase in cardiomyocytes area and significant upregulation of ANP and BNP gene expression. Roselle supplementation did not improve cardiomyocyte hypertrophy in the MI heart tissues in terms of cardiomyocyte area; however, roselle normalized gene expression of ANP. Values are presented as mean ± SEM for $n = 6$ per group. $^{*}p < .05$ relative to Control, $^{\#}p < .05$ relative to MI. (Color figure online)

found to significantly elevate the plasma levels of troponin-T. However, the plasma level of LDH was only slightly elevated. This could be due to short persistence of LDH in circulation post MI. LDH concentration normalize to baseline within 6–15 days [33]; hence, the relatively low level of LDH detected 7 days post MI in this study. Being a sensitive cardiac-specific regulatory protein, troponin-T often serves as gold standard in diagnosing myocardial injury [5]. A previous report stated that the level of troponin-T released into the circulation was well correlated with the severity of myocardial necrosis [34]. Interestingly, roselle supplementation managed to reduce the plasma level of troponin-T but not LDH in MI rats. This is probably due to anthocyanin content in roselle which was previously shown to be able to stabilize the integrity and functions of cells hence restricting leakage of cellular contents into the circulation [23].

Infiltration of inflammatory cells was observed in myocardium as soon as the first hour after MI [25]. Inflammation plays important role in the progression of cardiac repair by stimulating collagen deposition to compensate

myocyte loss. Prolonged, insufficiently suppressed inflammation may, therefore, lead to over deposition of collagen in cardiac, stiffening of heart wall hence compromised cardiac function [35]. Necrotic cardiomyocytes activate inflammatory signalling that in turn triggers resident and infiltrating inflammatory cells to injury sites. This leads to secretion of pro-inflammatory cytokines such as IL-6 and recruitment of neutrophils. Addition to its pro-inflammatory action, neutrophils may also trigger reparative macrophages. Besides clearing necrotic tissues, macrophages may exert anti-inflammatory effect by secreting IL-10 [36]. Upregulation of inflammatory cytokines such as IL-6 and IL-10 elevated in both animal models and human MI within few hours [37, 38]. Consistent with other studies [11, 39], this study showed that plasma levels of IL-6 and IL-10 were significantly upregulated in MI rats. This was further supported by the expression levels of IL-6 and -10 genes in MI heart tissues. In this study, roselle treatment was able to reduce the plasma levels of IL-6 and IL-10 and also the gene expression level of IL-10. IL-6 gene expression was heightened in MI group and was unaffected by 7-day roselle supplementation. It is speculated roselle might not act directly on IL-6 gene expression while the positive effects of roselle on IL-10 gene expression are presumably contributed by the presence of anthocyanin that has been proven to ablate inflammation in a myocardial ischemia–reperfusion injury model [40].

Apart from inflammation, cardiac fibrosis caused by myocardial remodelling also plays a major role in the development of heart failure. This is primarily caused by over-deposition of myocardial interstitial collagen, thereby altering heart functions. Deposition of Col-1, Col-3, and fibronectin builds up the extracellular matrix (ECM) network that provides structural and functional integrity to the heart. However, over-deposition of ECM reduces elasticity of heart wall, hence results in systolic and diastolic dysfunction [20]. This is worsened by the presence of cardiac fibroblasts which are the primary source of ECM [41]. In this study, isoprenaline-induced MI caused cardiac fibrosis by significant over-deposition of collagen that was substantiated by the elevated Col-3 gene expression. This is in line with that reported by a previous study [42] in which early cardiac remodelling in a hypertensive heart disease model exhibited initial increase in myocardial Col-3 and then followed by Col-1. Interestingly, roselle treatment ameliorated the occurrence of cardiac fibrosis. This correlates well with our recent study [20] that roselle, when given for 28 days, to obese-associated MI rats, was able to ablate myocardial fibrosis significantly. It is suggested that the anti-fibrotic activity of roselle in various animal models [43, 44] could be possibly due to its high anthocyanin content. However, the exact mechanism of anthocyanin contributing to anti-fibrotic activity remains unclear hence further study is needed to elucidate this.

Besides inflammation and fibrosis, hypertrophy is also responsible for adverse outcomes in post MI setting [45]. Cardiomyocytes hypertrophy is initiated as a compensatory mechanism secondary to pressure overload on the heart wall. However, irreversible cardiac hypertrophy often results in dilation of heart chamber hence impaired cardiac functions [10]. Cardiac hypertrophy is characterized by increase in the size of individual cardiomyocytes and re-expression of fetal cardiac genes, such as ANP and BNP [46]. Similar to other study, we have shown that MI rats showed significant upregulation of ANP and BNP gene expression hence suggesting cardiac hypertrophy. Consistent with previous study, roselle supplementation was able to modulate the ANP gene expression [20] although roselle is not effective in reducing cardiomyocytes area enlargement seen in MI rats.

Conclusion

This study proved that roselle was able to help ameliorate early cardiac remodelling by regulating inflammation, cardiac fibrosis, and hypertrophy in post MI setting. Although cardiac remodelling helps in maintaining cardiac performance during stress, over time, they can be maladaptive and eventually results in heart failure. Currently, there is no effective treatment agent available for managing MI complications; therefore, discovery of nutraceutical agents with protective properties against myocardial injury such as roselle may improve recovery of heart functions and prolong lifespan.

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Compliance with Ethical Standards

Conflict of interest The authors declare that there is no conflict of interest regarding the publication of this paper.

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