

Total Lipid Profile, Faecal Cholesterol, very Low Density Lipoprotein Cholesterol (VLDL-C), Atherogenic Index (A.I) and Percent Atherosclerosis with Aqueous Fruit Extract of *Solanum macrocarpum* in Chronic Triton-Induced Hyperlipidemic Albino Rats

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Abstract: Studies were undertaken to investigate the effect of the aqueous fruit extract of *Solanum macrocarpum* on the total lipid profile [total cholesterol, triglyceride, high density lipoprotein-cholesterol (HDL-C) and low density lipoprotein cholesterol (LDL-C)], faecal cholesterol, very low density lipoprotein-cholesterol (VLDL-C), atherogenic index (A.I) and percent atherosclerosis on chronic triton-induced hyperlipidemic rats. The increase in HDL-C was dose-dependent and statistically significant ($p < 0.05$) at both 24 and 72 h. There was no change ($p > 0.05$) with increase in extract dose for both total cholesterol and triglycerides throughout the period of study while the decrease in LDL-C was significant ($p < 0.05$) at 72h. The increase in faecal cholesterol with increase extract dose was significant ($p < 0.05$) at 48 and 72h of study. The VLDL-C and percent atherosclerosis reduced with increase in extract dose. The decrease in VLDL-C was only significant ($p < 0.05$) at 72h whilst that for percent atherosclerosis was significant ($p < 0.05$) at both 48 and 72h. There was no change in A.I. ($p > 0.05$). The results shows that the plant may be capable of reducing circulating lipids in chronic triton-induced hyperlipidemic rats probably by reducing absorption of lipids, thus, reducing hyperlipidemia. At the same time, the aqueous fruit extract probably has the potential to reduce the risk of development of heart diseases since VLDL-C has been shown to be beneficial and indicative of a lower risk of coronary heart diseases. Also, a reduction in percent atherosclerosis is desirable as this implies that atherosclerosis is reduced.

Key words: Atherogenic index, faecal-cholesterol, percent atherosclerosis, *Solanum macrocarpum*, total lipid profile, VLDL-cholesterol

INTRODUCTION

Extracts from the unripe fruits of *Solanum macrocarpum* (synonyms: *S. macrocarpon* L. sensu stricto and *S. daysphyllum* Schumacher and Thonn) (Grubben and Denton, 2004) called “Gorongu” in Kanuri have been used by the “Kanuris” of Nigeria and other West African countries (like Sierra Leone) and East African countries’ (like Kenya and Uganda) folk medicine (Grubben and Denton, 2004). In the traditional North East Arid zone of Nigeria, the unripe fruit of *S. macrocarpum* is known for its laxative, antihypertensive and hypolipidemic effects. The fruit and flowers are also used in cleaning the teeth (Bokhari and Ahmed, 1980; Grubben and Denton, 2004). Importantly, *S. macrocarpum* had been shown to display a wide spectrum of biological activities, with

experimental support for the empiric ethnopharmacological use of this plant in folk medicine (Sodipo *et al.*, 2008a, b, 2009a). Recently, we have demonstrated hepatoprotective effects of aqueous fruit extract of *S. macrocarpum* in diet-induced hypercholesterolemic rats (Sodipo *et al.*, 2009b) and acute triton-induced hyperlipidemic rats (Sodipo *et al.*, 2011c) However, the mechanism of hyperlipidemia had not been extensively studied. (Sodipo *et al.*, 2009c, 2011b) observed a favourable lipid profile in diet-induced hypercholesterolaemic and acute-triton induced hyperlipidemic rats administered with the aqueous fruit extract, the exact mechanism of cholesterol lowering and hypolipidemia was not known. Taking into account these data, we have conducted our research on total lipid profile, faecal cholesterol, very low density lipoprotein

cholesterol (VLDL-C), Atherogenic Index (A.I.) and percent atherosclerosis after oral administration of triton-X 100 (a non-ionic surfactant that interferes with uptake of lipids) for 90 days to induce chronic hyperlipidemia in rats in order to find out if the fruit of *S. macrocarpum* can indeed lower hyperlipidemia as an increase in faecal cholesterol corresponds to a decrease in cholesterol and lipid absorption (Moore *et al.*, 1968; Sodipo *et al.*, 2011b). Also, a plausible mechanism of the hypolipidaemic action of the plant, if there is any could probably be fashioned out. At the same time we will also try to find out if the extract has the potential to reduce the risk of development of coronary heart disease and atherosclerosis as low VLDL-C, low percent atherosclerosis and low atherogenic index have been shown to be beneficial and indicative of a lower risk of coronary heart diseases (Williamson *et al.*, 1996; Chander *et al.*, 2005).

It has been shown that intravenous injection of nonionic detergents such as triton WR-1339 (polymeric p-iso-octyl polyoxethylene phenol) in experimental animals, results in a progressive increase in the concentration of lipids in the blood (Otway and Robinson, 1967). The action is believed to be due at least in part, to the capacity of the detergent to associate with triglycerides in the plasma in such a way as to reduce their rate of hydrolysis by the enzyme, clearing factor lipase or lipoprotein lipase, and so to interfere with their uptake from the circulation by the extra-hepatic tissues (Robinson, 1963; Scanu, 1965). In the experiment that would be described, triton X-100 (polyoxethylene octyl phenyl ether) was administered orally to the rats and not parenterally like triton WR 1339 because the pilot study revealed that 400mg/kg of the triton-X administered intraperitoneally (i.p.) to 30 rats in the first day killed all of them, probably indicating high osmotic fragility and altered red blood cell (RBC) morphology, as to cause icterus, leading to the death of the rats. Oral administration of the triton however did not cause death in the rats (Sodipo, 2009; Sodipo *et al.*, 2011a, b, c).

MATERIALS AND METHODS

Plant collection and identification: The plant material (*Solanum macrocarpum* Linn.) used in this study was obtained from Alau in Konduga Local Government, Borno State, Nigeria, between October and November, 2007. The plant was identified and authenticated by Prof. S.S. Sanusi of the Department of Biological Sciences, University of Maiduguri, Maiduguri, Nigeria. Specimen voucher No. 548 was deposited at the Research Laboratory of the Department of Chemistry.

Extraction: The fruit of *S. macrocarpum* with the calyx removed was air dried and pulverized by grinding using

pestle and mortar. The 2.2 kg of the ground fruit was subjected to exhaustive Soxhlet-extraction in distilled water at 100°C to give the extract yield of 15.3% w/W (Mittal *et al.*, 1981; Fernando *et al.*, 1991; Lin *et al.*, 1999). The resultant solution was concentrated *in vacuo* and it was stored in a specimen bottle in a desiccator at room temperature until when required.

Animals: Thirty six (36) male albino rats of Wistar strain weighing 160-200g were used in this study. The animals were obtained from the Animal House Unit of the Department of Veterinary Physiology and Pharmacology and Biochemistry, Maiduguri. The animals were under standard laboratory condition in plastic cages. They were fed with commercial growers' mash feed (ECWA Feeds, Jos, Nigeria) and water was provided *ad libitum*. All the animals were handled according to the International Guiding Principles for Biomedical Research Involving Animals (CIOMS, 1985) as certified by the Animal Ethics Committee of the Faculty of Veterinary Medicine, University of Maiduguri.

Administration of triton and extract: Thirty (30) albino rats were made hyperlipidaemic by feeding them orally (p.o) for 90 days with normal feed diet and triton-X (Sigma Chemical Co. St. Louis, M.O. USA) at a dose of 400 mg/kg in saline suspension from the stock concentration of 535 g/mL. The rats were divided into 5 groups of 6 animals each. Another set of six (6) rats (Group 1) were fed with normal food and water only for 90 days. After 90 days, 24 of the rats were administered with graded doses of the fruit extract. Group I was the negative control and it was given distilled water only. Groups 3, 4, 5 and 6 were administered with geometrical doses (25, 50, 100 and 200 mg/kg, respectively) of the fruit extract intraperitoneally (i.p.) from a stock concentration of 200 mg/kg whilst group 2 (positive control) was not given the extract. After 24, 48 and 72 h, respectively of the effect of the extract on the hyperlipidemic rats, the total lipid profile, faecal cholesterol, VLDL-C, atherogenic index and percent atherosclerosis were determined (Williamson *et al.*, 1996). Before the rats were fed with triton-X, their weights were taken. The weights were subsequently taken after 30, 60 and 90 days, respectively of triton administration.

Determination of total lipid profile: Two rats from each of the groups were humanely sacrificed after 24, 48 and 72 h, respectively of the effect of the extract on chronic hyperlipidemic rats by cutting their throat with a sterile blade. Blood was collected into a clean, sterile, labelled centrifuge tubes without an anticoagulant and centrifuged at a rate of 12,000 revolutions per minute (rpm) for 10 min. The clear, yellow serum was then separated from

Table 1: Change in mean body weight of male albino rats after being administered orally with Triton-X (400 mg/kg) for 90 days

Group	Mean body weight ± S.D. (g)				% Increase in mean body weight
	Days of treatment				
	0	30	60	90	
One*	110.25±10.50 ^a	112.50±20.45 ^a	114.00±12.51 ^a	117.20±15.07 ^a	6.30±4.57 ^a
Two	100.20±26.64 ^a	135.80±41.26 ^b	203.44±52.97 ^b	214.20±58.61 ^b	113.78±32.37 ^b
Three	80.00±17.25 ^a	110.20±27.52	163.64±26.93 ^b	174.20±15.06 ^b	117.75±2.19 ^b
Four	99.40±29.19 ^a	131.40±41.58 ^b	184.80±37.58 ^b	216.80±41.05	^b 117.30±11.86 ^b
Five	116.60±42.58 ^a	129.00±11.92 ^b	172.78±17.03 ^b	194.80±19.74 ^b	67.07±22.84 ^b
Six	95.00±20.96 ^a	120.40±36.65 ^b	192.18±34.03 ^b	211.95±33.74 ^b	122.11±12.78 ^b

Within rows, means with different superscripts are statistically significant (p<0.05) when compared to day zero (0) using one way analysis of variance (ANOVA); 0 day: before triton-X administration; n: 6 rats; Group One*: Rats fed with normal diet and had free access to water throughout the 90 days but were not administered triton-X

Table 2: Effect of the aqueous fruit extract of *S. macrocarpum* on total lipid profile of hyperlipidaemic rats administered orally with triton-X for 90 days

Hours after extract administration	Group	Extract dose (mg/kg)	Total Cholesterol (mmol/L)	Triglycerides (mmol/L)	HDL-C (mmol/L)	LDL-C (mmol/L)
				Mean ± S.D.		
24	One	-ve control	1.70±0.28 ^a	1.35±0.07 ^a	0.35±0.07 ^a	0.60±0.14 ^a
	Two	+ve control	2.40±0.29 ^a	1.90±0.42 ^a	0.40±0.14 ^b	0.80±0.14 ^a
	Three	25.00	2.15±0.64 ^a	1.45±0.35 ^a	0.50±0.00 ^b	0.75±0.35 ^a
	Four	50.00	2.10±0.57 ^a	1.30±0.28 ^a	0.75±0.07 ^b	0.65±0.50 ^a
	Five	100.00	1.35±0.07 ^a	1.25±0.21 ^a	0.85±0.71 ^b	0.50±0.14 ^a
	Six	200.00	1.15±0.07 ^a	0.70±0.14 ^a	0.95±0.07 ^b	0.40±0.14 ^a
48	One	-ve control	1.70±0.14 ^a	1.25±0.07 ^a	0.45±0.07 ^a	0.45±0.50 ^a
	Two	+ve control	2.55±0.07 ^a	1.60±0.28 ^a	0.55±0.07 ^a	1.10±0.42 ^a
	Three	25.00	2.10±1.13 ^a	1.45±0.21 ^a	0.50±0.14 ^a	0.60±0.14 ^a
	Four	50.00	1.50±0.14 ^a	1.15±0.07 ^a	0.55±0.07 ^a	0.50±0.25 ^a
	Five	100.00	1.45±0.07 ^a	1.10±0.57 ^a	0.65±0.35 ^a	0.45±0.50 ^a
	Six	200.00	1.25±0.35 ^a	1.05±0.78 ^a	0.95±0.21 ^a	0.25±0.07 ^a
72	One	-ve control	1.90±0.14 ^a	1.35±0.07 ^a	0.60±0.14 ^a	0.80±0.14 ^a
	Two	+ve control	2.40±0.50 ^a	1.45±0.14 ^a	0.70±0.14 ^b	1.35±0.71 ^b
	Three	25.00	2.20±0.28 ^a	1.40±0.14 ^a	0.65±0.21 ^b	1.20±0.28 ^b
	Four	50.00	2.10±0.42 ^a	1.30±0.14 ^a	0.80±0.14 ^b	0.85±0.21 ^b
	Five	100.00	1.70±0.14 ^a	1.20±0.14 ^a	1.05±0.21 ^b	0.65±0.21 ^b
	Six	200.00	1.35±0.07 ^a	0.90±0.00 ^a	1.20±0.28 ^b	0.45±0.21 ^b

Within columns, means with different superscripts are statistically significant (p<0.05) when compared to Group I (-ve control); -ve control: Rats fed with normal feed diet and had free access to water; +ve control: Rats fed with normal feed diet and given triton-X

settled cellular elements and subjected to determination of total lipid profile. The total lipid profile estimated from the serum was total cholesterol, triglycerides, high density lipoprotein-cholesterol (HDL-C) and low density lipoprotein-cholesterol (LDL-C). Total cholesterol was assayed by Tindar's reaction (Evans and Stein, 1986; NIH, 1990) using commercial kits from Fortress Diagnostics Ltd; Antrim. Serum triglycerides level was determined as described by Fossati and Prencipe (1982) and Cole *et al.* (1997) using commercial kit (Human cholesterol using Liquicolour Test Kit, Germany) (Grove, 1970). The serum LDC-L level was calculated by Friedwald's formular (Friedwald *et al.*, 1972; Hardman and Limbird, 2001; Sood, 2006). The calculation for serum LDL-C is given by:

$$\text{Serum LDL-C} = \text{Total cholesterol} - \text{Triglycerides} \div 2.2 - \text{HDL} - \text{C.}$$

Determination of faecal cholesterol: Faeces from two rats in each of the groups were collected after 24, 48 and

72 h, respectively that the extract had acted on the hyperlipidaemic rats into clean, cellophane bags and deep frozen. They were homogenized and extracted with chloroform/methanol in the ratio 2:1 (Folch *et al.*, 1957) for analysis of faecal cholesterol. The faecal cholesterol determination was like that of the serum total cholesterol (Tindar's reaction) (Evans and Stein, 1986; NIH, 1990) using commercial kits from Fortress Diagnostic Ltd; Antrim.

Calculation of very low density lipoprotein cholesterol (VLDL-C) (mmol/L): VLDL-C was calculated using the formula (Henry, 1991; Igweh *et al.*, 2005; Sathesh and Paris, 2008):

$$\text{VLDL-C (mmol/L)} = \text{Triglycerides} / 2.2$$

Calculation of atherogenic index (A.I.): Although index was calculated with the formula given below (Williamson *et al.*, 1996) as:

$$\text{A.I.} = \text{VLDL-C} + \text{LDL-C} / \text{HDL-C}$$

Table 3: Effect of the aqueous fruit extract of *S. macrocarpum* on faecal cholesterol of hyperlipidaemic rats administered orally with triton-X for 90 days

Hours after extract administration	Group	Extract dose (mg/kg)	Faecal cholesterol
			(mmol/L) Mean ± S.D.
24	One	-ve control	0.40±0.14 ^a
	Two	+ve control	0.20±0.14 ^a
	Three	25.00	0.25±0.07 ^a
	Four	50.00	0.30±0.14 ^a
	Five	100.00	0.35±0.21 ^a
	Six	200.00	0.40±0.14 ^a
48	One	-ve control	0.40±0.13 ^a
	Two	+ve control	0.15±0.07 ^b
	Three	25.00	0.30±0.11 ^b
	Four	50.00	0.35±0.00 ^b
	Five	100.00	0.40±0.00 ^b
	Six	200.00	0.45±0.07 ^b
72	One	-ve control	0.50±0.00 ^a
	Two	+ve control	0.20±0.14 ^b
	Three	25.00	0.25±0.07 ^b
	Four	50.00	0.30±0.07 ^b
	Five	100.00	0.40±0.14 ^b
	Six	200.00	0.45±0.07 ^b

Within columns, means with different superscripts are statistically significant ($p < 0.05$) when compared to Group I (-ve control); -ve control: Rats fed with normal feed diet and had free access to water; +ve control: Rats fed with normal feed diet and given triton-X

Where,

VLDL-C is very low density lipoprotein cholesterol

LDL-C is low density lipoprotein cholesterol

HDL-C is high density lipoprotein cholesterol

Determination of atherosclerosis in rats (%): The atherosclerotic rats (treated with triton-X for 3 months) were sacrificed and subjected to pathological examination. Atheromatous lesions were visible and were measured. The area covered is known to be directly related to the circulating lipid levels. Aortas of rats were removed and fat and connective tissue were cleaned off for examination for pathological changes. Atheromatous plaques were measured and the area covered was expressed as a percentage of the aorta as follows (Williamson *et al.*, 1996):

$$\text{Atherosclerosis (\%)} = (\text{Length of atheromatous plaque in aorta} / \text{Total length of aorta}) \times 100$$

Statistical analysis: Data were expressed as the means±S.D. The results obtained were subjected to Analysis of Variance (ANOVA) using Graph Pad Software (1998).

RESULTS

Change in mean body weight of male albino rats (Wistar strain) after being administered orally with triton-X for 90 days: The effect of triton-X on mean body weight of albino rats fed orally with triton-X is shown in Table 1. The increase in body weight observed in the rats was statistically significant ($p < 0.05$) when compared to day zero in all the groups except in Group one. Group one was not administered with triton-X

throughout the period of the study. Also, there was a significant percentage weight gain ($p < 0.05$) in the hyperlipidemic rats (Groups two-six) when compared with Group one which received standard diet and water *ad libitum*.

Effect of the aqueous fruit extract of *Solanum macrocarpum* on total lipid profile of hyperlipidemic rats administered triton-X orally for 90 days: The effect of the aqueous fruit extract of *Solanum macrocarpum* on total lipid profile of hyperlipidemic rats are shown in Table 2. The increase in HDL-C was dose-dependent, and statistically significant ($p < 0.05$) at both 24 and 72 h. There was no change ($p > 0.05$) with increase in extract dose for both total cholesterol and triglycerides throughout the period of study whilst the decrease in LDL-C was significant ($p < 0.05$) at 72 h. Throughout the period of study, the lowest value of total cholesterol, triglycerides and LDL-C were recorded at 200 mg/kg extract dose. At 24 h of study, the values for the total cholesterol, triglycerides and LDL-C are (1.15±0.07), (0.07±0.14) and (0.04±0.14) mmol/L, respectively. Also, the highest increase in HDL-C was recorded at 200 mg/kg extract dose and at 24 h of study which was (0.95±0.07) mmol/L.

Effect of the aqueous fruit extract of *Solanum macrocarpum* on faecal cholesterol of hyperlipidemic rats administered triton-X orally for 90 days: The effect of the aqueous fruit extract of *Solanum macrocarpum* on faecal cholesterol of hyperlipidemic rats is shown in Table 3. Faecal cholesterol significantly increased ($p < 0.05$) in the hyperlipidemic rats with increasing dose of extract at 48 and 72 h of study. The values of faecal cholesterol for the positive control (Group two) throughout the period of the work remained lower than that of the negative control (Group one). At 24 h of study, the faecal cholesterol for Group two was (0.20±0.14) mmol/L whilst that for Group one was (0.40±0.14) mmol/L.

Effect of the aqueous fruit extract of *Solanum macrocarpum* on VLDL-C, atherogenic index (A.I.) and percent atherosclerosis of hyperlipidemic rats administered triton-X orally for 90 days: The effect of the aqueous fruit extract of *Solanum macrocarpum* on VLDL-C, atherogenic index (A.I.) and percent atherosclerosis are shown in Table 4. The VLDL-C and percent atherosclerosis reduced with increase in extract dose. The decrease in VLDL-C was only significant ($p < 0.05$) at 72 h whilst that for percent atherosclerosis was significant ($p < 0.05$) at both 48 and 72 h. There was no change in A.I. ($p > 0.05$).

DISCUSSION

The increase in mean body weight of the rats after triton-X administration for 90 days was significant

Table 4: Effect of the aqueous fruit extract of *S. macrocarpum* on VLDL-C, atherogenic index (A.I.) and atherosclerosis (%) of hyperlipidaemic rats administered orally with triton-X for 90 days

Hours after extract administration	Group	Extract dose (mg/kg)	VLDL-C (mmol/L)	Atherogenic index (A.I.)(no unit) Mean±S.D	Atherosclerosis (%)
24	One	-ve control	0.65±0.14 ^a	1.90±0.14 ^a	44.35±12.74 ^a
	Two	+ve control	0.95±0.21 ^a	2.69±0.48 ^a	64.25±1.06 ^a
	Three	25.00	0.73±0.11 ^a	2.05±0.42 ^a	59.36±5.31 ^a
	Four	50.00	0.65±0.14 ^a	1.90±0.14 ^a	56.55±3.18 ^a
	Five	100.00	0.63±0.11 ^a	1.71±0.56 ^a	48.52±9.92 ^a
	Six	200.00	0.35±0.07 ^a	1.65±0.50 ^a	40.35±9.93 ^a
48	One	-ve control	0.63±0.04 ^a	1.80±0.42 ^a	29.54±1.37 ^a
	Two	+ve control	0.80±0.14 ^a	3.71±2.30 ^a	63.50±6.61 ^b
	Three	25.00	0.73±0.11 ^a	2.55±0.64 ^a	52.75±3.89 ^b
	Four	50.00	0.58±0.04 ^a	2.42±0.12 ^a	39.23±1.09 ^b
	Five	100.00	0.55±0.04 ^a	2.34±1.29 ^a	29.60±1.57 ^b
	Six	200.00	0.38±0.18 ^a	1.80±0.42 ^a	13.92±0.83 ^b
72	One	-ve control	0.68±0.04 ^a	1.45±0.29 ^a	37.51±1.35 ^a
	Two	+ve control	0.73±0.11 ^b	3.80±1.81 ^a	51.38±1.95 ^b
	Three	25.00	0.70±0.07 ^b	2.16±0.14 ^a	46.09±0.83 ^b
	Four	50.00	0.63±0.11 ^b	1.77±0.21 ^a	37.36±1.50 ^b
	Five	100.00	0.55±0.71 ^b	1.70±1.49 ^a	32.67±1.53 ^b
	Six	200.00	0.45±0.00 ^b	1.39±0.37 ^a	9.25±1.34 ^b

Within columns, means with different superscripts are statistically significant (p<0.05) when compared to Group I (-ve control); -ve control: Rats fed with normal feed diet and had free access to water; +ve control: Rats fed with normal feed diet and given triton-X

(p<0.05) (Groups two to six). Table 1, whilst Group one fed with normal diet was not significant (p>0.05). The percentage weight gain in the hyperlipidemic rats (Groups 2 to 6) was significantly high (p<0.05) when compared to Group one. Excessive weight gain (obesity) has been implicated in hypertension and ischaemic heart disease (Nwanjo *et al.*, 2006). It probably suggests that the triton-X had induced atherosclerosis as atherosclerosis takes three to six months to be induced in rats (Williamson *et al.*, 1996).

The result from the administration of graded doses of the aqueous fruit extract of *S. macrocarpum* on triton-X induced hyperlipidemic rats revealed a significant reduction (p<0.05) in LDL-C at 72 h and significant elevation (p<0.05) in HDL-C at 24 and 72 h (Table 2). There was no reduction in the total cholesterol and triglycerides (p>0.05) throughout the period of study. Clinical studies in humans have shown that lowering levels of serum cholesterol (especially LDL-C) with diet or drugs decreases the incidence of coronary heart disease (Gotto *et al.*, 1990). The results obtained from serum lipid profile in this experiment agree with the findings of Nkanu *et al.* (2003), Chander *et al.* (2005) and Nwanjo *et al.* (2006). Also, increased levels of cholesterol are associated with Coronary Heart Disease (CHD), hyperlipoproteinemia, diabetes mellitus, cirrhosis and various liver diseases (Mukherjee, 1988; Odutola, 1992; Odetola *et al.*, 2004; Iweala and Okeke, 2005; Mshelia and Gahsua, 2007). In the present study, the cholesterol levels decreased (LDL-C). The implication of this is that the use of the aqueous fruit extract of *Solanum macrocarpum* will probably ameliorate the occurrence of coronary heart disease by lowering LDL-C level. Also, the phytochemistry revealed the fruit of *S. macrocarpum* to contain alkaloids, flavonoids and saponins (Sodipo

et al., 2008a) whose biological activities include among others, hypolipidemia and hypcholesterolemia (Cheeke, 1971; Mahato *et al.*, 1982; Satheesh and Paris, 2008; Chander *et al.*, 2005). *Solanum* species also contain plant steroids known as steroidal alkaloids and reports have shown that the *Solanum* alkaloids are said to be responsible for lowering hyperlipidemia (ANON, 2007). The result in this study tallies with that of Chander *et al.* (2005) who showed that administration of Indian black tea to Triton WR 1339 (polymeric p-iso-octyl polyoxyethylene phenol) induced hyperlipidemic rats caused a decrease in the plasma levels of cholesterol (LDL-C). So also the rise in serum, LDL-C and decrease in HDL-C in Triton-X induced hyperlipidemic rats is in conformity with the rise in plasma cholesterol levels observed in Triton induced hyperlipidemic rats carried out by Schurr *et al.* (1972). It has been shown that intravenous injection of nonionic detergents such as Triton WR 1339 in experimental animals, results in a progressive increase in the concentration of lipids in the blood (Kellner *et al.*, 1951; Friedman and Bryers, 1953; Otway and Robinson, 1967). The action is believed to be due at least, in part, to the capacity of the detergents to associate with triglycerides in the plasma in such a way as to reduce their rate of hydrolysis by the enzyme, clearing factor lipase or lipoprotein lipase, and so to interfere with their uptake from the circulation by the extra-hepatic tissues (Robinson, 1963; Scanu, 1965).

In the present study, the decrease in LDL-C was significant (p<0.05) at 72h of study. LDL-Cs are derived from the metabolism of VLDL-C and they have a very low half life (t_{1/2}), 3 to 4 days (Hardman and Limbird, 2001). The result buttressed the fact that the aqueous fruit extract of *Solanum macrocarpum* could probably lower the chronic hyperlipidemia induced in the rats. Flavonoids

present in the fruit (Sodipo *et al.*, 2008a) prevent the oxidation of the LDL-C which is atherogenic (Chander *et al.*, 2005; Khan, 2008).

Smaller LDL particles (LDL-III) are considered more atherogenic than larger more buoyant species because of their increased susceptibility to oxidation (Dejagar *et al.*, 1993; Igweh *et al.*, 2005) and their increased residence time in plasma (Rainwater, 2000; Igweh *et al.*, 2005). Plasma triglyceride concentration also has a determinative influence on the concentration of small dense LDL particles in normal population (Dejagar *et al.*, 1993; Mendelsohn and Kars, 1999; Igweh *et al.*, 2005). It has also been estimated that for any 0.026 mmol/mL (1 mL/dL) increase in HDL-C there is a 3% decrease in the risk of mortality from cardiovascular disease (Okonofua *et al.*, 1990; Igweh *et al.*, 2005).

The increase in faecal cholesterol with increase in extract dose was significant at 48 and 72h ($p < 0.05$) of study. This implies that with increase in extract dose, cholesterol excretion in the faeces increased. This agrees with the work of Moore *et al.* (1968) who demonstrated that the hypocholesterolaemic action of dietary unsaturated fatty acids in man is associated with an increase in the faecal loss of bile acids and neutral sterols. Saponins as found in this plant (Sodipo *et al.*, 2008a) are known to increase bile acid and other sterol production (Mahato *et al.*, 1982; Mac Donald *et al.*, 2005) which are subsequently excreted. This is in conformity with the hypothesis that increased faecal excretion of cholesterol corresponds to a decreased absorption (Moore *et al.*, 1968; Sodipo *et al.*, 2011b). Literature has shown that green tea leaves (*Camellia sinensis*) lowered cholesterol and inhibited lipid absorption and also decreased serum alanine amino transferase (ALT) activity in ovariectomised rats and obese mice respectively (Wang *et al.*, 2006; Bruno *et al.*, 2008). It therefore follows that since the aqueous fruit extract of *S. macrocarpum* increased faecal cholesterol excretion in chronic hyperlipidemic rats, the absorption probably decreased and this might explain the hypolipidaemic and hypocholesterolemic effects of the extract as claimed in traditional medicine. Also, the increase in faecal cholesterol excretion might be due to its decreased absorption which may be due to the inhibition of pancreatic lipolytic enzymes such as lipase and phospholipase A₂ as demonstrated in green tea leaves (Bruno *et al.*, 2008), leading to an increase in β -oxidation in mice fed high-fat diet, may also be plausible mechanism in non-accumulation of cholesterol and hence increased excretion of fat in the faeces in chronic hyperlipidemic rats administered aqueous fruit extract of *S. macrocarpum*. The increase in faecal excretion of cholesterol was significant at 48 and 72 h of study, probably implying that the maximal hypolipidemic effect was felt at 72 h of extract administration. This may be compared with the pattern observed in the acute triton-

induced hyperlipidemic rats in which the maximal faecal excretion was at 48 h (Sodipo *et al.*, 2011a). The increased faecal cholesterol excretion as demonstrated by the aqueous fruit extract of the plant may be responsible for its hypolipidemic effects. The human bile consists of primary bile acid (31% cholic acid and 45% chenodeoxycholic acid in the liver) and 24% secondary bile (which is made up of deoxycholic acid and lithocholic acid) produced from the action of intestinal bacteria on primary bile acids. In primates and other animals such as the rat, cholic acid is dominant (Mead *et al.*, 1986).

The hyperlipidemia induced by triton-X in rats was reduced by graded doses of aqueous fruit extract of *S. macrocarpum* as shown in the statistical reduction ($p < 0.05$) in VLDL-C at 72 h and percent atherosclerosis at 48 and 72 h (Table 4). The values for the positive control (Group two) i.e. hyperlipidemic rats not treated with extract were high; these are atherogenic and undesirable. These values reduced in groups of rats treated with varying doses of aqueous extract of the fruit of *S. macrocarpum*. This shows that the aqueous extract has the potential to reduce the risk of development of heart diseases since low VLDL-C and low % atherosclerosis have been shown to be beneficial and indicative of a lower risk of coronary heart diseases (Williamson *et al.*, 1996; Chander *et al.*, 2005). In the liver, activation of peroxisome proliferator activated receptor α -isoform (PPAR α) is predominantly involved in fatty acid and lipid catabolism. It is also involved in the import and activation of genes involved in fatty acid oxidation in the liver, heart, kidney and skeletal muscles (Gilde and Van Bilsen, 2003; Satheesh and Paris, 2008). In the liver, activation of PPAR α leads to increased β -oxidation of fatty acids and decreased triglyceride and VLDL synthesis (Fruchart and Duriez, 2004; Satheesh and Paris, 2008).

A reduction [Length Of Plaque in aorta/ Total length of aorta]×100

is desirable as this implies that atherosclerosis is reduced (Williamson *et al.*, 1996). Chander *et al.* (2005) have shown that oxidized LDL is atherogenic, thus a reduction in LDL is anti-atherogenic as found in Indian black tea, Tata Tea Co. Ltd., India on Triton WR 1339 induced hyperlipidemia in rats. Oxidation of LDL could have been prevented by the flavonoids present in the plants as flavonoids are antioxidants (Khan, 2008).

CONCLUSION

The present study shows that the aqueous fruit extract of *Solanum macrocarpum* may be capable of reducing lipids in chronic triton-induced hyperlipidemic rats probably by reducing absorption of lipids and increasing

faecal cholesterol excretion, thus reducing hyperlipidemia, thus, buttressing the claim in traditional medicine that the unripe fruit lowers hyperlipidemia. In addition, the extract probably has the potential to reduce the risk of development of coronary heart disease and atherosclerosis.

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