

Endocrine Function with Aqueous Fruit Extract of *Solanum macrocarpum* Linn. In Albino Rats Chronically Administered Triton-X to Induce Hyperlipidemia

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ABSTRACT

Studies were conducted on the effect of the aqueous fruit extract of *Solanum macrocarpum* Linn. in chronic triton-induced hyperlipidemic rats. The plant was Soxhlet-extracted with distilled water and the extract concentrated *in vacuo* with a yield of 15.34 % ^{w/w}. The extract was stored in a specimen bottle in a desiccator at room temperature until when required. The endocrine function parameters: thyroid hormones (thyroxine ie. T₄, 3, 5, 3' triiodothyronine i.e. T₃ and thyroid stimulating hormone ie. TSH), testosterone, 17β – oestradiol and insulin were determined. The results showed that with increase in extract dose there was a significant (p<0.05) increase in TSH and T₄ at 72 hrs whilst there was no change for T₃ (p>0.05) throughout the period of study. Oestradiol levels increased significantly (p<0.05) at 24 hrs and 72 hrs when compared to the negative control (Group one), but the levels of the oestradiol level in the negative control was however higher than that of the rats not administered the extract (positive control or Group two). The positive control had an oestradiol level of (15.00±4.24) pg/ml, (12.00±1.49) pg/ml and (13.50±2.54) pg/ml at 24 hrs, 48hrs and 72 hrs respectively whilst the values of the oestradiol in the negative control rats (group one) are (36.50±2.12) pg/ml, (20.00±4.24) pg/mol and (24.50±2.12) pg/mol at 24 hrs, 48 hrs and 72 hrs respectively. The increase in the testosterone level with increase in extract dose was significant (p<0.05) at 48 hrs whilst the insulin levels increased significantly (p<0.05) at 48 hrs and 72 hrs when compared to the negative control. The aqueous fruit extract of *S. macrocarpum* by increasing the level of the thyroid hormones, testosterone, 17β oestradiol and insulin in hyperlipidemic rats in this study probably imply a lipid lowering effect.

Keywords: *Solanum macrocarpum* Linn, aqueous extract, triton-X endocrine function, chronic hyperlipidemic rats.

INTRODUCTION

The control of metabolism, growth and reproduction is mediated by a combination of neural and endocrine systems located in the hypothalamus and pituitary gland (Katzung, 2004). Some of the endocrine hormones are thyroid hormones (thyroxine ie. T₄, triiodothyronine i.e. T₃ and thyrotrophin ie. TSH), reproductive hormones (testosterone and oestrogens) and the pancreatic hormone, insulin (Anthony, 2003).

The thyroid gland consists of two types of cells, the follicular and parafollicular cells (Gali, 2007). The follicular cells are responsible for the production of thyroxine also called 3, 5, 3', 5' tetraiodothyronine or T₄ and 3, 5, 3' triiodothyronine or T₃). T₄ can be considered a pro-hormone and T₃ is produced by extrathyroidal deiodination of T₄ by the Type 1 deiodinase enzyme. T₄ is biologically inactive and must be converted to T₃ before biological activity is exerted (Oppenheimer, 1972). Thus, it is not surprising that the thyroid produces 100% of circulating thyroxine (T₄) but only about (5–10%) of circulating T₃ (Gali, 2007).

Thyroid hormones are indispensable for growth, mental development and sexual maturation in mammals. Conversely, excess thyroid secretion leads to body wasting, nervousness, tachycardia, tremor and excess heat production. Thyroid hormone absence leads to mental and physical slowing, poor resistance to cold and in children, mental retardation and dwarfism (Braverman, 1996; Gali, 2007). Thyroid hormone is required for the normal functions of nearly all tissues, with major effect on oxygen consumption and metabolic rate (Oppenheimer *et al.*, 1987). These hormones also stimulate protein, carbohydrate and lipid metabolism (Gali, 2007).

Thyroid function is controlled by the thyroid stimulating hormone (TSH) or thyrotrophin of the anterior pituitary (Jackson, 1982; Gali, 2007). Thyroid hormones increase the expression of LDL receptors on the hepatocytes and increase the activity of lipid lowering liver enzymes, resulting in a reduction in LDL (Ness and Lopez, 1995; Ness *et al.*, 1998; Shin and Timothy, 2003; Gali, 2007). Thyroid hormones also increase the expression of apolipoprotein A₁, a major component of HDL (Taylor *et al.*, 1996; Gali, 2007). T₃ also regulates post-transcriptional editing of apolipoprotein mRNA (Davidson *et al.*, 1998). In addition, T₃ is important in hepatic degradation of cholesterol into bile acids by increasing the transcription of the rate-limiting enzyme in the process, the cholesterol – 7 α – hydroxylase (Ness *et al.*, 1990; Pandak *et al.*, 1997; Gali, 2007). The above effect of the thyroid hormones could be beneficial in reducing the onset of atherosclerosis, if they were elicited without deleterious effects, particularly the cardiac effect (Dillman, 1990; Woeber, 1992; Gali, 2007).

Oestrogens are C₁₈ steroids with aromatic configuration in the A ring. 17 β oestradiol, oestrone and oestradiol are the three naturally occurring oestrogens found in the matured female body (Katzung, 2004; Aliu, 2007). Oestradiol is the most potent naturally occurring oestrogen. Besides human ovary, oestrogens are also secreted by the placenta and to a lesser extent by the adrenal cortex and the testis (Katzung, 2004). Oestradiol levels have been shown to be decreased in patients with myocardial infarction (Odutola, 1992; Williamson *et al.*, 1996). Oestrogens have also been shown to decrease low density lipoprotein cholesterol (LDL-C) by 5 – 10%. This lipid effect accounts for 25 – 40 % in the reduction of coronary heart disease after oestrogen therapy (ANON, 2008). Oestrogens also give a favourable lipid profile i.e. increase high density lipoprotein cholesterol (HDL-C), decrease low density lipoprotein cholesterol (LDL-C) and reduce in total cholesterol (Katzung, 2004) which in turn may contribute to retardation of atherosclerotic cardiovascular disease.

Insulin is produced by the β -cells of the islet of Langerhans in the pancreas. Absolute or relative lack of insulin leads to diabetes mellitus (Katzung, 2004). Diabetes mellitus is a group of metabolic and endocrine disorders characterized by chronic hyperglycaemia and eventual glycosuria. It is caused by the inability of tissues to carry out normal metabolism of carbohydrates, fats and protein. Insulin is a small protein with a molecular weight in humans of 5,808. It contains 51 amino acids arranged in 2 chains (A and B) and linked by disulphide bridges. There are species differences in the amino acids of both chains (Katzung, 2004). In diabetes mellitus, insufficiency or deficiency of insulin leads to decreased synthesis of protein, lipids and glycogen. Increased mobilization of fat from adipose tissue leads to increased levels of cholesterol, lipids, free fatty acids and triglycerides i.e. hyperlipidaemia (Katzung, 2004; Aliu, 2007). Also there is decreased high density lipoprotein cholesterol (HDL-C) in diabetes mellitus patients (Cabezas *et al.*, 1993; Syvanne and Taskinen, 1997; Sodipo, 2009). At this same time, there seems to be few studies relating diabetes to hyperlipidemia (Sjoholm, 1998a; Sodipo, 2009).

Testosterone is the most important androgen secreted by the testis. It is synthesized from cholesterol in the Leydig cells (95%) and only 5% by the adrenal cortex. In men, approximately 8 mg testosterone is produced daily (Katzung, 2004). Plasma levels of testosterone in males daily drops to approximately 0.6 μ g/dl after 50 years. Testosterone is also present in the plasma of women and is approximately 0.03 μ g/dl (i.e. about 1/20 times less) and is derived from the ovaries, the adrenal cortex and the peripheral conversion of other hormones (Katzung, 2004). It is reduced in males with myocardial infarction (Williamson *et al.*, 1996; Katzung, 2004). The active form of the enzyme is dihydrotestosterone and this is achieved using the enzyme 5 α -reductase (Katzung 2004; Sodipo, 2009). Also testosterone has been found at lower levels with a high fat diet (Williamson *et al.*, 1996. Sodipo, 2009) and this is of note because high fat diet may eventually lead to hyperlipidaemia. In other words, it may be possible for hyperlipidemia to lower testosterone level which in turn may probably facilitate myocardial infarction.

In the traditional North East Arid zone of Nigeria, the unripe fruit of *S. macrocarpum* (synonyms: *S. macrocarpum* L. sensu stricto and *S. daysphyllum* Schumacher and Thonn) [Grubben and Denton, 2004] called “Gorongong” in Kanuri is known for its laxative, antihypertensive and hypolipidaemic effects. 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; 2012a).

Recently, we have demonstrated hepatoprotective effects of aqueous fruit extract of *S. macrocarpum* in diet-induced hypercholesterolaemic rats (Sodipo *et al.*, 2009b), acute and chronic triton-induced hyperlipidemic rats respectively (Sodipo *et al.*, 2011b, 2012a). However, the mechanism of hypolipidemia had not been extensively studied. Sodipo *et al.*, (2009c; 2011a, 2012a) observed a favourable lipid profile in diet-induced hypercholesterolaemic acute and chronic-triton-induced hyperlipidaemic 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 endocrine function tests (T₃, T₄, TSH, 17 β -oestradiol, testosterone, insulin) after oral administration of triton-X 100 (a non-ionic surfactant that

interferes with uptake of lipids) for 90 days to induce chronic hyperlipidaemia/ atherosclerosis in rats in order to find out if the fruit of *S. macrocarpum* can indeed lower hyperlipidaemia, as an increase in endocrine hormones corresponds to a decrease in LDL – C and increase in HDL-C (Odutola 1992; Williamson *et al.*, 1996; Katzung, 2004; Gali, 2007; ANON, 2008; Sodipo, 2009). Also a plausible mechanism of the hypolipideamic 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 development of coronary heart disease and atherosclerosis as increase in thyroid hormones, oestrogen, testosterone and insulin have been shown to be beneficial and indicative of a lower risk of coronary heart diseases (Odutola, 1992; Wiliamson *et al.*, 1996; Katzung, 2004; Gali, 2007; ANON 2008; Sodipo, 2009).

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 and kept in a desiccators 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, Pharmacology and Biochemistry, University of Maiduguri. They were housed under standard laboratory condition in plastic cages. They were fed 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 (Approved on October 15th 2008 at its 12th Ethical Committee meeting).

Administration of triton and extract

Thirty (30) albino rats were made hyperlipidemic 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 thirty six (36) rats for the experiment were divided into 6 groups of 6 animals each. After ninety (90) days, 24 of the 30 hyperlipidemic rats were administered with graded doses of the fruit extract. Group I was the negative control and it was given normal feed and distilled water only. Group two was the positive control and it was given normal feed and triton-X with distilled water only and normal feed. Groups 3, 4, 5, and 6 were administered with geometrical doses (25, 50, 100 and 200 mg/kg) of the fruit extract intraperitoneally (i.p.) from a stock concentration of 200 mg/ml. After 24, 48 and 72 hrs, respectively of the effect of the extract on the hyperlipidemic rats, the endocrine function tests were determined. Before the rats were fed with triton –X, their weights were taken. The weights were subsequently taken after 30, 60 and 90 days of triton administration (adapted from Williamson, *et al.*, 1996).

Endocrine function tests

Two rats from each of the groups were humanely sacrificed after 24 hrs, 48 hrs and 72 hrs respectively of the effect of the extract on chronic hyperlipidaemic rats by cutting their throat with a sterile blade. Blood was collected into clean, sterile labelled centrifuge tubes without an anticoagulant and centrifuged at a rate of 12,000 revolutions per minute (rpm) for 10 minutes. The clear, yellow serum was then separated from settled cellular elements and subjected to determination of endocrine function tests.

The endocrine function tests estimated from the serum were thyroid hormones (which included 3, 5, 3' triiodothyronine or T₃, 3, 5, 3', 5' tetraiodothyronine or T₄, thyroid stimulating hormone, TSH) reproductive hormones (which include testosterone and 17β-oestradiol) and insulin.

T₃ was determined by the Enzyme Link Immunosorbent Assay (Microwell Elisa) using competitive enzyme immunoassay reaction as described by Chopra *et al.*, (1971b). T₄ also called thyroxine was also determined by Enzyme Linked Immunosorbent Assay (Microwell Elisa) method using monoclonal antibodies specific for T₄ as described by Chopra *et al.*, (1971a). TSH called thyrotrophin was determined by the enzyme Linked Immunosorbent Assay (Elisa) method for the quantitative determination in serum as described by Hopton and Harrap, (1986); Bravermann (1996); Fisher (1996). The direct immunoenzymatic assay (Elisa method) described by Sood, (2006) was used for the determination of testosterone and that described by Tsang *et al.*, (1980) was used for oestrogen determination. Insulin determination was carried out by the immunoenzymometric assay described by Sood, (2006).

Determination of total cholesterol

Two rats in each group were humanely sacrificed by cutting the throat with a sterile blade. Blood was collected from the vena cava into clean, labelled centrifuge tubes without anticoagulant after the extract had been allowed to act for 24, 48, 72 hrs respectively. The blood was centrifuged at a rate of 12,000 rotations per minute (rpm) for 10 minutes. The clear, yellow serum was then separated from settled cellular elements. Cholesterol was assayed by Tindar's reaction (Evans and Stein, 1986; NIH, 1990) using commercial kits, from Fortress Diagnostic Ltd, Antrim.

Statistical analysis

Test of significance between control and treatment means were carried out by 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 study. Also, there was a significant percentage weight gain ($p < 0.05$) in the hyperlipidaemic rats (Groups two-six) when compared with Group one which received standard diet and water *ad libitum*.

Effect on Thyroid Hormones

The results of the effect of the aqueous fruit extract of *Solanum macrocarpum* on thyroid hormones are shown in Table 2. The TSH and T₄ increased with increase in extract dose. The increase in TSH and T₄ were significant ($p < 0.05$) at 72 hrs, whilst there was no change for T₃ ($p > 0.05$) throughout the period of study.

Effect on Testosterone, Oestradiol and Insulin

The effect of the aqueous fruit extract of *Solanum macrocarpum* on testosterone, oestradiol and insulin are shown in Table 3. Oestradiol levels increased significantly ($p < 0.05$) at 24 hrs and 72 hrs when compared to the negative control (Group one), but the levels of the oestradiol level in the negative control (Group one) were however higher than that of the rats not administered the extract (positive control or Group two). The positive control had an oestradiol level of (15.00 ± 4.24) pg/ml, (12.00 ± 1.49) pg/ml and (13.50 ± 2.54) pg/ml at 24 hrs, 48 hrs and 72 hrs respectively whilst the values of the oestradiol in the negative control rats (Group one) are (36.50 ± 2.12) pg/ml, (20.00 ± 4.24) pg/ml and (24.50 ± 2.12) pg/ml at 24 hrs, 48 hrs and 72 hrs respectively.

The increase in the testosterone level with increase in extract dose was significant at 24 hrs and 48 hrs of study ($p < 0.05$). The testosterone levels of the positive control (Group two) however remained below 0.20 mg/ml (i.e. very low) throughout the period of study.

Insulin levels increased significantly ($p < 0.05$) at 48 hrs and 72 hrs when compared to the negative control (Group one). The levels of insulin in the negative control (Group one) were higher than that of the rats not administered the extract (positive control or Group two).

Effect of extract on total cholesterol

The effect of the aqueous fruit extract of *Solanum macrocarpum* on total cholesterol of hyperlipidaemic rats administered orally with triton-X for 90 days is shown in Table 4. There was a non-significant ($P > 0.05$) increase in total cholesterol when compared to the positive control with increase in extract dose at 24, 48 and 72 hrs respectively. The oral

administration of triton-X resulted in a rise in serum cholesterol of rats in the positive control group (i.e. those administered only Triton-X).

DISCUSSION

The increase in mean body weight of the rats after triton-X administration for 90 days was significant ($p < 0.05$) [Groups two to six], whilst Group one fed with normal diet was not significant ($p > 0.05$). The percentage weight gain in the hyperlipidaemic rats (Groups two to six) 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 hyperlipidaemic rats fed with graded doses of the aqueous fruit extract of *S. macrocarpum* had significant increase ($p < 0.05$) in the level of the thyroid hormones TSH and T_4 at 72 hrs (Table 2). There was no change in T_3 ($p > 0.05$) throughout the period of study. Thyroid hormones increase the expression of LDL receptors on the hepatocytes and increase the activity of lipid-lowering enzymes, resulting in a reduction in LDL (Ness and Lopez, 1995; Ness *et al.*, 1998; Shin and Timothy, 2003; Gali, 2007). Thus, increase in TSH and T_4 levels as demonstrated by the aqueous fruit extract of *S. macrocarpum* on triton-X induced hyperlipidaemic rats at 72 hrs increased further the activity of these lipid-lowering enzymes, thus resulting in a reduction of LDL which is responsible for hyperlipidaemia. In addition, thyroid hormones also increase the expression of apolipoprotein A_1 , a major component of HDL (Taylor *et al.*, 1996; Gali, 2007). Since increased thyroid hormones leads to increased HDL, which in turn leads to reduced hyperlipidaemia, then the aqueous fruit extract of *S. macrocarpum* could probably reduce hyperlipidaemia and could thus be beneficial. T_3 (the active hormone) is important in hepatic degradation of cholesterol into bile acids by increasing the transcription of rate-limiting enzyme in the process, the cholesterol 7-hydroxylase (Ness *et al.*, 1990; Pandak *et al.*, 1997; Gali, 2007). However, graded doses of *S. macrocarpum* aqueous fruit extract led to no change in T_3 levels. Nonetheless, the effects of the thyroid hormones (TSH and T_4) could be beneficial in reducing the onset of atherosclerosis. The TSH of the anterior pituitary controls the thyroid function (Gali, 2007). Generally, thyroid hormones are found at lower levels with a high fat diet (Williamson *et al.*, 1996). It is therefore not surprising that the hyperlipidaemic rats which were not administered the aqueous fruit extract (Group two) had reduced levels of thyroid hormones when compared to the extract-treated rats (Groups three to six). Thyroid hormones stimulate the synthesis, mobilisation and degradation of lipids. They also lower cholesterol level, and for this reason, D-thyroxine is used in the treatment of hyperlipoproteinaemia (Aliu, 2007).

Testosterone has been found at lower levels with a high fat diet (Williamson *et al.*, 1996). Testosterone the dominant androgen found in the adrenal glands, brain, ovary, pituitary, skin, kidney and testes is reduced in male with myocardial infarction (Williamson *et al.*, 1996). Thus the decrease in testosterone observed in the untreated male hyperlipidaemic rats (Group two) in this study agrees with this report. There was a dose-dependent increase ($p < 0.05$) in the level of testosterone on extract administration at 24 hrs and 48 hrs implying therefore that the hyperlipidaemia might probably have reduced in the rats on administration of the aqueous fruit extract of *S. macrocarpum*.

Oestradiol levels have been shown to be decreased in patients with myocardial infarction (Odotola, 1992; Williamson *et al.*, 1996). On administration of the aqueous fruit extract of *Solanum macrocarpum* to the hyperlipidaemic rats, the levels of oestradiol increased correspondingly ($p < 0.05$) from 25 mg/kg to 200 mg/kg dose at 24 hrs and 72 hrs with the highest increase being obtained at 200 mg/kg. The increase in oestradiol implies therefore that the hyperlipidaemia is probably being reduced. The positive control (Group two) i.e. the rats not administered extract, had lower levels of oestradiol when compared to the extract-treated rats (Groups three to six), indicating hyperlipidaemia in the untreated rats. Oestrogens have been shown to decrease LDL-C by only 5-10%. It is therefore believed that the lipid effect account for only 25-40% of the reduction in the incidence of coronary heart disease after oestrogen therapy. A possible mechanism may be a direct effect on the vessel wall, improving blood flow and inhibiting the atherogenic mechanism, independent on effect on plasma lipids (ANON 2008). However, data on men suggest that a 35% reduction in LDC-C is required to achieve a 50% reduction in cardiovascular disease i.e. coronary atherosclerosis provided that LDC reductions are the sole cause of cardio-protection (ANON, 2008). Apart from maintaining friendly lipid profile, oestrogen changes the vascular tone by increasing nitric oxide production. Nitric oxide activates guanyl cyclase, increasing intravascular levels of cyclic guanosine 3', 5'-monophosphate (cyclic GMP), and thereby produces vasodilation (Hardman and Limbird, 2001; Sodipo, 2007). They reduce myocardial oxygen requirement by decreasing peripheral vascular resistance (PR) or by reducing cardiac output (CO) or by decreasing both PR and CO, as $BP = TPR \times CO$ (Katzung, 2004). Oestrogen also stabilizes the endothelial cells. In Australia, the natural oestrogen grazing of grass is implicated in causing vaginal prolapse in ewes and balanoposthitis in wethers.

Also, oestrogenmimetic compounds (flavonoids) are found in many plants and studies have shown that eating these plant products may produce slight oestrogen effects (Katzung, 2004). Sodipo *et al.*, (2008a) has shown that the fruit of *S. macrocarpum* contains flavonoids which have also been separated, isolated and purified in the ethanol extract of the plant (Sodipo *et al.*, 2012b). Thus the fruit of this plant may be eaten as a nutraceutical in lowering LDL-C which in turn lowers hyperlipidemia. Naturally occurring steroidal oestrogens are subject to first-pass hepatic metabolism and show low bioavailability when administered orally (Aliu, 2007). This study on *S. macrocarpum* may be expanded further to see if there is any way of isolating oestrogens from the fruit or other parts of the plant if present, so that it can be a ready source of oestrogens, especially in our menopausal women.

Insulin levels have been found to be decreased in patients with myocardial infarction (Williamson *et al.*, 1996). From the present study (Table 3), the extract significantly increased ($p < 0.05$) the level of insulin in hyperlipidaemic rats at 48 hrs and 72 hrs and the increase was dose-dependent. In addition to promoting systemic atherosclerosis, dyslipidaemia may also contribute to the development of glomerulosclerosis and therefore chronic renal failure (John, 1991; Mshelia and Gashau, 2007). Furthermore, islet- β -cell dysfunction caused by lipotoxicity may result in hyperglycaemia, which also leads to hyperlipidaemia-vicious cycle (Sjoholm, 1998a; Aliu, 2007; Mshelia and Gashau, 2007). Also the decreased HDL-C found in diabetes mellitus patients are not typically associated with marked increase in plasma total cholesterol and LDL-C (Cabezes *et al.*, 1993; Syvanne and Taskinen, 1997). Also there seems to be few studies relating diabetes to hyperlipidaemia (Sjoholm, 1998a). VLDL contains more triglycerides and less cholesterol, phospholipids and protein. They transport triglycerides from the liver to other tissues for utilization or storage. Triglycerides are removed from VLDL and chylomicrons by lipoprotein-lipase. The activity of this enzyme is increased by insulin and thyroxine (Hardman and Limbird, 2001). Thus, by increasing level of insulin at 48 hrs and 72 hrs, the aqueous fruit extract of *S. macrocarpum* increases the conversion of VLDL to triglycerides and thus reduces hyperlipidaemia as triglycerides are being utilized. It should be noted that chylomicrons are synthesized from fatty acids of dietary triglycerides and cholesterol absorbed from the small intestine by epithelial cells using the enzyme, diacylglycerol transferase (Hardman and Limbird, 2001). Overproduction of insulin (hyperinsulinaemia) is rare, but increased levels in the blood have been incriminated in bovine ketosis and also in snorter dwarfism of beef cattle (Aliu, 2007).

CONCLUSION

The aqueous fruit extract of *S. macrocarpum* increased the level of endocrine function indices—thyroid hormones (T_3 , T_4 , TSH), reproductive hormones (testosterone and 17 β -oestradiol) and insulin, the pancreatic hormone under the conditions of this study, probably contributing to the lipid lowering effect of the plant.

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

Group	Mean body Weight \pm S.D. (g)				% Increase in Mean Body Weight
	Days of Treatment				
	0	30	60	90	
One*	110.25 \pm 10.50	112.50 \pm 20.45 ^a	114.00 \pm 12.51 ^a	117.20 \pm 15.07 ^a	6.30 \pm 4.57 ^a
Two	100.20 \pm 26.64 ^a	135.80 \pm 41.26 ^b	203.44 \pm 52.97 ^b	214.20 \pm 58.61 ^b	113.78 \pm 32.37 ^b
Three	80.00 \pm 17.25 ^a	110.20 \pm 27.52	163.64 \pm 26.93 ^b	174.20 \pm 15.06 ^b	117.75 \pm 2.19 ^b
Four	99.40 \pm 29.19 ^a	131.40 \pm 41.58 ^b	184.80 \pm 37.58 ^b	216.80 \pm 41.05 ^b	117.30 \pm 11.86 ^b
Five	116/60 \pm 42.58 ^a	129.00 \pm 11.92 ^b	172.78 \pm 17.03 ^b	194.80 \pm 19.74 ^b	67.07 \pm 22.84 ^b
Six	95.00 \pm 20.96 ^a	120.40 \pm 36.65 ^b	192.18 \pm 34.03 ^b	211.95 \pm 33.74 ^b	122.11 \pm 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 not administered triton-X

Table 2: Effect of the aqueous fruit extract of *S. macrocarpum* on thyroid hormones

Hours after administration	Group	Extract dose (mg/kg)	TSH (ng/ml)	T ₃ (ng/ml)	T ₄ (μ U/ml)
			Mean \pm S.D.		
24	One	-ve control	0.45 \pm 0.21 ^a	0.70 \pm 0.14 ^a	3.60 \pm 0.57 ^a
	Two	+ve control	0.30 \pm 0.00 ^a	0.45 \pm 0.21 ^a	3.25 \pm 0.35 ^a
	Three	25.00	0.35 \pm 0.00 ^a	0.60 \pm 0.28 ^a	3.40 \pm 0.14 ^a
	Four	50.00	0.40 \pm 0.14 ^a	0.70 \pm 0.14 ^a	4.30 \pm 0.43 ^a
	Five	100.00	0.45 \pm 0.14 ^a	0.75 \pm 0.07 ^a	4.35 \pm 0.50 ^a
	Six	200.00	0.50 \pm 0.14 ^a	0.80 \pm 0.00 ^a	4.50 \pm 0.71 ^a
48	One	-ve control	0.50 \pm 0.00 ^a	0.70 \pm 0.14 ^a	3.60 \pm 0.35 ^a
	Two	+ve control	0.25 \pm 0.00 ^a	0.35 \pm 0.07 ^a	3.25 \pm 0.59 ^a
	Three	25.00	0.40 \pm 0.14 ^a	0.50 \pm 0.14 ^a	3.40 \pm 0.14 ^a
	Four	50.00	0.50 \pm 0.00 ^a	0.60 \pm 0.14 ^a	4.30 \pm 0.43 ^a
	Five	100.00	0.65 \pm 0.50 ^a	0.70 \pm 0.14 ^a	4.35 \pm 0.50 ^a
	Six	200.00	0.85 \pm 0.21 ^a	0.80 \pm 0.00 ^a	4.50 \pm 0.71 ^a
72	One	-ve control	0.50 \pm 0.28 ^a	0.80 \pm 0.14 ^a	3.65 \pm 0.21 ^a
	Two	+ve control	0.45 \pm 0.07 ^b	0.30 \pm 0.14 ^a	2.65 \pm 0.50 ^b
	Three	25.00	0.55 \pm 0.35 ^b	0.55 \pm 0.07 ^a	2.75 \pm 0.35 ^b
	Four	50.00	0.65 \pm 0.35 ^b	0.85 \pm 0.21 ^a	3.80 \pm 0.28 ^b
	Five	100.00	0.80 \pm 0.28 ^b	0.90 \pm 0.28 ^a	4.20 \pm 0.42 ^b
	Six	200.00	0.75 \pm 0.36 ^b	0.95 \pm 0.35 ^a	4.45 \pm 0.07 ^b

Within columns, means with different superscripts are statistically significant ($p < 0.05$) when compared to Group 1 (-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

Table 3: Effect of the aqueous fruit extract of *S. macrocarpum* on testosterone oestradiol and insulin

Hours after administration	extract Group	Extract dose (mg/kg)	Testosterone (ng/ml)	Mean ± S.D.	
				Oestradiol (pg/ml)	Insulin (µU/ml)
24	One	-ve control	1.50±0.28 ^a	36.50±2.12 ^a	4.00±1.41 ^a
	Two	+ve control	<0.20	15.00±4.24 ^b	2.00±0.00 ^a
	Three	25.00	<0.20	18.00±4.24 ^b	1.20±0.05 ^a
	Four	50.00	0.20±0.07 ^b	19.00±1.41 ^b	2.00±0.00 ^a
	Five	100.00	0.50±0.42	20.50±0.71 ^b	2.75±0.35 ^a
	Six	200.00	1.05±0.01 ^a	25.00±2.85 ^b	3.00±0.00 ^a
48	One	-ve control	1.55±0.43 ^a	20.00±4.24 ^a	5.00±0.00 ^a
	Two	+ve control	<0.20	13.00±1.49 ^a	2.50±0.07 ^b
	Three	25.00	<0.20	14.00±4.24 ^a	1.00±0.00 ^b
	Four	50.00	0.25±0.35 ^b	14.50±2.12 ^a	2.50±0.71 ^b
	Five	100.00	0.50±0.42 ^b	15.50±0.71 ^a	3.00±0.00 ^b
	Six	200.00	1.35±0.21 ^b	18.00±2.83 ^a	4.00±0.71 ^b
72	One	-ve control	1.60±0.35 ^a	24.50±2.12 ^a	3.00±0.00 ^a
	Two	+ve control	<0.20	13.50±3.54 ^b	2.05±0.07 ^b
	Three	25.00	<0.20	15.00±1.41 ^b	<1.00
	Four	50.00	<0.20	19.00±4.24 ^b	1.00±0.00 ^b
	Five	100.00	0.55±0.07 ^a	21.50±2.12 ^b	2.15±0.07 ^b
	Six	200.00	1.05±0.21 ^a	24.50±2.12 ^b	3.00±0.00 ^b

Within columns, means with different superscripts are statistically significant (p<0.05) when compared to Group 1 (-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

<0.20ng/ml = Very low for testosterone, <1.00µUL/ml = Very low for insulin

Table 4: Effect of the aqueous fruit extract of *S. macrocarpum* on total cholesterol of hyperlipidaemic rats administered orally with triton-X for 90 days

Hours after extract Administration	Group	Extract dose (mg/kg)	Total Cholesterol (mmol/L)
			Mean ± S.D.
24	One	-ve control	1.70±0.28 ^a
	Two	+ve control	2.40±0.29 ^a
	Three	25.00	2.15±0.64 ^a
	Four	50.00	2.10±0.57 ^a
	Five	100.00	1.35±0.07 ^a
	Six	200.00	1.15±0.07 ^a
48	One	-ve control	1.70±0.14 ^a
	Two	+ve control	2.55±0.07 ^a
	Three	25.00	2.10±1.13 ^a
	Four	50.00	1.50±0.14 ^a
	Five	100.00	1.45±0.07 ^a
	Six	200.00	1.25±0.35 ^a
72	One	-ve control	1.90±0.14 ^a
	Two	+ve control	2.40±0.50 ^a
	Three	25.00	2.20±0.28 ^a
	Four	50.00	2.10±0.42 ^a
	Five	100.00	1.70±0.14 ^a
	Six	200.00	1.35±0.07 ^a

Within columns, means with the same superscript are not statistically significant ($p>0.05$) when compared to Group 1 (-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

