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Abstract: Hyperlipidemia and atherosclerosis are among major health risk concerns. The aim of the present study was to evaluate the possible improving potentials of *Moringa oleifera* extract (*MOE*), in hydroethanol 30%, on lipid metabolic profile in hyperlipidemic rat model. Rats were rendered hyperlipidemic by 6-week supplementation of high-fat diet *ad libitum*. Rats were assigned into six groups; with different treatments (group-1 received normal diet and kept as normal control; group-2 were fed high-fat (cholesterol 2% + coconut oil 2%) diet for 6 weeks and kept as diseased control; group-3 were kept on high-fat diet but received ezetimibe (1 mg/Kg, orally, daily) and kept as standard; while rats of groups-4, -5 and -6 were kept on high-fat diet and received *MOE* at doses of 200, 400 and 600 mg/Kg, orally, daily, and kept as treated groups. Blood samples for serum were taken for clinicochemical analysis on the days 21 (3 weeks) and 42 (6 weeks) of the experiment and tissue specimens from the aorta were taken for histopathology. *MOE* extract significantly ($P < 0.05$) decreased the elevated serum lipid parameters, including total lipids, tri-acylglycerols (TAGs), cholesterol, LDL-C, VLDL-C, but significantly ($P < 0.05$) normalized the serum HDL-C concentrations of rats kept on high-fat diet. Administration of *MOE* significantly improved atherosclerotic index in samples taken from animals kept on high-fat diet compared to the diseased untreated ones. Histopathological examination of the aortic specimens revealed down accumulation of lipids in the arterial wall. These data may give scientific evidence that *MOE* has a good improving potential on hyperlipidemia and atherosclerosis disease conditions.

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1. Introduction

Among metabolic disorders, dyslipidemia, particularly hypercholesterolemia is considered as a major contributor in cardiovascular disease, including atherosclerosis and atherosclerosis-associated conditions as coronary heart disease, ischemic cerebrovascular disease and peripheral vascular disease (Nelson 2013). The best target for therapy in these conditions is to normalize blood and/or tissue cholesterol and lipid values. The conventional anti-hyperlipidemic chemical drugs are, usually, associated with common some adverse effects such as malaise, gastric irritation, nausea/vomiting/diarrhea, hyperuricemia, muscle aches, red dry skin, and altered liver function (Gurib-Fakim 2006) and (Kumar *et al.*, 2008).

Natural products with health improving impacts constitute, therefore, an interesting research area in minds of concerned personnel, including patients, physicians and researchers based on their safety, efficacy and easy access (Ververidis *et al.*, 2007).

Moringa oleifera, Family *Moringaceae*, is a fast-growing, drought-resistant tree, native to tropical and subtropical regions including KSA, Egypt and India that is considered as the major producer of *Moringa*.

Moringa plant parts, especially leaves, is thought to have a nutraceutical benefits (Leone *et al.*, 2015). Different parts and extracts of the plant were reported to possess variable beneficial health activities, including, pesticide (Ashfaq *et al.*, 2012), antioxidant (Chumark *et al.*, 2008), anti-inflammatory (Coppin *et al.*, 2013), immunomodulatory (Gupta *et al.*, 2010), antidiabetic (Mbikay 2012), hepatorenal protecting (Oyagbemi *et al.*, 2013, Asiedu-Gyekye *et al.*, 2014), and anticancer (Sikder *et al.*, 2013) activities.

The present study was designed to evaluate, using clinicochemical and histopathological tools, the hyperlipidemia and atherogenic index improving profile of *MOE* as a natural remedy to the control of dyslipidemia and atherosclerosis in rat hyperlipidemic model by high-fat diet supplementation.

2. Material and Methods

Moringa oleifera extract

The extract was obtained and standardized according to the classical methodological stepwise described by Harborne (1973) with minor modifications. The green leaves of *the plant* (Figure 1) were collected from our local environment and identified by a plant specialist. Plant parts were

refluxed in running tap water and then with bi-distilled water, shade dried at room temperature and chopped. Extract was prepared by maceration the chopped plant parts (150 g) in a known volume (1.5 Liter) of water/ethanol (70:30, v/v). Maceration continued for 72 hours in refrigerator with frequent shaking. The hydro-ethanolic extract was then strained through muslin mesh and then concentrated using a shaking water bath at 56 °C in a wide-mouthed containers and the semiliquid obtained (yield) was then weighed and re-constituted freshly every day by dissolving in measured amount of bi-distilled water. Three stock solutions were prepared; 20, 40 and 60 mg/mL. *MOE* was administered to rats at dosage rate of 200 (small dose; SD), 400 (medium dose; MD) and 600 (high dose; HD) mg/Kg, daily, adjusted so that each rat (weighing 200 g) receives 2 ml of *MOE* orally using a rat gastric tube to the corresponding groups as explained below.



Figure 1: Green and dry leaves of *Moringa oleifera* plant used for extraction.

Ezetimibe

EZE, a standard inhibitor of cholesterol absorption, (Lipka 2003) used in the present study was kindly gifted by SIGMA pharmaceuticals, Quesna Industrial Zone, Egypt. It was obtained as a pure powder. EZE was dissolved in 20% ethanol; where 10 mg of EZE were dissolved firstly in 10 ml absolute alcohol, and then the alcoholic solution of EZE was completed up to 50 ml by bi-distilled water. Each rat within the target group received 1 ml of the prepared solution which is equivalent to a dosage rate of 1 mg/Kg, orally, once daily (Patel 2004).

Experimental animals and protocol

Thirty-six white male rats, of age 6-8 weeks and of average weight of 200 g were used in a parallel study design. Rats were divided in separate cages and gained access to clean water and diets *ad libitum* at room temperature. After a week for acclimatization, rats were subjected to various experimental treatments, as follows:

Group-1: Rats kept on normal diet and administered no drugs but their vehicles; and served as normal control.

Group-2: Rats kept on high-fat diet and administered no drugs but their vehicles; and served as diseased control.

Group-3: Rats kept on high-fat diet and administered EZE at a dose rate of 1 mg/Kg, orally, once daily, for 6 weeks; and used as a standard anti-hyperlipidemic.

Groups-4,5 & 6: Rats kept on high-fat diet and administered *MOE* at doses of 200, 400 and 600 mg/Kg, orally, once daily, for 6 weeks; and kept as test.

Treatments were conducted simultaneously; and all procedures were ethical to animals and performed with merciful and humane manner under light ether anesthesia.

Sampling

Blood for serum was collected on the 21st and the 42nd days from the start of the experiment. Blood was harvested into plain sampling tubes from the retro-orbital venous plexus using heparinized capillary tubes. The harvested blood was left to coagulate at 37 °C for an hour; and then the clot was allowed to shrink by refrigeration for further an hour. Centrifugation at 1000 *xg* for 10 minutes were performed to separate clean sera which were collected into Eppendorf's tubes using clean and dry Pasteur pipettes. The obtained samples were at -50 °C till analysis of the following clinicochemical parameters: Total lipids (TL), total cholesterol (TC), triacylglycerols (TAGs), high density lipoprotein-cholesterol (HDL-C), low density lipoprotein-cholesterol (LDL-C) and very low density lipoprotein-cholesterol (VLDL-C). On the last day of the experiment, and after blood collection, animals were humanely sacrificed under ether anesthesia, and aortic tissue specimens were taken into 10% formalin solution for histopathological examination.

Clinicochemical analysis

The serum total lipids were determined according to the method described by Chabrol and Castellano (1961) using a kit supplied by Spinreact® (Sant Esteve De Bas, Spain); total cholesterol was determined enzymatically according to the method described by Meiattini *et al.*, (1978) using a kit supplied by Spinreact®; HDL-L was determined according to the precipitation method described by Friedewald *et al.*, (1972) using a kit supplied by Spectrum® (Obour city, Egypt); TAGs were measured enzymatically depending on the method explained after Young *et al.*, (1975) using a kit Spinreact®. LDL-C and VLDL-C values were calculated using the formulae described by Friedewald *et al.*, (1972) and Bauer (1982), respectively. All laboratory steps were

according to the instructions supplied by the kit manufacturers.

Atherogenic index

The Atherogenic index of plasma (AIP) was mathematically calculated according to the formula described by Dobiášová (2004): $AIP = \text{Log} (\text{TAGs} / \text{HDL-C})$

The TAGs and HDL-C parameters must be expressed in molar concentrations, where each 100 mg/dL of TAGs = 1.13 mmol/L; while each 20 mg/dL of HDL-C = 0.518 mmol/L. Outcomes were interpreted as follows: $AIP < 0.11$: Low Risk; $AIP = 0.11-0.21$: Intermediate Risk; $AIP > 0.21$: High Risk.

Histopathological examination

Aortic specimens picked out from dissected rats in all groups were immediately placed in 10% formalin solution; processed for sectioning, staining and microscopical examining as described by Bancroft and Gamble (2008). Samples were allowed to fix for a 24 h period. The fixed samples were then gently washed under slowly running water for overnight. The clean fixed samples were then allowed to dehydrate in a series of increased-concentrations of ethanol starting in 70% and finalizing in absolute alcohol. The fixed, clean, dehydrated samples were placed in xylol for 3 h to clear and then placed in melted paraffin wax tissue boxes. The wax containing tissue specimens was left to solidify and then sections of 4 - 6 μm thickness were obtained using a rotary microtome. Staining procedure started with removal of wax from the microsections by two changes in ethanol (absolute; 5 minutes each). Ethanol was washed away with water. Sections were stained with H & E for 10 minutes, and then the extra stains were gently flushed under running water for 15 minutes. The stained microsections were dehydrated by alcohol series as mentioned above, then allowed to clear in xylol and finally covered with DBX. The prepared microsection slides were examined microscopically and interpreted by a specialist.

Data statistical analysis

Data are presented as mean \pm S.E of 6 observations. The obtained data were statistically

analyzed using repeated-measures ANOVA at two timing points (3 and 6 weeks) with Tukey's post-hoc to determine differences between groups at probability level of 5%. All statistical procedure was done by SPSS software, version 20.

3. Results

Effect of MOE on serum lipid profile

As shown in Tables 1, 2, 3, 5, 6, there were significant increases in serum total lipids, cholesterol, tri-acyl-glycerols, LDL-C, VLDL-C values in samples obtained from rats kept on high-fat diet, compared to those obtained from rats on normal diet. While administration of MOE to control rats revealed insignificant alterations in these parameters throughout the experimental period, yet, its administration significantly ($P < 0.05$) decreased their serum levels in samples of rats kept on high-fat diet in a dose-dependent manner. Nevertheless, as presented in Table 4, there was a significant ($P < 0.05$) decrease in HDL-C values of samples of rats kept on high-fat diet. Such decrease was significantly ($P < 0.05$) increased upon administration of MOE with no significant changes in rats kept on basal diet.

Effect of MOE on Atherogenic index of plasma

Data of the present study (Table 7) demonstrate a significant risk values of API down to 0.1 in samples taken from rats kept on high-fat diet throughout the experiment, compared to those in samples taken from the normal control rats which were kept on normal diet. Administration of MOE significantly ($P < 0.05$) improved API values to the safe limits in animals kept on high-fat diet compared to the diseased ones.

Effect of MOE on arterial wall structure

Data of the present study showed that there were no changes in aortic samples picked out from rats of normal control group. Yet, subjecting experimental rats to high-fat diet (diseased-control group) resulted in fatty infiltration in-between the arterial tunics. The severity and development of such change was impeded, to a great extent, in specimens picked out from MOE-treated groups and EZE-treated rats, as well (Figure 2a, b, c and d).

Table 1: Serum total lipids concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm \text{S.E}$; n=6):

	1	2	3	4	5	6
Day 21	343.20 \pm 11.05	585.67 ^a \pm 22.13	426.24 ^b \pm 16.31	447.33 ^b \pm 23.33	422.33 ^b \pm 21.82	401.20 ^b \pm 15.65
Day 42	358.13 \pm 17.23	753.67 ^a \pm 24.33	448.33 ^b \pm 21.67	495.66 ^b \pm 24.84	443.00 ^b \pm 22.67	425.25 ^b \pm 16.85

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 2: Serum cholesterol concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6
Day 21	70.17 \pm 5.20	162.18 ^a \pm 10.06	98.33 ^b \pm 8.40	110.33 ^b \pm 7.66	97.22 ^b \pm 7.85	81.17 ^b \pm 6.10
Day 42	81.35 \pm 7.78	218.35 ^a \pm 15.67	131.34 ^b \pm 11.55	144.21 ^b \pm 10.78	126.22 ^b \pm 6.80	108.35 ^b \pm 6.78

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 3: Serum Triacylglycerol concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6
Day 21	78.85 \pm 7.84	181.33 ^a \pm 15.36	110.66 ^b \pm 24.53	119.65 ^b \pm 13.70	111.78 ^b \pm 11.06	101.55 ^b \pm 8.66
Day 42	81.12 \pm 10.13	210.67 ^a \pm 18.74	119.16 ^b \pm 10.48	128.35 ^b \pm 14.44	120.32 ^b \pm 13.11	109.12 ^b \pm 13.08

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 4: Serum HDL-C concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6
Day 21	34.67 \pm 3.15	19.82 ^a \pm 1.67	29.46 ^b \pm 2.43	24.89 \pm 3.48	28.33 ^b \pm 1.81	35.85 ^b \pm 3.35
Day 42	41.53 \pm 3.88	16.53 ^a \pm 1.33	34.22 ^b \pm 2.02	28.16 \pm 2.14	36.77 ^b \pm 2.05	42.63 ^b \pm 3.71

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 5: Serum LDL-C concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6
Day 21	16.66 \pm 1.66	66.33 ^a \pm 5.67	31.66 ^b \pm 4.33	45.79 ^b \pm 4.85	32.11 ^b \pm 4.03	17.97 ^b \pm 2.67
Day 42	18.12 \pm 1.43	85.82 ^a \pm 9.12	45.21 ^b \pm 4.65	58.10 ^b \pm 4.54	46.89 ^b \pm 4.15	20.42 ^b \pm 2.36

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 6: Serum VLDL-C concentration (mg/dL) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6
Day 21	24.6 \pm 1.80	49.32 ^a \pm 4.33	33.83 ^b \pm 2.24	38.79 ^b \pm 1.94	32.73 ^b \pm 2.18	25.33 ^b \pm 2.10
Day 42	26.26 \pm 2.37	56.54 ^a \pm 5.28	41.1 ^b \pm 6.65	44.52 ^b \pm 3.37	39.11 ^b \pm 3.11	28.17 ^b \pm 2.03

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

Table 7: Atherogenic index of plasma (AIP) in rats under different treatments, Normal Control (1), Hyperlipidemic control (2), Hyperlipidemic with Ezetimibe 1 mg/Kg, orally (3), Hyperlipidemic with MOE 200, 400 and 600 mg/Kg, orally (4, 5 & 6), daily for 42 days ($\bar{X} \pm S.E$; n=6):

	1	2	3	4	5	6	
Day 21	TAGs (mmol/L)	0.870 \pm 0.008	2.045 ^a \pm 0.15	1.243 ^b \pm 0.07	1.335 ^b \pm 0.08	1.255 ^b \pm 0.075	1.141 ^b \pm 0.066
	HDL-C (mmol/L)	1.140 \pm 0.031	0.513 \pm 0.004	0.764 \pm 0.008	0.644 \pm 0.005	0.738 \pm 0.006	0.932 \pm 0.009
	API Log (TAGs/TC)	0.053 \pm 0.001	0.601 \pm 0.031	0.209 \pm 0.010	0.316 \pm 0.021	0.230 \pm 0.011	0.087 \pm 0.002
Day 42	TAGs (mmol/L)	0.918 \pm 0.009	2.380 ^a \pm 0.081	1.344 ^b \pm 0.046	1.434 ^b \pm 0.052	1.356 ^b \pm 0.067	1.232 ^b \pm 0.033
	HDL-C (mmol/L)	1.062 \pm 0.008	0.427 \pm 0.005	0.888 \pm 0.051	0.738 \pm 0.062	0.958 \pm 0.045	1.087 \pm 0.061
	API Log (TAGs/TC)	0.063 \pm 0.002	0.746 \pm 0.025	0.179 \pm 0.003	0.288 \pm 0.005	0.151 \pm 0.002	0.055 \pm 0.002

Superscripts ^a & ^b mean significantly ($P < 0.05$) different from corresponding Control (1) and Diseased (2), respectively.

4. Discussion

Pathological alterations in the lipid metabolic profile especially long standing hyperlipidemia is a

direct cause of various disease conditions including atherosclerosis (Ross and Harker 1976). The fact is supported by Ross (1986) and Crowther (2005).

Findings of the current study show that hyperlipidemia, induced by 42-day supplementation of high-fat (cholesterol and coconut oil 2% wt/wt, each) diet caused, as expected, marked alterations in the measured lipid parameters (Tables 1~6) of rat groups kept on such diet. In addition, the established dyslipidemia was associated with abnormally elevated Atherogenic index (API > 0.21; Table 7), together with considerable histopathological change in the form of massive fatty infiltration in all arterial tunics (Figure 2). These rats were applied, in the present study, as a model for hyperlipidemia predisposing to atherosclerosis to evaluate the possible protection against atherosclerosis mediated by a hydro-ethanolic extract prepared from *Moringa oleifera* green leaves, in context of its anti-hyperlipidemic potential.

Data presented in this study (Table 1) show a significant ($P < 0.05$) increase in serum total lipid value of rats received high-fat diet throughout the 42 days of the experiment, compared to the normal control rats fed on a balanced diet. Similar findings were reported previously in rats (Csont *et al.*, 2002); (El-Mahmoudy *et al.*, 2013); (El-Mahmoudy *et al.*, 2014) and rabbits (Diaz *et al.*, 2000).

Administration of *MOE* significantly decreased serum total lipid concentration in animals received high-fat diet compared to the diseased untreated ones. The decrease in serum total lipid concentration in *MOE*-treated animals is logic after recording the improved serum values of TAGs, TC, LDL-C and VLDL-C that were observed simultaneously in this study. Lowering serum lipid profile recorded in the present study may be parallel with that reported by (Czerwiński *et al.*, 2004) who found that oat and amaranth meals positively affect plasma lipid profile in rats fed cholesterol-containing diets. The authors attributed such effect to the contents of the bioactive components and the antioxidant activities of the studied plant samples.

Findings presented in the current study (Table 2) show a significant ($P < 0.05$) elevation in serum TC concentration in samples from rats kept on high-fat diet throughout the 42-day experimental period, compared to that of the normal control animals. This finding is in accordance with that achieved by (Diaz *et al.*, 2000) who found that rabbits kept on atherogenic diet exhibited marked elevation in TC in plasma. Elevated serum cholesterol level is expected after high-fat diet supplementation and the inhibited clearing rate of LDL-C from the blood due to some defect in LDL receptors associated with elevated plasma total cholesterol values above normal levels (Zulet *et al.*, 1999). Administration of *MOE* significantly ($P < 0.05$) decreased serum cholesterol concentration in animals received high-fat diet compared to the diseased untreated ones. The hypo-

cholesterolemic effect of *MOE* may be explained on the basis of decreasing cholesterol intestinal absorption with less dietary cholesterol is delivered to the pool of cholesterol to the liver; and/or greater clearance of LDL-cholesterol particles by the liver.

Findings presented in the current study (Table 3) show a significant ($P < 0.05$) increase in TAGs in samples from animals kept on high-fat diet if compared with those of the normal control. Such significant elevation in serum TAGs may be explained on the basis of the diminished activity of lipase - insulin-dependent enzyme- contributing in TAGs clearance from blood by enhancing their hydrolysis to glycerol and free fatty acids (Yost *et al.*, 1995). Daily oral administration of *MOE* significantly decreased serum TAGs concentration in animals received high-fat diet compared to that of diseased untreated ones. This finding may be parallel to those of Maier *et al.*, (2000) and Czerwiński *et al.*, (2004) in women and rats, respectively. The significant decrease in plasma TAGs has been explained previously by Bennani-Kabchi *et al.*, (2000) who related that decrease to the higher rate of lipolysis mediated by enhanced plasma lipase activity. Nevertheless, more earlier, Griffin *et al.*, (1982) stated that the lower level plasma TAGs might also reflect a lower rate of lipogenesis in the liver.

Tables 4,5 and 6 present significant increases in serum LDL-C and VLDL-C and a significant decrease in HDL-C in rats kept on high-fat diet, compared to the corresponding normal control ones. The elevated serum LDL-C and VLDL-C seemed to occur upon over-production of LDL beyond the capacity of LDL-receptors expressed on hepatocyte cell membranes. In addition, the dietary fat and cholesterol may alter the serum lipoprotein pattern and increases the cholesterol content in VLDL (Mahley and Holcombe 1977). *MOE* administration revealed significant decreases in serum LDL-C and VLDL-C concentrations with a significant increase in HDL-C when compared to those of the rats received high-fat diet. Such improving effect of *MOE* may be attributed to the decreased absorption of fats supplemented to rats and/or increased peripheral and hepatic breakdown of cholesterol esters from VLDL-C and LDL-C. The compositional change of HDL-C might be speculated to be due to a probable activation of Lecithin-cholesterol acyltransferase (LCAT) that is stimulated firstly by exogenous cholesterol.

Tables 7 present significant ($P < 0.05$) elevation in API above 0.21 in samples taken from rats kept on high-fat diet, if compared with those of the control, indicating a high exposure risk to atherosclerosis in the former. *MOE* administration significantly protected against elevations of API compared to the diseased untreated rats in a dose dependent manner. The extract improved the treated rats' status from high

to moderate and to mild risk for exposure to atherosclerosis, dose-dependently. This protecting effect was explained on the bases of improved lipid profile, namely decreased TAGs and increased HDL-C concentrations in the extract-treated groups. Supportive studies conducted by Adisakwattana and Chanathong (2011) and Toma *et al.*, (2014) indicated that MOE decreases lipid absorption by inhibiting pancreatic cholesterol esterase, pancreatic lipase, cholesterol micellization and bile binding capacity *in vitro*.

Histopathological findings come supportive to the biochemical analysis, where the fatty degenerative changes observed in aortic specimens from rats kept on high-fat diet were gradually diminished upon concurrent administration of MOE (Figure 2). Atherosclerosis extent and type was categorized morphometrically by Gore and Tejada (1957). According to the type of the lesion, the authors graded

atherosclerosis as follows: lipid streaks, spots and patches as Grade-I; elevated smoothly surfaced fibrous plaques of variable lipid content as Grade-II; plaques with ulceration, necrosis or hemorrhage as Grade-III; and finally calcified plaques as Grade-IV. According to the extent of lesions, the authors graded atherosclerosis as follows: less than 5% or negligible as Grade-0; 6-15% or minimal as Grade-A; 16-33% or mild as Grade-B; 34-50% or moderate as Grade-C; and finally more than 50% or severe as Grade-D. By applying this appraisal, histopathological findings in the present study may approximately indicate that MOE, especially at the large dose, has improved the atherosclerosis/risk from Grade-II/D to Grad-I/A.

The recorded beneficial effects of MOE may be related to the active pharmacological constituents present in the extract, including flavonoids, glucosinolates, catechins, phenolics, tannins and gallic acid (Stohs and Hartman 2015).

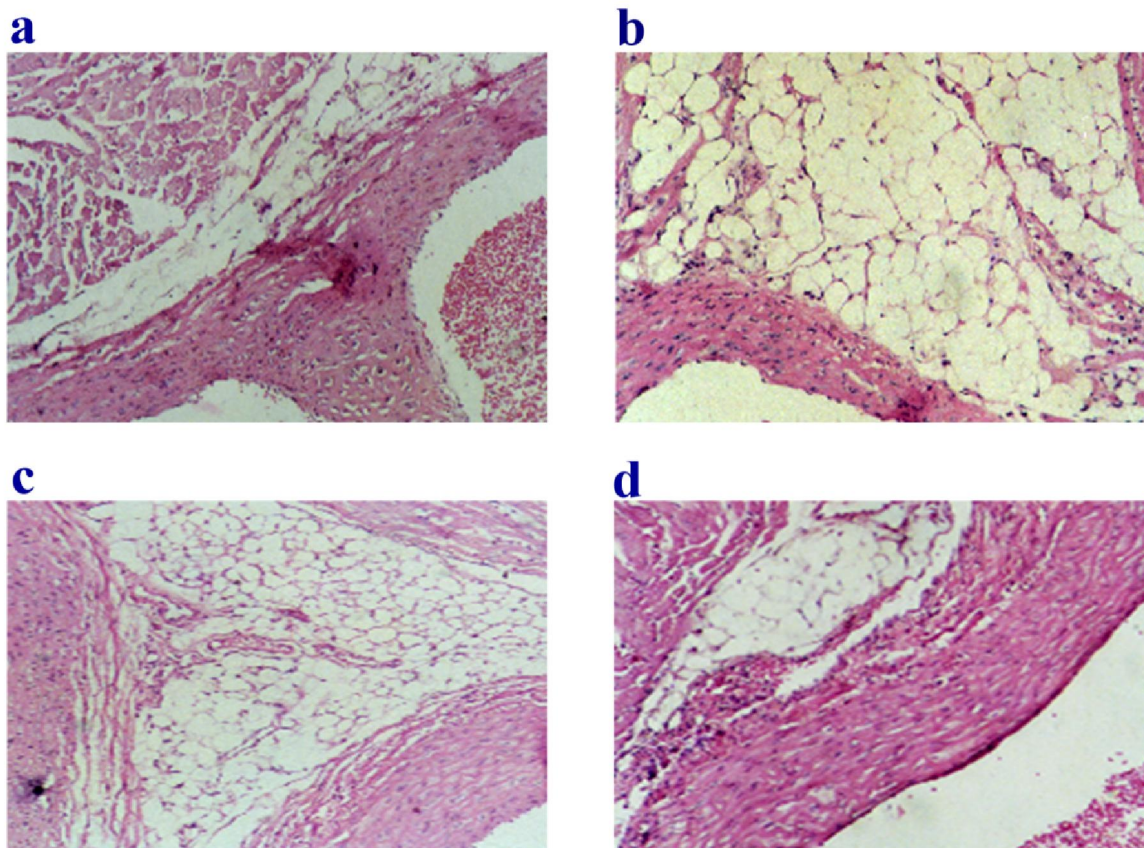


Figure 2: Histopathological picture of aortic sections from normal (a, showing no microscopic alterations), hyperlipidemic (b, showing massive fatty infiltration and vacuolar degeneration), EZE-treated (c, showing moderate fatty infiltration) and 600 mg/kg MOE-treated (d, showing mild fatty infiltration) rats.

A perusal of entries reported in table 1 reveals statistically significant difference in positive mental health of elderly females on the basis of physical

activity status. It was observed that positive mental health of elderly females engaged in regular physical activity was significantly superior as compared to

elderly females with sedentary lifestyle. The calculated $t=22.09$ also proves their finding scientifically at 0.01 level of statistical significance.

Beyer et al. (2002) in a study reported that regular participation in physical activity enhance the non-adrenaline levels which regulates mood and this positive mood changes are also seen in behaviour and thinking of an individual. After continued physical activity person accepts his/her weakness and becomes emotionally more stable. These reflect in positive mental health of an individual.

In conclusion, the present findings may suggest that *Moringa oleifera* extract may protect against hyperlipidemia and development of atherosclerosis, indicated by clinicopathological analysis. The extract, therefore, may have a good health impact in dyslipidemia and related disease conditions.

No-Conflict-Interest

The authors hereby declare that there is no conflict of interest related to the present study.

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