



## ASSESSMENT OF WILD AFRICAN VEGETABLES ON CARDIAC FUNCTIONS, ANTIOXIDANTS AND BLOOD LIPID PROFILE IN A RAT MODEL OF MYOCARDIAL INFARCTION

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**Abstract** The heart is susceptible to many chronic diseases including myocardial infarction (MI). This study assessed the cardio-protective potential of five wild vegetables: *Basella alba*, *Crassocephalum crepidioides*, *Launaea taraxacifolia*, *Senecio bialafrae*, and *Solanum nigrum* in MI rat model, thus revealing its biochemical activities *in vivo*. Nourished animals (84) were randomized into 14 groups (n=6 rats per group). Groups 2 to 6 and groups 9 to 13 orally received 200 mg kg<sup>-1</sup>bw of each vegetable's aqueous extract while groups 7 and 14 received equal ratio of extracts combination. Groups 1 and 8 served as normal and isoproterenol (ISO) control respectively. Only groups 9 to 14 were subcutaneously administered ISO (20 mg kg<sup>-1</sup> bw) on days 29 and 30 after 28 days extracts pre-treatment. Antioxidants, haematology and lipid markers were assessed while excised hearts were histologically examined. One-way analysis of variance (ANOVA) with least significance difference (LSD) tests were used to analyse data statistically ( $p < 0.05$ ). ISO control (group 8) significantly ( $p < 0.05$ ) decreased blood indices, antioxidants, high-density lipoprotein (HDL) and increased low-density lipoprotein (LDL), lipid peroxidation with myocardial aberrations as opposed to vegetables pre-treated groups. All the groups pre-treated with vegetables (singly and combination) prevented MI with increase in blood parameters, antioxidants, HDL and decreased LDL, lipid peroxidation as well as normal and lesser degree of myocardia distortion in histological examination. Induction of MI by ISO and the protective effects of the vegetables' extracts on cardiac functions via mechanism(s) involving oxidative stress reduction and improvement of blood lipid profile were confirmed from the study.

**Keywords:** *Basella alba*, *Crassocephalum crepidioides*, *Launaea taraxacifolia*, *Senecio bialafrae*, *Solanum nigrum*, wild vegetables, myocardial infarction

### Introduction

Cardiovascular disease (CVD) which majorly comprises coronary heart disease, cerebrovascular disease (or stroke), hypertension and peripheral vascular disease, remains a global health challenge. Undeniably, CVD is the main cause of death globally because every year, an estimated 17.9 million people die from CVDs. Coronary heart disease (CHD), which accounts for 7.3 million of CVD deaths, results from the build-up of plaques in the inner walls of the coronary arteries (WHO, 2020).

Accumulation of plaque in the arterial wall limits the flow of blood to heart muscles thereby causing ischemia. Prolonged restriction of blood supply to cardiac myocytes in turn leads to necrosis of cardiac myocytes and, in pathology, this condition is known as myocardial infarction or MI. In addition to necrosis of cardiac myocytes, MI has also been associated with increased or decreased levels of C-reactive protein in plasma (Salisu *et al.*, 2018).

Essentially, treatment and prevention of CVDs involves targeting the risk factors and maintaining a healthy lifestyle respectively. Apart from cessation from smoking, moderate alcohol consumption and frequent exercise, other preventive therapies recommended by physicians to individuals at high risk of CVDs are targeted at the diet. Reducing intake of sugar and saturated fats as well as eating a diet rich in fruits and vegetables contribute to cardiovascular health (Aggarwal *et al.*, 2017). Whilst it has been clearly shown that fruit and vegetable intakes are associated with reducing the risk of CVDs, cancer and all-cause mortalities (Aune *et al.*, 2017), only a few scientific studies have been done to investigate the protective potential of African wild leafy vegetables against MI. We have previously identified chemical compounds in these vegetables and also proved their effects on blood pressure and cardiac biomarkers in myocardial infarction – induced male Wistar rats (Salisu *et al.*, 2018; Salisu *et al.*, 2019). These studies suggest a preventive role of each vegetable and their combination in ISO-induced MI rats, but their biochemical activities have not been

documented. In this study, we demonstrated the protective properties of five African wild underutilized leafy vegetables- *Launaea taraxacifolia* (LT), *Crassocephalum crepidioides* (CC), *Solanum nigrum* (SN), *Basella alba* (BA) and *Senecio biafrae* (SB) in a rat model of myocardial infarction, and investigated the mechanism(s) underlying their cardiac function protective role.

## Materials and Methods

### Plant collection, voucher numbers and preparation of aqueous leaf extracts

Fresh leaves of *L. taraxacifolia* (LUH5806A), *C. crepidioides* (LUH1229A), *S. nigrum* (LUH1228A), *B. alba* (LUH5807A) and *S. biafrae* (LUH1227A) were plucked from Dr. Salisu's home garden, Ota, Ogun State, Nigeria (Latitude 6.7°N and longitude 3.2°E), during the rainy season. Identification and authentication with voucher specimen numbers in parenthesis above was done by Dr. A. B. Kadiri, a taxonomist at the Herbarium of the Department of Botany, Faculty of Science, University of Lagos, Nigeria. The leaves were washed with distilled water, air-dried under room temperature (26 °C) for a week and then coarsely (30–40 mesh sieves) pulverized using electric blender (Binatone, BLG-451). The ground plant materials and mixture in equal ratio 1:1:1:1:1 were macerated in three litres of cold distilled water for 72 hours at room temperature (26 °C), filtered, evaporated under reduced pressure and freeze dried at 4 °C for the study (Handa, 2008).

### Animals and Treatment

All animal experiments and protocols were approved by the Animal Care and Use/Research Ethics Committee (ACUREC) of the College of Medicine, University of Lagos, Idi-Araba, Lagos with ethical approval number CM/COM/08/VOL.XXV prior to research. Purchased Male Wistar albino rats (150g to 200g) from Experimental Animal Unit of the Faculty of Veterinary Medicine, University of Ibadan were housed to acclimatize (2 weeks before the study) in standardized conditions with constant temperature (27 °C), humidity, and a 12-h light/dark cycle, fed with sterilized rat pellets and distilled water *ad libitum*. Rats were randomized into 14 groups (6 per group) and categorized into two: vegetables category (Table 1) and ISO-treated category (Table 2). In vegetable category, Group 1 (normal control) received normal saline for 28 days while groups 2 to 7 were administered orally 200 mg kg<sup>-1</sup> bw<sup>-1</sup> of each plant extract including the mixture (1:1:1:1:1) for 28 days. In the ISO-treated category, group 8 served as ISO control while groups 9 to 14 were also administered 200 mg kg<sup>-1</sup> bw<sup>-1</sup> of each plant extract

including the mixture (1:1:1:1:1) orally for 28 days and then ISO (Sigma Chemical Company, St. Louis, MO, USA) drug dose of 20 mg kg<sup>-1</sup> bw<sup>-1</sup> was subcutaneously administered within 10 minutes of preparation on day 29 and 30 according to the method of Radhika *et al.* (2011).

### Blood and heart samples collection

At the end of the experimental phase, a cardiac puncture was performed to obtain blood samples for haematological and biochemical studies from the rats sacrificed by cervical dislocation. The heart of each rat was carefully removed for histological examination.

### Haematological measurement

Red blood cells (RBC), packed cell volume (PCV), white blood cells (WBC) and blood platelets (PLT) were measured by injecting blood sample (13µl) into an automatic haematology analyser (Mindray BC3200 model, Shenzhen, China) to sort and size blood cells on the basis of changes in light pulses as the cells pass in front of a laser beam.

### Biochemical measurement

Superoxide dismutase (SOD) was assayed by its ability to inhibit the auto-oxidation of epinephrine determined by the increase in absorbance at 480nm as described by method of Sun and Zigma (1978). Catalase (CAT) activity was determined as described by Usuh *et al.* (2005) by measuring the decrease in absorbance at 240nm due to the decomposition of H<sub>2</sub>O<sub>2</sub> in a UV recording spectrophotometer. The glutathione (GSH) content was estimated according to the method described by Sedlak and Lindsay (1968) while malondialdehyde (MDA) was measured as an index of lipid peroxidation as described by Hosseinzadeh *et al.* (2009). High density lipoproteins (HDL), low-density lipoproteins (LDL), total cholesterol (TC) and triglycerides (TG) assays were photometrically performed on an Automatic Biochemistry Analyser (902 Roche/Hitachi) by commercial test kits (Roche diagnostics, Sandhofer Strasse, Mannheim, Germany) following the protocol of the manufacturer.

### Histopathological Examination

The histopathology procedure of Taylor *et al.* (2003) was used. Small slices of the heart tissues of the excised animals were washed with normal saline, fixed in 10% buffered formalin, embedded in paraffin and sectioned (5 µm thick). Each section was stained with hematoxylin and eosin dye for examination under light microscope with a camera for histopathological changes and photomicrographs.

### Statistical Analysis

All data are presented as mean ± standard error of the mean (SEM) and analysed by Statistical package for Social Sciences (SPSS) software (version 20, IBM Corporation, Armonk, NY, USA). One way

Analysis of variance (ANOVA) coupled with a post-hoc test of least significance difference (LSD) was used to analyse the data. The results were considered significant at  $p$  values of less than 0.05 ( $p < 0.05$ ).

## Results and Discussion

### Effect of aqueous leafy vegetables' extracts on haematological parameters

Tables 3 and 4 below showed the effect of vegetables' extracts in pre-treated rats (vegetables and ISO-treated categories respectively). The mean RBC count was significantly increased ( $p < 0.05$ ) in the mixture group but slightly significantly decreased ( $p < 0.05$ ) in *B. alba* and *L. taraxacifolia* groups as compared to normal control (Table 3). However, the mean PCV was elevated significantly ( $p < 0.05$ ) in the *L. taraxacifolia* pre-treated rats while no significant difference ( $p > 0.05$ ) was observed in other groups as compared with the control group. The mean WBC increased ( $p < 0.05$ ) in all the groups except in *C. crepidioides* group with no significant difference ( $p > 0.05$ ) as compared to control group. A significant increase in mean PLT was observed in all the groups when compared to control (Table 3). The mean RBC values in ISO-treated category (Table 4) of all the pre-treated groups were significantly increased ( $p < 0.05$ ) when compared with ISO control. No significant difference ( $p > 0.05$ ) in mean PCV values of *L. taraxacifolia*, *S. nigrum* and mixture groups in comparison with ISO control group but significant increases ( $p < 0.05$ ) were recorded in the remaining groups (*B. alba*, *C. crepidioides* and *S. bialfræ* groups) as compared to normal and ISO groups (Table 4). A significant increase in mean WBC counts was observed in all the groups as compared to ISO control group but no significant differences ( $p > 0.05$ ) in PLT counts of all pre-treated groups when compared with controls. Haematological parameters have been described as metrics to assess physiological, nutritional and pathological status of animals (Muhammad *et al.*, 2000). The elevated mean RBC, PLT, PCV and WBC of the animals pre-treated with plant extracts compared to untreated groups corroborates previous reports that vegetables may contain blood forming vitamins and some compounds capable of increasing production of blood cells in fighting stress and maintaining normal physiological state of the animals (Eburnlomo *et al.*, 2012; Adias *et al.*, 2013). The five African wild vegetables and their mixture have powerful haemopoietic potential that suppressed effects of oxidative stress on haematological indices in Wistar rats in which myocardial infarction was induced.

### Effect of aqueous leafy vegetables' extracts on biochemical measurements

As shown in Figure 1 below, a significant increase ( $p < 0.05$ ) in SOD activities was observed in

vegetable category while the contrary was the case in ISO-treated category when compared to ISO control (Figure 1). CAT activities in figure 2 and GSH concentration in figure 3 were significantly increased ( $p < 0.05$ ) in all the groups as compared to normal control and ISO control groups (Figures 2 to 3). MDA concentration displayed in figure 4 was reduced significantly ( $p < 0.05$ ) in all the groups when compared to the two control groups (Figure 4). Figure 5 to 7 depicts plasma lipid profile of both categories of pre-treated rats. The mean HDL was significantly increased ( $p < 0.05$ ) while mean LDL was significantly decreased ( $p < 0.05$ ) in all the groups in the vegetable category as compared to normal control (Figure 5). The ISO-treated category on the other hand, exhibited a drastic significant increase ( $p < 0.05$ ) in mean LDL (Figure 5), TC (Figure 6) and TG (Figure 7) of ISO control group while in the other pre-treated groups these parameters were significantly lowered ( $p < 0.05$ ). However, there was no significant difference ( $p > 0.05$ ) in the mean HDL of all the pre-treated groups as compared to normal control group (Figure 5). Endogenous antioxidant enzymes (SOD and CAT) and non-enzymatic antioxidant biomolecules (GSH) of the heart homogenates (supernatant) were functionally impaired in ISO group resulting in a decline in their activities. This result is in consonance with earlier report of Patel *et al.*, (2010) that accumulation of oxidants as a result of decrease in antioxidants (SOD, CAT and GSH) leads to susceptibility of myocardial cell membranes to oxidative damage. Conversely, increase in these antioxidants (SOD, CAT and GSH) activities in vegetables pre-treated groups may be attributed to the role of the vegetables' extracts in regulating antioxidant activities as previously reported (Gunja *et al.*, 2010). MDA is a major end product of lipid peroxidation and marker of oxidative stress (Sailaja *et al.*, 2013).

In the present study, increased lipid peroxidation was characterised by increased MDA levels in ISO-treated animals. This elevated lipid peroxidation may cause damage to cardiac constituents. This is in line with the previous reports that increased MDA contents may be as a result of increased generation of free radicals and/or decreased activities of antioxidant defence mechanism (Rajadurai and Prince, 2005; Priscilla and Prince, 2009). Lowered MDA content in all the vegetables' pre-treated groups as compared to controls could be interpreted as a mechanism of protection against oxidative stress induced by ISO and this is indicative of antioxidant activity of the plant extracts.

Enhanced lipid biosynthesis resulting in hyperlipidemia has been postulated to be due to administration of ISO which then stimulates adenylate cyclase activity (Patel *et al.*, 2010). Hyperlipidemic effects of ISO was characterised by

elevated LDL (bad cholesterol) levels, total cholesterol (TC), Triglycerides (TG) and lowered HDL (good cholesterol) levels as compared to normal control. Accumulation of these cholesterol and TG in the heart tissue is usually accompanied by cardiovascular damage (Gokkusu and Mostafazadeh, 2003). All the vegetable pre-treated groups displayed reduced levels of these cholesterol (LDL, TC and TG) and increased level of good cholesterol (HDL) supporting earlier studies of preventing damage to myocardium (Kareem *et al.*, 2009). A strong positive association has been documented between the risk of developing ischemic heart disease and serum LDL level, whereas a negative association has been reported with HDL-cholesterol (Radhiga *et al.*, 2012). Significantly lowered TC and TG by pre-treatment with the vegetable's extracts could be due to non-atherogenic redirection of circulating lipoproteins by efficient lipolysis of TG-rich lipoprotein in the heart (Sailaja *et al.*, 2013).

#### Effect of aqueous leafy vegetables' extracts on histological examination

Myocardial aberrations characterised by necrosis, oedema (widening of inter fibre space) and slender fibres were observed in ISO control group (Figure 9) as compared to normal control in Figure 8. Rats pre-treated with BA (BA+ISO-treated) in figure 10 restored normal myocardium in comparison with ISO control group in figure 9. CC +ISO-treated rat (Figure 11) was also restored due to 28 days pre-treatment of CC as compared to ISO control group in figure 9. However, a wavy myocardium was noticed in LT+ISO-treated rats (Figure 12) which is near normal myocardium when compared to the rats treated with only ISO (Figure 9). Development of MI was prevented in animals that were pre-treated with SB with a slight elongation of myocardial fibres

(Figure 13) as opposed to ISO control group in figure 9. Protection of MI was also visible in SN +ISO-treated group (Figure 14) with normal myocardium architecture as compared to ISO-treated group only (Figure 9). Animals in the mixture group that were pre-treated with the combination of all the vegetables' extract in equal ratio (Figure 15) showed absolutely normal myocardial fibre while other vegetables' extracts pre-treated groups (Figure 10 to 14) displayed near to normal myocardium architecture. Histopathological examination of myocardial tissue in normal control showed clear integrity of the myocardial cell membrane associated with syncytium of myocardial fibers and absence of inflammatory cell infiltration. ISO-treated rats in contrast, showed necrosis, oedema, slenderer fibers and infiltration of inflammatory cells. Hearts of animals pre-treated with vegetables' extracts exhibited near to normal myocardium architecture with normal myocardial fibres and the intervening interstitium thus confirming the cardioprotective effects of the plant extracts as reported in literature (Fathiazad *et al.*, 2012).

#### Conclusion

This study provided scientific basis for a more effective approach than treatment for chronic diseases such as myocardial infarction. The study also revealed usefulness of African wild leafy vegetables for cardioprotective purposes and preventive medicine ultimately, justifying the need to create awareness about diets that are rich in less-used indigenous green leafy vegetables in promoting health among the populace.

#### Conflict of interest

The authors declare no conflict of interests as regards publication of this paper.

**Table 1: Animal grouping for 30 days (Vegetables category)**

Groups	Number of rats	Plant extracts
1	6	Normal control (no extract)
2	6	<i>B. alba</i> (BA)
3	6	<i>C. crepidioides</i> (CC)
4	6	<i>L. taraxacifolia</i> (LT)
5	6	<i>S. biafrae</i> (SB)
6	6	<i>S. nigrum</i> (SN)
7	6	Mixture (1:1:1:1:1)

**Table 2: Animal grouping for 30 days (ISO-treated category)**

Groups	Number of rats	Plant extracts
8	6	ISO (ISO control)
9	6	<i>B. alba</i> (BA) + ISO
10	6	<i>C. crepidioides</i> (CC) + ISO
11	6	<i>L. taraxacifolia</i> (LT) + ISO
12	6	<i>S. biafrae</i> (SB) + ISO
13	6	<i>S. nigrum</i> (SN)+ ISO
14	6	Mixture (1:1:1:1:1) + ISO



**Table 3: Effects of vegetables' extracts pre-treatment (28 days) on haematological parameters in Wistar rats (vegetables category)**

Blood Indices	Normal control						
	(Grp1)	BA (Grp 2)	CC (Grp 3)	LT (Grp 4)	SB (Grp 5)	SN (Grp 6)	Mixture (Grp 7)
RBC ( $\times 10^6 \mu\text{L}^{-1}$ )	7.62 $\pm$ 0.12 <sup>ab</sup>	6.61 $\pm$ 0.33 <sup>bc</sup>	7.70 $\pm$ 0.10 <sup>ab</sup>	5.61 $\pm$ 0.37 <sup>c</sup>	7.2 $\pm$ 0.25 <sup>ab</sup>	7.72 $\pm$ 0.13 <sup>ab</sup>	7.88 $\pm$ 0.26 <sup>a</sup>
PCV (%)	42.3 $\pm$ 0.78 <sup>b</sup>	40.2 $\pm$ 1.46 <sup>b</sup>	41.6 $\pm$ 0.10 <sup>b</sup>	48.7 $\pm$ 0.89 <sup>a</sup>	42.2 $\pm$ 0.51 <sup>b</sup>	44.1 $\pm$ 0.59 <sup>b</sup>	42.9 $\pm$ 1.10 <sup>b</sup>
WBC ( $\times 10^3 \mu\text{L}^{-1}$ )	14.3 $\pm$ 0.40 <sup>d</sup>	22.1 $\pm$ 0.50 <sup>bc</sup>	14.1 $\pm$ 0.51 <sup>d</sup>	23.9 $\pm$ 0.99 <sup>b</sup>	29.2 $\pm$ 1.98 <sup>a</sup>	17.5 $\pm$ 0.77 <sup>cd</sup>	18.6 $\pm$ 1.50 <sup>cd</sup>
PLT ( $\times 10^3 \mu\text{L}^{-1}$ )	639 $\pm$ 75.1 <sup>b</sup>	873 $\pm$ 11.6 <sup>a</sup>	720 $\pm$ 2.11 <sup>ab</sup>	664 $\pm$ 7.12 <sup>ab</sup>	717 $\pm$ 17.8 <sup>ab</sup>	756 $\pm$ 29.9 <sup>ab</sup>	795 $\pm$ 90.4 <sup>ab</sup>

Values are means  $\pm$  SEM, n = 6 per treatment group. Means in a row without a common superscript letter differ ( $p < 0.05$ ) as analysed by one-way ANOVA and the LSD test

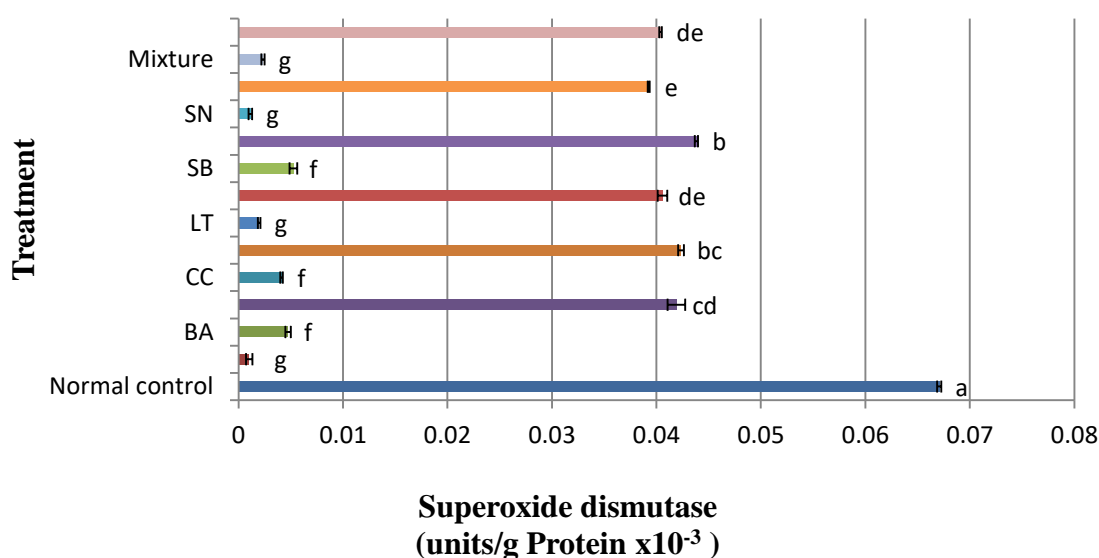
BA= *Basella alba*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*, SB= *Senecio bialfrae*, SN= *Solanum nigrum*, Grp = Group.

**Table 4: Effects of vegetables' extracts pre-treatment (28 days) on haematological parameters in Wistar rats (ISO-treated category)**

Blood Indices	Normal control							
	(Grp 1)	ISO control (Grp 8)	BA+ISO (Grp 9)	CC+ ISO (Grp 10)	LT + ISO (Grp 11)	SB + ISO (Grp 12)	SN + ISO (Grp 13)	Mixture +ISO (Grp 14)
RBC ( $\times 10^6 \mu\text{L}^{-1}$ )	7.62 $\pm$ 0.12 <sup>a</sup>	6.37 $\pm$ 0.07 <sup>b</sup>	8.31 $\pm$ 0.16 <sup>a</sup>	8.67 $\pm$ 0.22 <sup>a</sup>	7.97 $\pm$ 0.46 <sup>a</sup>	8.68 $\pm$ 0.26 <sup>a</sup>	8.09 $\pm$ 0.19 <sup>a</sup>	8.00 $\pm$ 0.22 <sup>a</sup>
PCV (%)	42.3 $\pm$ 0.78 <sup>b</sup>	47.6 $\pm$ 0.85 <sup>ab</sup>	49.4 $\pm$ 0.95 <sup>a</sup>	49.2 $\pm$ 1.26 <sup>a</sup>	45.9 $\pm$ 2.92 <sup>ab</sup>	49.4 $\pm$ 1.11 <sup>a</sup>	47.7 $\pm$ 0.41 <sup>ab</sup>	45.8 $\pm$ 1.62 <sup>ab</sup>
WBC ( $\times 10^3 \mu\text{L}^{-1}$ )	14.3 $\pm$ 0.40 <sup>ab</sup>	8.58 $\pm$ 0.29 <sup>c</sup>	17.6 $\pm$ 1.23 <sup>ab</sup>	20.6 $\pm$ 1.12 <sup>a</sup>	16.6 $\pm$ 0.63 <sup>ab</sup>	17 $\pm$ 1.14 <sup>ab</sup>	18.5 $\pm$ 0.66 <sup>ab</sup>	13.9 $\pm$ 0.67 <sup>b</sup>
PLT ( $\times 10^3 \mu\text{L}^{-1}$ )	639 $\pm$ 75.1	652.0 $\pm$ 51.6	818.0 $\pm$ 37.3	807.0 $\pm$ 40.3	775.0 $\pm$ 49.3	888.0 $\pm$ 135.0	912.0 $\pm$ 81.0	769.0 $\pm$ 46.3

Values are means  $\pm$  SEM, n = 6 per treatment group. Means in a row without a common superscript letter differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

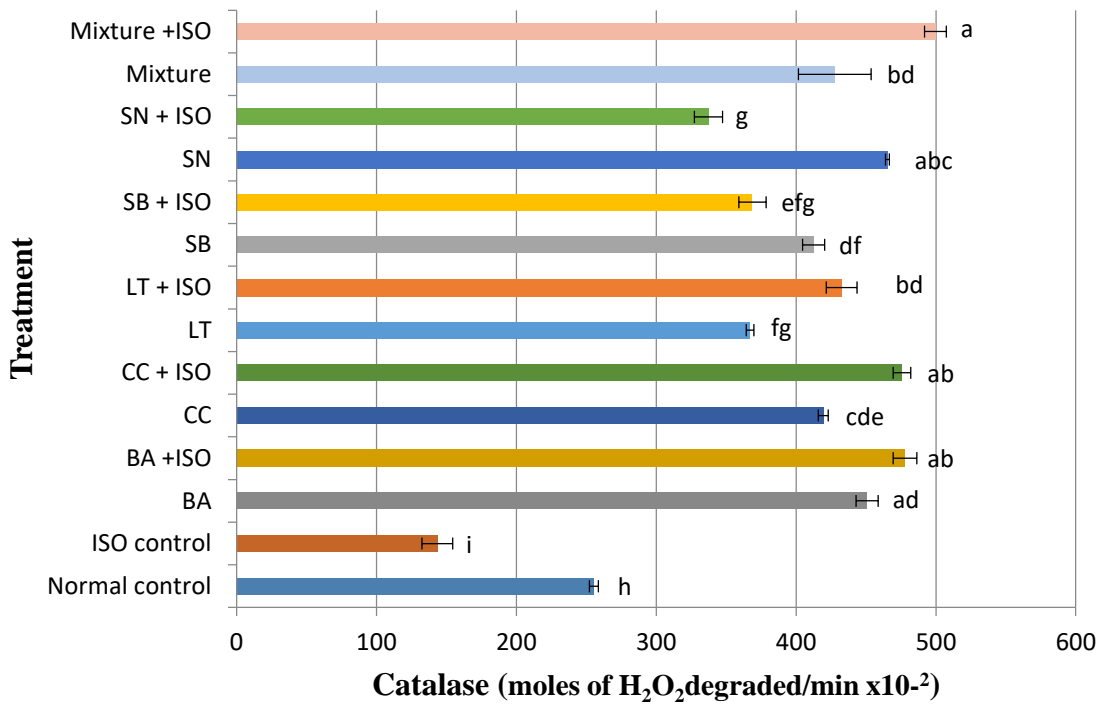
BA= *Basella alba*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*, SB= *Senecio bialfrae*, SN= *Solanum nigrum*, Grp = Group.



**Figure 1: Effects of aqueous leafy vegetables' extracts pre-treatment on Superoxide dismutase (SOD) activity in isoproterenol-induced myocardial infarction rats (vegetable and ISO-treated categories) for 28 days**

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

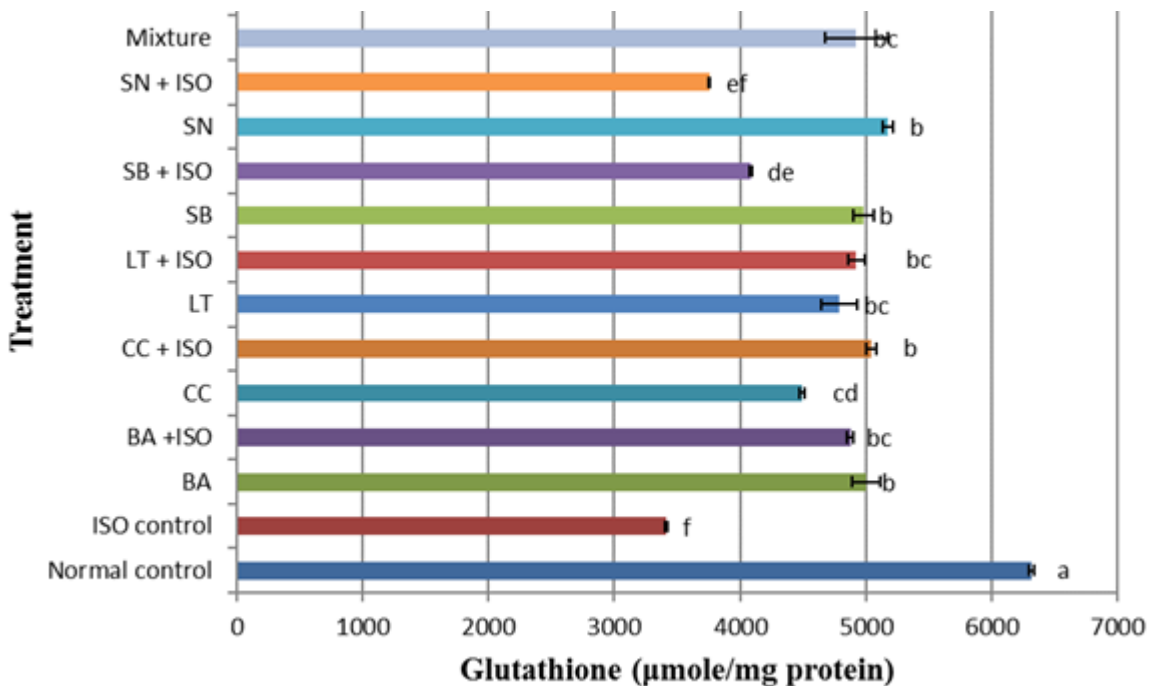
BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio bialfrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.



**Figure 2:** Effects of aqueous leafy vegetables' extracts pre-treatment on catalase (CAT) activity in isoproterenol-induced myocardial infarction rats (vegetable and ISO-treated categories) for 28 days

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.



**Figure 3:** Effects of aqueous leafy vegetables' extracts pre-treatment on glutathione (GSH) concentration in isoproterenol-induced myocardial infarction rats (vegetable and ISO-treated categories) for 28 days

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.

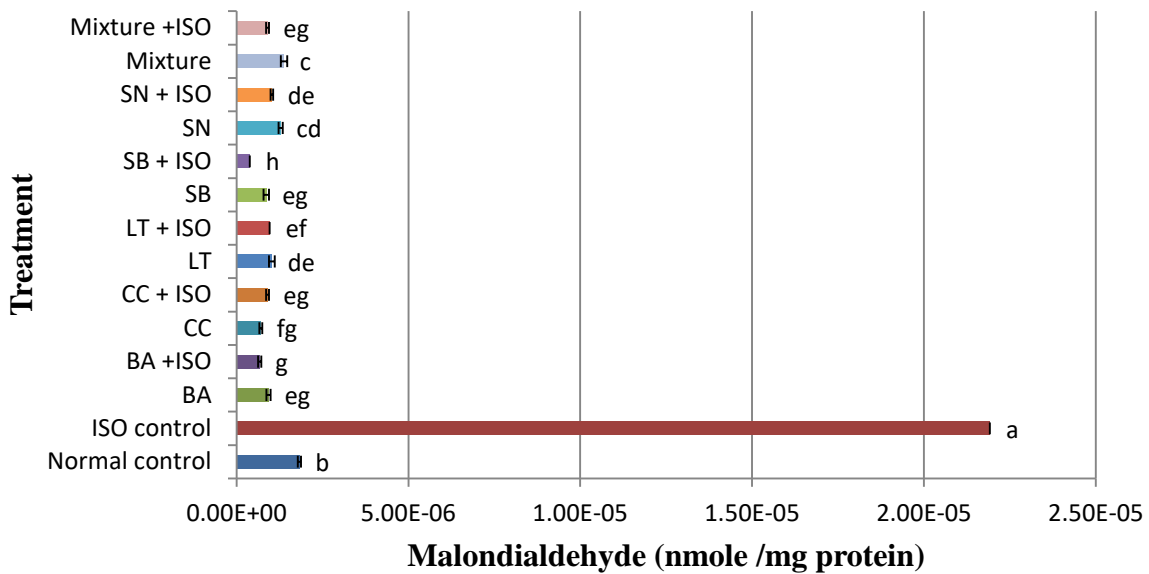


Figure 4: Effects of aqueous leafy vegetables' extracts pre-treatment on malondialdehyde (MDA) concentration in isoproterenol-induced myocardial infarction rats (vegetable and ISO-treated categories) for 28 days

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.

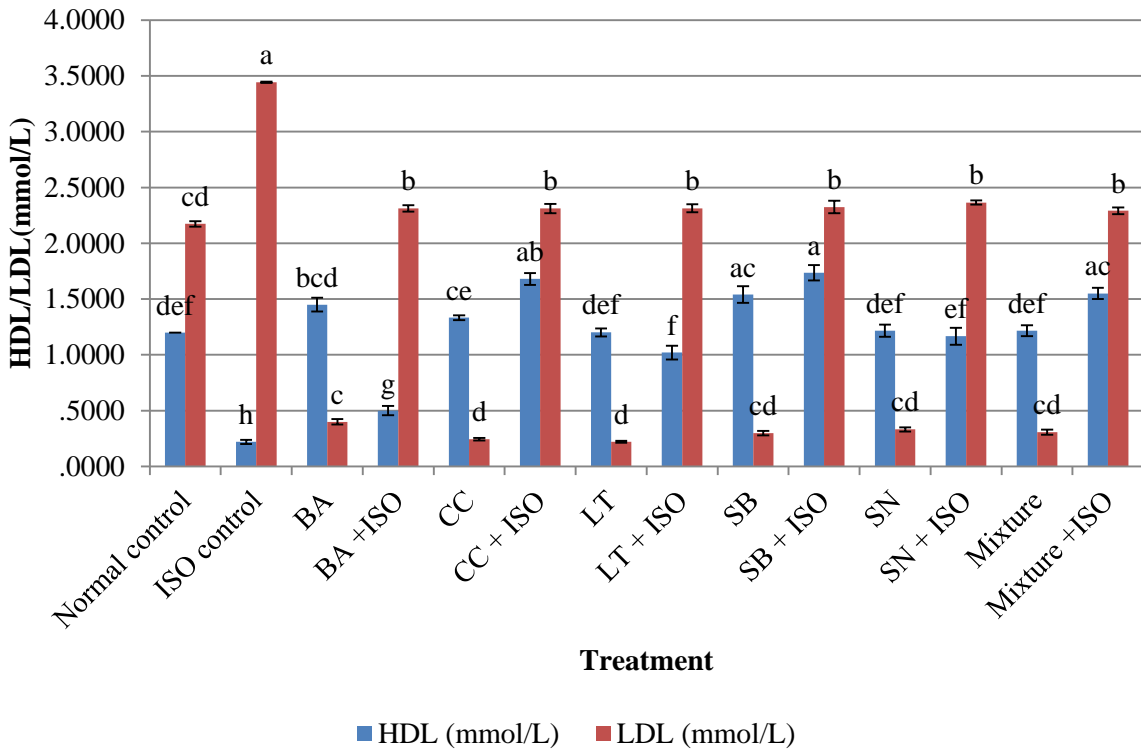
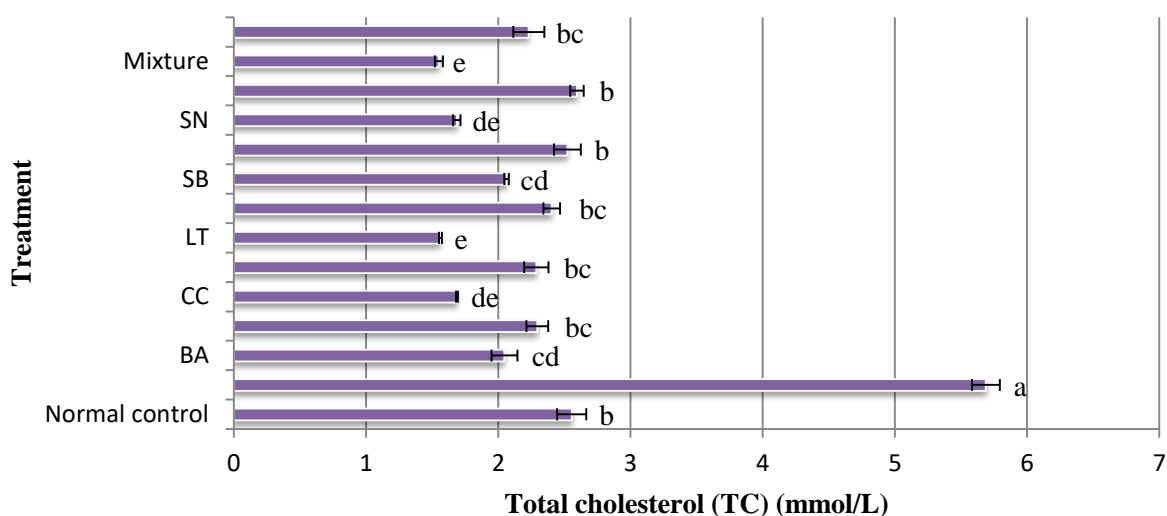


Figure 5: Effects of aqueous leafy vegetables' extracts pre-treatment on lipoprotein lipid profile (HDL: high density lipoproteins and LDL: low density lipoproteins) in isoproterenol-induced myocardial infarction rats (vegetable and ISO-treated categories) for 28 days

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

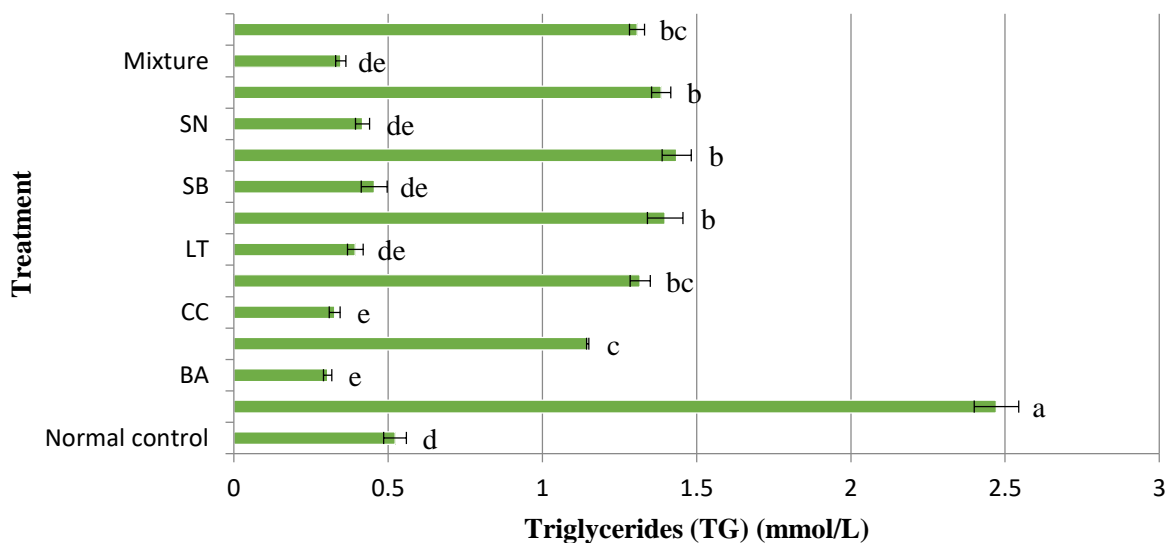
BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.



**Figure 6: Effects of vegetables' extracts pre-treatment (28 days) on TC in vegetable and ISO-treated categories**

Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.

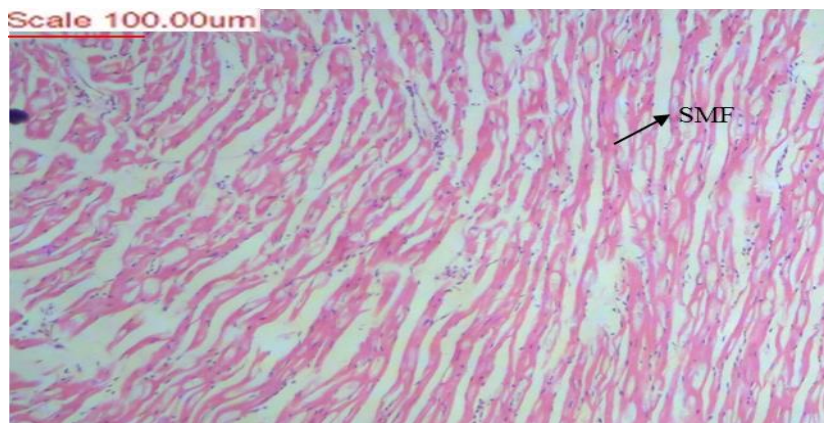


**Figure 7: Effects of vegetables' extracts pre-treatment on TG in vegetable and ISO-treated categories**

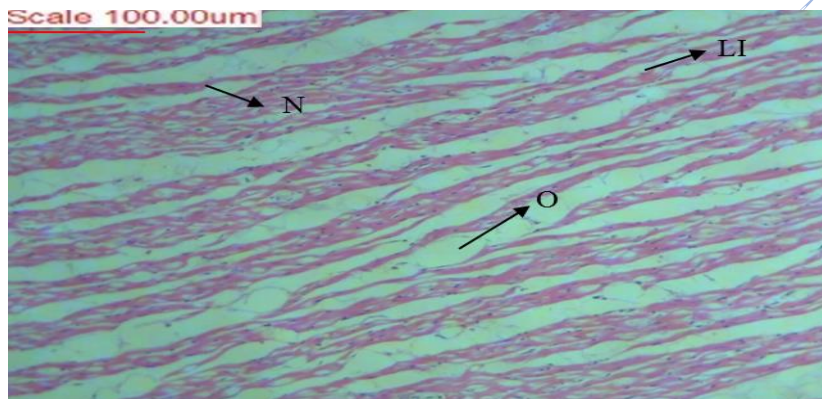
Means without a common letter(s) differ ( $p < 0.05$ ) as analyzed by one-way ANOVA and the LSD test.

BA= *Basella alba*, SN= *Solanum nigrum*, SB= *Senecio biafrae*, CC= *Crassocephalum crepidioides*, LT= *Launaea taraxacifolia*.

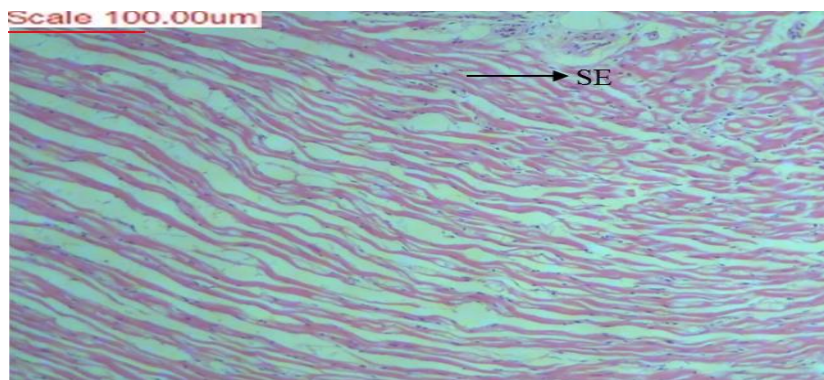




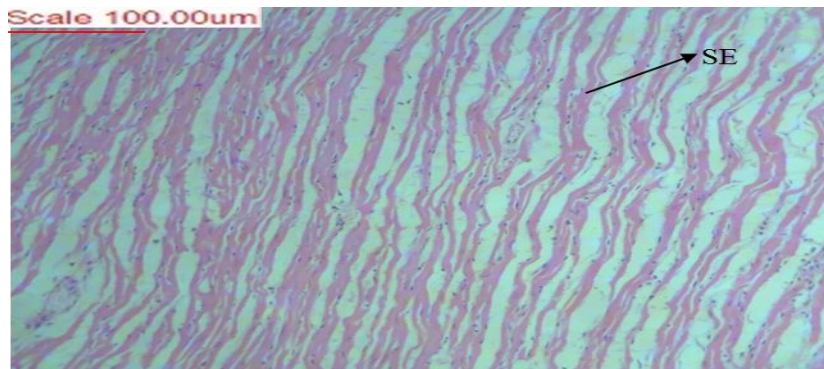
**Figure 8:** A representative photo-micrograph of normal myocardium in normal control. Rats in normal control with no treatment showing normal architecture of heart tissues. Normal heart muscles (MAG X40) with syncytium of myocardial fibres (SMF).



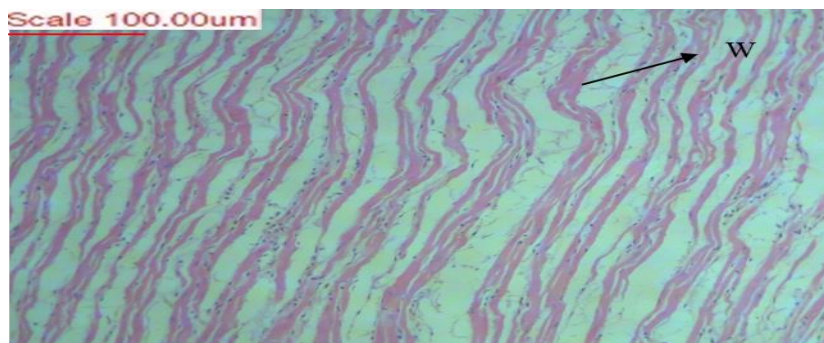
**Figure 9:** A representative photo-micrograph of abnormal myocardium in ISO control. Rats in ISO control showing myocardial infarction architecture of heart tissues associated with slender myocardial fibres damage (O-oedema (widening of interfibre space), LI-Leucocyte infiltration, N-Necrosis (MAG X40). ISO=Isoproterenol.



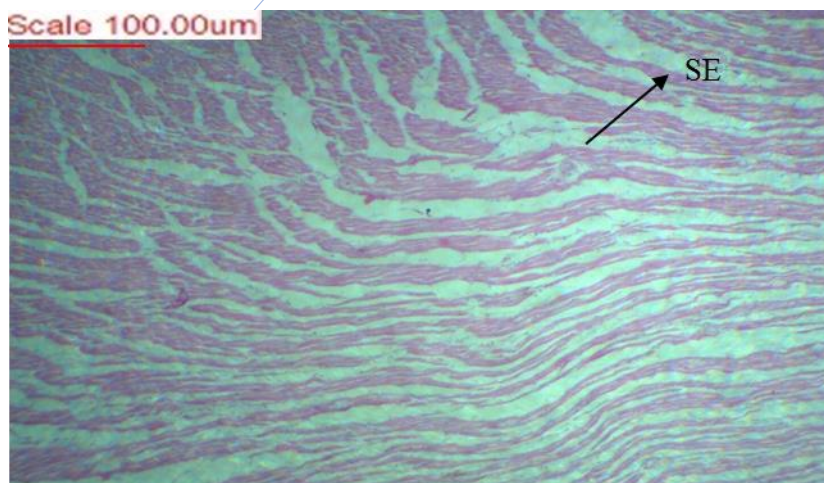
**Figure 10:** A representative photo-micrograph of normal architecture of heart tissues in BA + ISO treated rats. Normal architecture of heart tissues in BA + ISO treated rats was restored due to 28 days pre-treatment of BA. Slightly elongated (SE) myocardial fibres (MAG X40). BA= *Basella alba*, ISO=Isoproterenol.



**Figure 11: A representative photo-micrograph of CC+ISO-treated rats.** Normal architecture of heart tissues in CC+ISO-treated rats was restored due to 28 days pre-treatment of CC. Slightly elongated (SE) myocardial fibres (MAG X40). CC= *Crassocephalum crepidioides*, ISO=Isoproterenol.

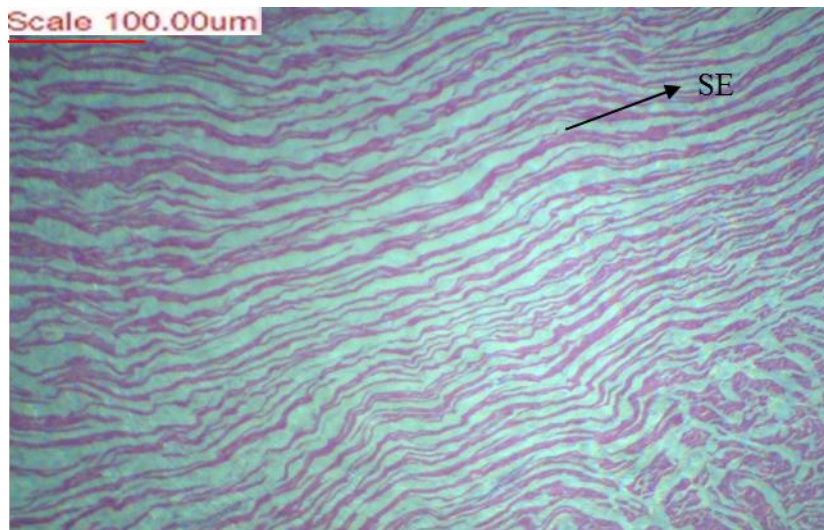


**Figure 12: A representative photo-micrograph of LT + ISO-treated rats.** Near to normal architecture of heart tissues in LT + ISO-treated rats was restored due to 28 days pre-treatment of LT. Wavy (W) myocardial fibres (MAG X40). LT= *Launaea taraxacifolia*, ISO=Isoproterenol.

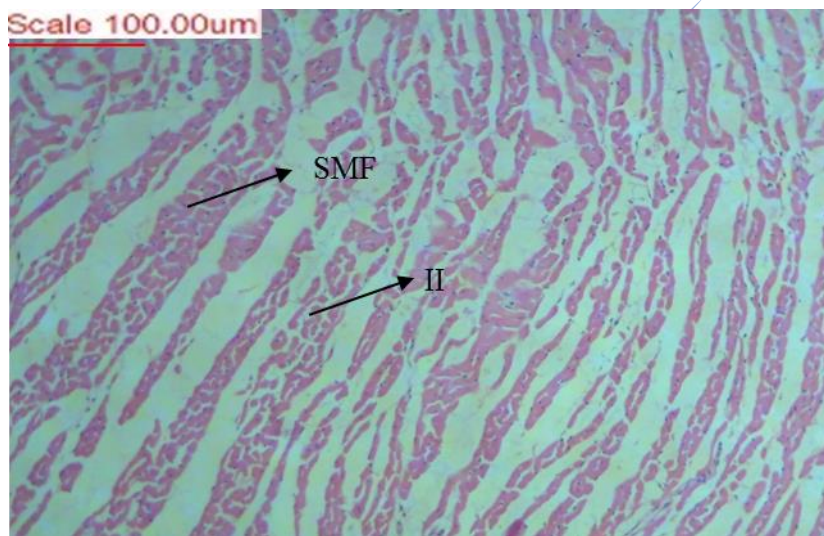


**Figure 13: A representative photo-micrograph of SB+ISO-treated rats.** Normal architecture of heart tissues in SB+ISO-treated rats was restored due to 28 days pre-treatment of SB. Slightly elongated (SE) myocardial fibres (MAG X40). SB= *Sarcocolla bicolor*, ISO=Isoproterenol.





**Figure 14: A representative photo-micrograph of SN+ISO-treated rats.**  
 Normal architecture of heart tissues in SN +ISO-treated rats was restored due to 28 days pre-treatment of SN. Slightly elongated (SE) myocardial fibres (MAG X40). SN= *Solanum nigrum*, ISO=Isoproterenol.



**Figure 15: A representative photo-micrograph of Mixture + ISO-treated rats.**  
 Normal architecture of heart muscles was completely restored with syncytium of myocardial fibres (SMF) and the intervening interstitium (II) (MAG X40) due to 28 days pre-treatment of vegetables' mixture. ISO=Isoproterenol.

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