Caliphate Journal of Science & Technology (CaJoST)

ISSN: 2705-313X (**PRINT**); 2705-3121 (**ONLINE**) **Research Article**

Open Access Journal available at: https://cajostssu.com and https://www.ajol.info/index.php/cajost/index

DOI: https://dx.doi.org/10.4314/cajost.v4i1.10

Article Info

Received: 21st August 2021

Revised: 26th January 2022

Accepted: 29th January 2022

Multi-plant (*Phoenix dactylifera* and *Cyperus esculentus*) Effects on Regulating Diet Induced-hypercholesterolemia in Female Sprague-Dawley Rats

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Hypercholesterolemia also known as amplified blood cholesterol represent other forms of hyperlipidemia secondary to atherosclerosis is rising fast in our society. Natural remedies without potential harmful effects are increasing as well. It is against this backdrop that we investigated combined effects of *Phoenix dactylifera* and *Cyperus esculentus* against diet induced hypercholesterolemia in rats. In-bred, healthy non-pregnant female Sprague-Dawley rats (11-17weeks, 153-177g) were grouped and orally administered (A1, B1, and C1 =300mg/kg b.w.; A2, B2, and C2 =600mg/kg b.w.; A3, B3, and C3 =900mg/kg b.w.; A4, B4, and C4 =1200mg/kg b.w and D = untreated, n=3) daily for 45days by oral intubation. Polypropylene cages in a sanitized aerated facility, bedded with sawdust housed all animals. They observed 7days adaptation to environmental temperature (25±5ºC), humidity (45±5%), and photoperiod (12:12 hr. day/night). Rats consumed high fat-dietary cholesterol diet to induce hypercholesterolemia and water provided *ad libitum*. Sera were used for lipid profiling (before, during, and after) following varying treatments plan. Lipid profile: TC (total cholesterol), ST (serum triglycerides), LDLC (low-density lipoprotein cholesterol), HDLC (high-density lipoprotein cholesterol) were abnormal in pre-experimental phases (both test and control). Values were regulated after treatment particularly in 1200mg/kg b.w in rats administered *P. dactylifera*: TC50.17±2.7/dL, ST27.7±0.6mg/dL, LDL23.2±2.5mg/dL, HDL89.1 ± 3.9/dL; *C. esculentus*: TC41.03±2.3mg/dL, ST27.4±2.9mg/dL, LDL27.1±1.3mg/dL, HDL94.3±1.8mg/dL; and mix: TC32.77±3.8mg/dL, ST23.5±2.4mg/dL, LDL21.3±2.9mg/dL, HDL97.8±3.9mg/dL excluding control: TC92.62±0.3mg/dL, ST71.3±1.9mg/dL, LDL64.0±0.2mg/dL, and HDL21.3±1.1mg/dL ($p < 0.05$). Therefore, synergy of *P. dactylifera* and *C. esculentus* regulate hypercholesterolemia in rats while *C. esculentus* particularly is the super active constituent in the mix.

Cite this: *CaJoST*, 2022, 1, 78-86

Keywords: *C. esculentus*, *P. dactylifera*, lipid profile, hyperlipidemia, medicinal crops, experimental rats

1. Introduction

Hypercholesterolemia is also known as high cholesterol content in blood (Durrington, 2003). Hypercholesterolemia is another form of hyperlipidemia, which indicates the onset of abnormal lipid metabolism secondary to manifestation and progression of atherosclerosis (Sinha and Gosh, 2018). Death rate resulting from coronary heart disease has been globally reported, and epidemiologists suggest that there is a link to hypercholesterolemia being a major predisposing factor (Saryono *et al.,* 2017). Increased level of cholesterol results to atherosclerosis due to an elevated atherogenic index leading to poor blood circulation and subsequent to untimely death of heart muscles (Ghuge and Zine, 2012). A rise in total cholesterol level and low density lipoprotein (LDL) increases the risk of coronary heart disease and has been linked to high intake of saturated fatty meals, low intensity of physical activities including unhealthy lifestyle in humans (Madamanchi *et al.,* 2005). The use of herbs in regulating high cholesterol in blood or abnormal lipid parameters subsequent to hyperlipidemia has been useful with improved activities through regulation of varying mechanisms of action (Sinha and Gosh, 2018). A high consumption of fruits and vegetables is beneficial to human health by preventing chronic diseases like: atherosclerosis, cancer, cardiovascular,

diabetes, and neurodegenerative diseases (Saryono *et al.,* 2016). The antioxidant content in fruits and vegetables may decrease the risk of chronic diseases and protect human health (Saryono *et al.,* 2016). Nonetheless, natural remedies are increasingly used to resolve complicated health issues believing that nature has no potential harmful effects to humans. Among the numerous medicinal plants are *P. dactylifera* popularly called date palm fruit and *C. esculentus* known globally as tiger nut. Date palm fruit belongs to the family *Arecaceae*, while the component parts have anticancer, antioxidant, hepatoprotective, antidiabetic, anti-inflammatory, antibacterial, antifungal and antiviral activities (Lim, 2012).

P. dactylifera is an important traditional crop in Iraq, Arabia, North Africa and Morocco where it is reportedly used in treating various illnesses including atherosclerosis and coronary heart disease (Saryono *et al.,* 2017). Phytochemicals of *P dactylifera* suggest that it is rich in antioxidants, vitamins, steroids, flavonoid, saponins, and simple sugars (Ezeh *et al.*, 2014). On the other hand, *C. esculentus* from *Cyperaceae* family propagated by rhizome, basal bulb, and tuber is a pleasurable and exciting crop to taste (Ogeyemhe *et al.,* 2018). The crop is commonly known by several names like chufa, earth almond, and tiger nut in English considered as one of the earliest food sources known to humanity (Ezeh *et al.*, 2014). Tiger nut juice has been reportedly used in preventing arteriosclerosis, since its consumption can help to prevent heart problems and thrombosis and activate blood circulation mainly because it contained unsaturated fatty acid, which is similar to olive oil (Chukwuma *et al.* 2010). There is a clear distinction in the individual use of the plant as against the mix, which is yet to be harnessed for regulating lipid parameters either in animals or humans. This is a novel study as far as we are aware being that most literature we came across focused solely on either *P. dactylifera* or *C. esculentus* against other ailments. It is against this backdrop that we investigated the multi-plant effects of *P. dactylifera* and *C. esculentus* mix against diet induced hypercholesterolemia toxicity in rat's model.

2. Materials and Methods

2.1 Collection of Plant Materials

Six hundred and twelve grams of *P. dactylifera* and seven hundred and eighty-two grams of *C. esculentus* were purchased from Hausa quarters at Aduwawa cattle market in Benin City, Nigeria. Plant materials were identified and authenticated by an expert taxonomist at the Department of Plant and Biotechnology, University of Benin, Nigeria. Plants were assigned voucher referencing UBH_P191 and UBH_C192 respectively.

2.2 Preparation and Extraction Process

Plant materials were washed in a basket under running tap water and left to drain dry in the laboratory, shaded from direct sunlight for 24 hours and oven dried thereafter. Extracts of *P. dactylifera* and *C. esculentus* were obtained by pulverizing samples of each plant with an electric blender (Kenwood 1.6L, BL480 Prestons, Australia) repeatedly till it achieved a smooth powdery substance following the methods of extraction earlier published (Ogeyemhe *et al.* 2018; Ogeyemhe *et al.,* 2019). Multi-plant extract was obtained by mixing extracts of *P. dactylifera* and *C. esculentus* in equal proportion (Ogeyemhe *et al.,* 2019).

2.3 Experimental animals

In-bred and healthy non-pregnant female Sprague Daley rats of about 11-17weeks, weighing 153-177g were obtained from the animal facility of the Department of Animal and Environmental Biology, University of Benin, Nigeria after obtaining ethics approval for the study. Males were excluded to minimize physiological interactions in female rats resulting from animal mating. Rats were handled in line with international best practices for animal experimentation (NRC, 2011); and maintaining the rights of animals updated in the committee's guides for the care and usage of experimental rat's gazette (2011). Rats were housed in a sanitized environment were animals are nursed from birth. Aerated cages were designed with polypropylene and bedded with wood-dust, which are often cleaned mornings and evenings to maintain same sanitary condition from start to finish. Rats observed 14days adaptation to environmental temperature (25±5ºC), humidity (45±5%) and photoperiod (12:12 hr. day/night). Rats had unrestricted access to specially arranged animal fat containing feeds and water at ease.

2.4 Hypercholesterolemia in rats

Animal feeds contained huge proportion of fat constituents (saturated and trans fats) including extremely high dietary cholesterol, specifically demanded from Livestock Feeds PLC, Lagos. Feeds were fed to experimental rats throughout the experiment to sustain hypercholesterolemia.

2.5 Experimental Design and Conduct

Due to the large number of groups in this study (13 cages), n=3 rats were utilized to minimize the use of experimental animals according to lay down regulations (NRC, 2011). Hence, Rats were grouped and administered extract for 45days by oral intubation in the following order: Group D - untreated group serving as control

Group A_1:- administered 300mg/kg b.w. of *P. dactylifera* extract orally per day
Group A_2:- administered 600mg/kg b.w. of *P. dactylifera* extract orally per day
Group A_3:- administered 900mg/kg b.w. of *P. dactylifera* extract orally per day
Group A_4:- administered 1200mg/kg b.w. of *P. dactylifera* extract orally per day
Group B_1:- administered 300mg/kg b.w. of *C. esculentus* extract orally per day
Group B_2:- administered 600mg/kg b.w. of *C. esculentus* extract orally per day
Group B_3:- administered 900mg/kg b.w. of *C. esculentus* extract orally per day
Group B_4:- administered 1200mg/kg b.w. of *C. esculentus* extract orally per day
Group C_1:- administered 300mg/kg b.w. of combined extract orally per day
Group C_2:- administered 600mg/kg b.w. of combined extract orally per day
Group C_3:- administered 900mg/kg b.w. of combined extract orally per day
Group C_4:- administered 1200mg/kg b.w. of combined extract orally per day

2.5.1 Sub-Acute toxicity study

Following earlier reports on plant extract, the present study adopted LD_{50} described in a similar study while sub-acute toxicity test was conducted in line with a modified version of Lorke's method for multi-plant extract (Ogeyemhe *et al.* 2018; Ogeyemhe *et al.,* 2020).

2.5.2 Empirical and Physical Measurements

We measured all experimental rats before and after experimentation with a digital Mettler weighing balance (Mettler Toledo Type BD6000, Mettler-Toledo GmbH, Greifensee, Switzerland). Differences after in reference to weight before experimentation were calculated per group. All animals were monitored sporadically on daily basis for physical signs of abnormality both in behavior and activity for the entire period.

2.5.3 Collection of Blood Samples

Blood samples were collected from all animals before and halfway to the experiment from the marginal ear vein using a 2ml needle and syringe to determine lipid profile of all animals with a lipid panel diagnostic kit (Solana Health Inc. USA), which serve as a pointer to whether hypercholesterolemia was induced before and sustained during experimentation. In the end, all animals fasted all-night and were sacrificed by cervical dislocation. Again, blood samples were collected aseptically via cardiac puncture, and were immediately emptied into a sterile centrifuge tube, which was left to clot at room temperature for 1hr. Samples were later spun (3,000rpm) for ten minutes with a centrifuge (BROADBENT, UK). This process was conducted at room temperature and stored at -20ºC until it was needed for lipid profile testing. Enzymatic end point method was used to evaluate the lipid profile: serum triglycerides (ST), total cholesterol (TC), high density lipoproteins cholesterol (HDLC) and low density lipoproteins cholesterol (LDLC).

2.5.4 Estimation of Lipid Profile

Sera for fasting blood was obtained by centrifuging clotted blood as described earlier. Chemical analyzer (Erba Chem 5X Analyzer) based on spectrophotometric principle was applied for lipid analysis: serum triglycerides (ST), total cholesterol (TC), high density lipoproteins cholesterol (HDLC) and low density lipoproteins cholesterol (LDLC). Commercialized kits (Solana Health Inc. USA) were used with a wavelength of 546 nm and an optical path of 1cm respectively.

2.5.5 Histopathology

Organs like liver, kidney, pancreas and heart, said to be associated with hyperlipidemia disorder (Csonka *et al.,* 2017) were excised and grossed accordingly. About 3-5mm thickness of tissues were cut and processed histologically. Five microns of serial sections were cut with a rotatory microtome (Hestion ERM 4000 Germany). Sections were deparaffinized and stained according to H&E method, and viewed with a light Binocular microscope®.

2.5.6 Data processing and analysis

Result expressions are presented as means ± S.E.M. Data are analyzed using Instat Statistic Package version 3. Analyses ANOVA was used to compare the mean differences between and within the groups. Dunneth's post hoc test compared the means within experimental groups and against varying controls including before, during and after experimentation. Variants within the groups were considered statistically significant at $P \leq 0.05$.

3. Results and Discussion

Hyperlipidemia associated lipid disorders are considered to cause atherosclerotic cardiovascular diseases ranked as one of the risk factors contributing to prevalence and severity of coronary heart diseases (Niharika, 2017). It is characterized by elevated serum total cholesterol, low density lipoprotein cholesterol (LDLC), very low density lipoprotein cholesterol (VLDLC) and decreased high density lipoprotein cholesterol (HDLC) respectively (Mushtaq *et al*., 2017). Cholesterol is abundant in the liver, adrenal glands as well as the brain and the nervous system, but the liver has the capability of synthesizing sufficient amount of cholesterol for

normal body functions carried in the blood as lipoprotein while streaming in the body (Osmund, 2000). This may suggest partly why histopathology of liver, kidneys, pancreas and heart in this study did not show pathological concerns as against the report that these organs are often affected in end-stage hyperlipidemia through metabolic and biologic activities (Csonka *et al.,* 2017). Considering that hypercholesterolemia was induced with diet, we suggest that exerted impacts may be insufficient to cause any pathological effects. There were no observable toxicological signs in experimental animals, no morbidity and mortality before, during and after experimentation while mixed extract was found to have a wide safety margin. *P. dactylifera* treated rats showed remarkable weight gain in all treatment groups while *C. esculentus* showed slight gain in body weight only in animals that consumed low doses of extract which suggests that *C. esculentus* may support weight loss if included in diet plans in large quantity. This observation is strongly supported by Ogeyemhe *et al.,* (2019) in which similar weight losses were reported. Though negligible weight gain was demonstrated in rats treated with mixture of extracts (Table 1), which may translate to both plants being competitively antagonizing one another (Ogeyemhe *et al.,* 2020).

Table 1: Empirical and Physical Measurement of Experimental Rats

EXTRACT	Groups	Dosages (mg/kg) b.w.	Initial Average Weight (g)	Final Average Weight (g)	Physical Activities
Distilled water	D	0	153.21± 1.9	237.28±3.6	±
P. dactylifera	A1	300	159.32± 2.7	173.24±2.8	+
P. dactylifera	A2	600	161.17±1.8	174.12±3.5	++
P. dactylifera	A3	900	172.25± 2.6	184.33±1.4	++
P. dactylifera	A4	1200	181.16±2.4	192.72±2.7	++
C. esculentus	B1	300	155.77±1.9	163.66±2.8	+
C. esculentus	B2	600	163.22±3.3	170.86±3.7	+
C. esculentus	B3	900	168.02±1.3	173.74±1.5	++
C. esculentus	B4	1200	176.29±3.8	181.62±1.9	++
Combined extract	C1	300	161.15±1.3	166.11±2.6	+
Combined extract	C2	600	162.62±1.1	166.83±2.7	++
Combined extract	C3	900	170.03±3.9	175.11±1.1	++
Combined extract	C4	1200	176.83±4.5	180.02±1.8	++

All values expressed as mean ± standard error of the mean, while 0mg/kg = control.
Combined extract = P. dactylifera + C. esculentus; body weight = b.w.

Table 2: Serum Lipid Panels of Experimental Animals Before Administration

Grouping	TC mg/dL	ST mg/dL	LDLC mg/dL	HDLC mg/dL
0mg/ kg	61.61 ± 0.3	47.3 ± 1.9	41.0 ± 0.2	33.0 ± 2.1
P. dactylifera				
300mg/ kg b.w. / day	65.14 ± 3.02	45.7 ± 2.8	43.9 ± 4.1	35.2 ± 4.5
600mg/ kg b.w. / day	54.22 ± 1.05	49.8 ± 0.8	40.8 ± 3.1	32.7 ± 3.6
900mg/ kg b.w. / day	57.96 ± 1.33	51.1 ± 2.7	47.2 ± 3.9	30.6 ± 2.5
1200mg/ kg b.w. / day	61.77 ± 3.48	48.2 ± 5.6	47.9 ± 3.7	29.4 ± 1.3
P-value	0.178	0.163	0.189	0.127
C. esculentus				
300mg/ kg b.w. / day	63.01 ± 2.4	49.1 ± 4.4	40.2 ± 2.8	34.6 ± 1.5
600mg/ kg b.w. / day	63.17 ± 2.7	47.3 ± 1.3	43.75 ± 0.1	33.9 ± 3.1
900mg/ kg b.w. / day	64.82 ± 3.6	47.4 ± 1.3	48.77 ± 3.5	31.3 ± 1.5
1200mg/ kg b.w. / day	62.53 ± 2.8	49.5 ± 2.6	46.88 ± 1.7	36.3 ± 2.2
P-value	0.165	0.117	0.167	0.145
Mixed extract				
300mg/ kg b.w. / day	61.83 ± 3.9	49.5 ± 0.6	43.5 ± 2.7	33.8 ± 2.9
600mg/ kg b.w. / day	61.62 ± 2.3	46.7 ± 4.1	51.07 ± 2.2	34.4 ± 1.3
900mg/ kg b.w. / day	59.13 ± 1.8	48.1 ± 2.2	45.2 ± 4.4	29.8 ± 2.5
1200mg/ kg b.w. / day	63.77 ± 3.8	49.7 ± 1.2	44.8 ± 2.8	36.3 ± 1.1
P-value	0.113	0.165	0.243	0.215

Values are expressed as mean ± standard error of the mean, while 0mg/kg = control
Values are not statistically significantly at P ≥ 0.05 (one-way ANOVA) before treatment options
Note: Lipid profile was calculated in mg/dL in all the parameters. TC = total cholesterol;
ST = triglycerides; LDL= low density lipoprotein; HDL= High density lipoprotein
Mixed Extract (P. dactylifera + C. esculentus) in equal proportion

Table 3: Serum Lipid Panels of Experimental Animals Midline (During) Administration

Grouping	TC mg/dL	ST mg/dL	LDLC mg/dL	HDLC mg/dL
0mg/ kg	79.17 ± 0.3	57.3 ± 1.9	49.0 ± 0.2	27 ± 2.1
P. dactylifera				
300mg/ kg b.w. / day	62.03 ± 3.15	41.0 ± 3.3	39.3 ± 2.2	42.1 ± 3.3
600mg/ kg b.w. / day	51.98 ± 0.33	44.3 ± 2.1	31.3 ± 2.9	59.9 ± 2.4
900mg/ kg b.w. / day	56.13 ± 2.11	44.1 ± 2.2	33.7 ± 0.6	62.8 ± 3.2
1200mg/ kg b.w. / day	60.05 ± 2.03	43.8 ± 1.2	34.6 ± 9.1	66.3 ± 3.4
P-value	0.053	0.011	0.146	0.051
C. esculentus				
300mg/ kg b.w. / day	50.56 ± 3.13	46.6 ± 2.1	41.36 ± 1.1	53.4 ± 2.3
600mg/ kg b.w. / day	46.03 ± 1.18	41.2 ± 2.4	40.28 ± 2.8	57.7 ± 3.9
900mg/ kg b.w. / day	53.21 ± 2.32	41.1 ± 0.1	33.6 ± 1.7*	71.7 ± 2.2*
1200mg/ kg b.w. / day	51.82 ± 3.49	42.1 ± 2.7	31.28 ± 2.5*	71.9 ± 2.5*
P-value	0.157	0.111	0.025	0.053
Mixed extract				
300mg/ kg b.w. / day	56.58 ± 1.1	43.0 ± 3.7	36.0 ± 0.3	60.8 ± 4.9
600mg/ kg b.w. / day	53.04 ± 3.5	40.5 ± 1.8	46.0 ± 0.2	63.2 ± 4.1
900mg/ kg b.w. / day	46.05 ± 0.1*	41.9 ± 0.8	36.2 ± 1.4	67.1 ± 3.9*
1200mg/ kg b.w. / day	42.29 ± 2.7*	44.4 ± 2.3	33.8 ± 2.8	74.2 ± 2.8*
P-value	0.011	0.065	0.076	0.004

Values are expressed as mean ± standard error of the mean, while 0mg/kg = control
*Significantly different from values before treatment and control groups at $p \leq 0.05$
Note: Lipid profile was calculated in mg/dL in all the parameters. TC = total cholesterol; ST = triglycerides; LDL= low density lipoprotein; HDL= High density lipoprotein
Mixed Extract (P. dactylifera + C. esculentus) in equal proportion

Table 4: Serum Lipid Panels of Experimental Animals After Administration

Grouping	TC mg/dL	ST mg/dL	LDLC mg/dL	HDLC mg/dL
0mg/ kg	92.23 ± 1.3	71.3 ± 1.9	64.0 ± 0.2	21.3 ± 1.1
P. dactylifera				
300mg/ kg b.w. / day	55.93 ± 2.6	33.8 ± 1.5	28.7 ± 1.3	68.3 ± 2.8
600mg/ kg b.w. / day	47.58 ± 2.5	31.6 ± 0.3*	22.4 ± 1.1*	69.8 ± 3.6*
900mg/ kg b.w. / day	48.84 ± 1.4*	32.3 ± 3.5*	24.8 ± 2.4	76.7 ± 2.5*
1200mg/ kg b.w. / day	50.17 ± 2.7*	27.7 ± 0.6*	23.2 ± 2.5*	89.1 ± 3.9*
P-value	0.041	0.028	0.036	0.001
C. esculentus				
300mg/ kg b.w. / day	47.11 ± 1.6	31.1 ± 1.4	29.2 ± 0.1	71.9 ± 3.7*
600mg/ kg b.w. / day	41.17 ± 1.1	35.2 ± 2.2	31.0 ± 1.2	74.6 ± 1.6*
900mg/ kg b.w. / day	42.99 ± 2.9*	26.7 ± 0.1*	29.1 ± 1.2*	83.9 ± 0.2*
1200mg/ kg b.w. / day	41.03 ± 2.3*	27.4 ± 2.9*	27.1 ± 1.3*	94.3 ± 1.8*
P-value	0.001	0.003	0.134	0.001
Mixed extract				
300mg/ kg b.w. / day	41.62 ± 1.3*	31.6 ± 0.4	25.4 ± 2.9	75.5 ± 2.6*
600mg/ kg b.w. / day	44.45 ± 2.6	29.3 ± 1.9*	27.3 ± 1.1	79.5 ± 2.2*
900mg/ kg b.w. / day	33.13 ± 3.8*	38.3 ± 1.5	25.2 ± 1.2	87.1 ± 1.3*
1200mg/ kg b.w. / day	32.77 ± 2.4*	23.5 ± 2.4*	21.3 ± 2.9*	97.8 ± 3.9*
P-value	0.004	0.001	0.016	0.001

Values are expressed as mean ± standard error of the mean, while 0mg/kg = control
*Significantly different from values midway to treatment and control groups at $p \leq 0.05$
Note: Lipid profile was calculated in mg/dL in all the parameters. TC = total cholesterol; ST = triglycerides; LDL= low density lipoprotein; HDL= High density lipoprotein
Mixed Extract (P. dactylifera + C. esculentus) in equa proportion

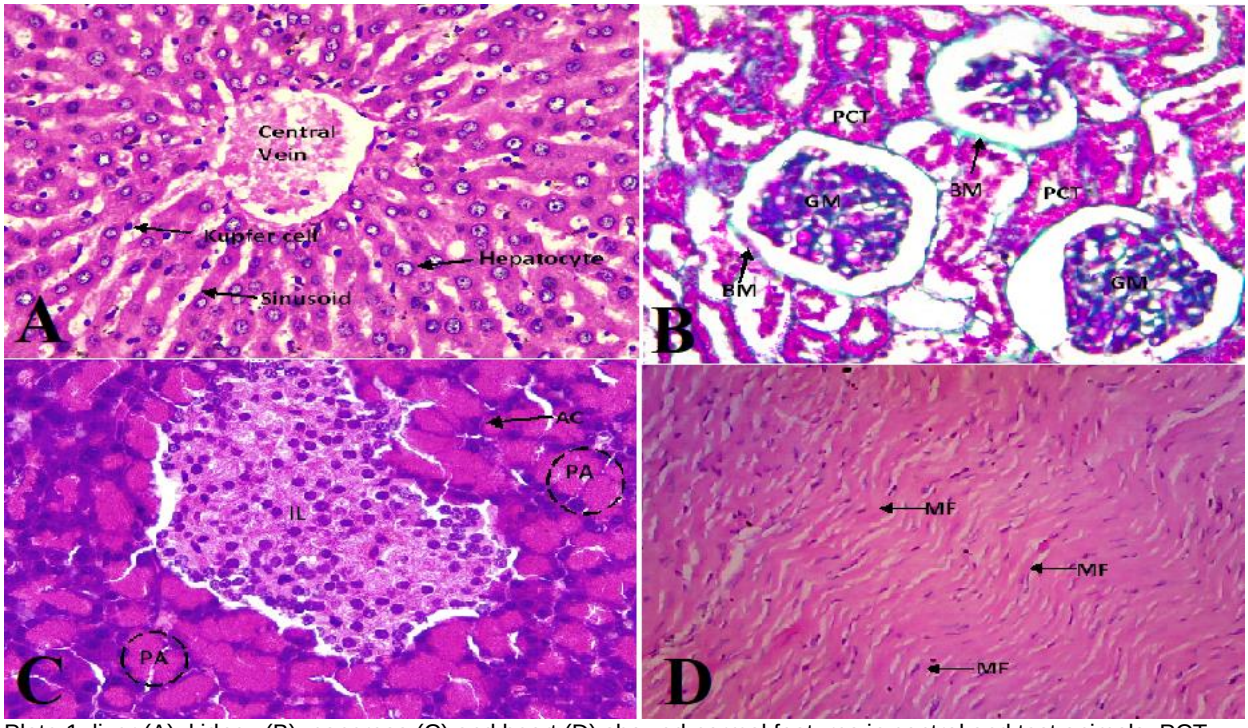

Plate 1: liver (A), kidney (B), pancreas (C) and heart (D) showed normal features in control and test animals. PCT = Proximal Convoluted tubule, BM = Bowman capsule, GM= Glomeruli, PA=, Pancreatic Acini, IL= Islet of Langerhans, AC= Acini cell, MF= myocardium fibre. Haematoxylin & Eosin stains at x400 Magnification

Lipid profile of rats were fully regulated after treatment with *P. dactylifera* extracts compared with results before treatment, midway to experimentation, and controls in this study. We suggest that *P. dactylifera* fruit extract exerts a long term effect rather than short as the overall result was laudable after 45 days treatment. This observation is comparable with reports on capability of *P. dactylifera* fruit to improve serum lipids in rat's model (Saafi *et al.* 2011; Arshad *et al.* 2015; Gunay *et al.* 2016). In a randomized controlled study, it was reported that serum HDLC increased while LDLC, triglycerides and total cholesterol significantly decreased after investigation (Mushtaq *et al.*, 2017). Reductions in serum LDLC and elevated HDLC values after treatment were reported to be due partly to free radical scavenging activities of *P. dactylifera* fruit (Chaira *et al.,* 2007). In another report, atherogenic index with LDLC / HDLC ratio in plasma has been found to be a good prognostic factor for cardiovascular risk in the presence of co-morbid situations as reported (Gunay *et al.,* 2016). Regulation of lipid profile by *P. dactylifera* extract may also be traced to some active components in the plant like flavonoids and polyphones (antioxidants), and has been confirmed to exist richly in *P. dactylifera* fruit as against other component parts of the plant (Arshad *et al.,* 2015). *P. dactylifera* fruit fiber content is relatively high and has been reported to captivate dietary fats which in-turn help in regulating lipid profiles (Juhaimi *et al.* 2012; Osman, 2014).

All parameters after treatment with high dose *C. esculentus* extract indicated a statistical significance ($p \leq 0.05$) compared with those obtained before treatment / control animals. This observation suggests that the extract performs better after a prolonged administration, and embraces the report of Hassan (2007), which suggests that *C. esculentus* extract has varying health benefits that are yet to be harnessed. It appeared that *C. esculentus* assists in inducing the good lipid (elevation of HDL-cholesterol levels) in animals via inhibition of biosynthesis of cholesterol concentration (Zommara and Imaizumi, 2017). Elevation of HDL-C / reduction in LDL-C during treatment with *C. esculentus* may have resulted from high amount of mono unsaturated fatty acids such as oleic acid content of the plant. This acid (oleic acid) is largely available in *C. esculentus* leading to increasing levels of HDL/ reduced LDL-C in rats primarily by delaying the clearance of HDL apo A-I from plasma compartment (Brousseau *et al.,* 1995). On the other hand, poly unsaturated fatty acids (linoleic acid), which is abundantly available in *C. esculentus* oil was found to decrease LDL-C and VLDL in rats and has been reported to signify an inhibitory effect on hepatic synthesis including secretion of triglyceride-rich VLDL in blood (Nenseter *et al.* 1992; Rustan *et al.,* 1993).

Effects of high dose multi-plant extract on lipid parameters revealed that all values were markedly controlled during investigation (Table 3) while further regulation of parameters were observed after treatment (Table 4) compared

with values before administration / controls and were statistically significant ($p \leq 0.05$). This report is strongly suggestive of an anti-hypercholesterolemia action, which means that the effect may be due to synergy between *P. dactylifera* / *C. esculentus* including individual actions of the plant. It was observed that the mix may possess therapeutic, regulatory, and anti-hyperlipidemia properties than individual extracts going by the quick healing exhibited in animals after sample collections from marginal ear lobe followed by treatment with multi-plant extract compared with control animals. But the mechanism of this act has not been well understood in the present work. Notwithstanding, the action buttresses earlier reports on multiple plant's extracts functioning effectively than individual components (Mahunnah *et al.* 2006; Otieno *et al.,* 2008). Thus justifying reports that combination of plants of similar actions has more curative potentials compared with its individual actions (Ogeyemhe *et al.,* 2019).

4. Conclusion

Multi-plant extracts regulate hypercholesterolemia in rats while *C. esculentus* appeared to be the major active anti-hypercholesterolemia component in the mix. The present combined extracts may just be a welcome development in the management of hypercholesterolemia.

Acknowledgements

Authors acknowledge the technical assistance provided by Mr. Anthony Igiebor; Department of Medical Laboratory Science, University of Benin, Nigeria and Prof. H.A. Akinnibosun of Plant Biology and Biotechnology, University of Benin, Nigeria for identification of the plants.

Conflict of interest

The authors declare no conflict of interest.

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