EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 5
Issue: 2
440-444
Year: 2018

ANTIHYPERTRIGLYCERIDEMIC PROPERTIES OF *SORGHUM VULGARE* LEAF SHEATH ON HAEMATOLOGICAL INDICES AND LIVER FUNCTION MARKERS ON HIGH FAT DIET DYSLIPIDEMIC WISTAR-ALBINO RATS

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Article Received on 11/12/2017 **Article Revised on 01/01/2018** **Article Accepted on 21/01/2018**

ABSTRACT
Hypertriglyceridemia is a condition in which the concentration of triglyceride in the blood is high. Hypertriglyceridemia can lead to atherosclerosis and severe conditions, acute pancreatitis. The aim of this study was to investigate the antihypertriglyceridemic properties of *Sorghum vulgare* leaf sheath on haematological indices and liver function markers on high fat diet dyslipidemic wistar-albino rats. Thirty-six Wistar-albino rats weighing 110-130g were used for the study. The animals were distributed randomly into six groups of six animals each. Group 1 (control), group II to group VI were induced with high fat diet; group II(untreated), groups III was treated with atorvastatin (0.2mg/kg), whereas groups IV to VI received 400mg/kg, 800mg/kg and 1200mg/kg of the plant extract respectively. The results of the study showed that aqueous extract of *Sorghum vulgare* leaf sheath (at 800mg/kg) significantly reduced ($p \leq 0.05$) triglyceride, Very low density lipoproteins and bilirubin concentrations. Aspartate aminotransferase activity and Alanine aminotransferase activities also reduced significantly($p \leq 0.05$). Total cholesterol, Low Density Lipoprotein, High density lipoprotein, Red blood cell, Haemoglobin, Packed cell volume, Neutrophils and Lymphocytes concentrations do not differ significantly ($p \leq 0.05$). The results suggest that aqueous extract of *Sorghum vulgare* leaf sheath (at 800mg/kg) could be used in the treatment of hypertriglyceridemia and its related complications in murine models.

KEYWORDS: *Sorghum vulgare*, hypertriglyceridemia, cardiovascular diseases.

INTRODUCTION
Hypertriglyceridemia is a dyslipidemic condition in which there is elevated triglyceride concentration in the blood. Other dyslipidemic features include elevated serum concentrations of total cholesterol, low density lipoprotein (LDL), very low density lipoprotein (VLDL) and reduced high density lipoprotein (HDL) concentration (Olorunnisola *et. al.*, 2012).

High concentrations of triglyceride is associated with atherosclerosis, even in the absence of hypercholesterolemia and predispose to cardiovascular disease. Very low density lipoprotein (VLDL) and intermediate density lipoprotein (IDL) have been found in atherosclerotic plagues. Very elevated concentrations of triglyceride can also increase the risk of acute pancreatitis. Hypertriglyceridemia is essential in metabolic syndrome, which include low concentration of High density lipoprotein (HDL-C), insulin resistance, hypertension and abdominal obesity. The triglyceride's concentration can be increase by total fat, alcohol and excess calories. Lifestyle changes such as healthy diet, regular exercise, avoidance of tobacco smoking can favourably affect total cholesterol, HDL cholesterol, LDL cholesterol and triglyceride concentrations (ACP, 2010). Dyslipidaemia can also result to non-alcoholic liver fatty disease (*Kneeman et. al.*, 2011).

Sorghum vulgare belongs to the grass family. Phytochemical analysis of *Sorghum vulgare* leaf sheath shows the presence of tannins, flavonoids, phenols, saponins and phytate (Oyetayo and Ifedayo 2012). Some of these phytochemicals have antioxidant properties and chelating properties that could help in reducing hyperlipidaemia (Nwiloh et.al., 2016).

2.0 METHODLOGY
2.1 Collection and Preparation of aqueous extract of the plants
The dry plant was gotten from mile 3 market Port Harcourt, Nigeria. The plant was ground into fine powder with a blender and stored in an air tight container. The ground powder was macerated in distilled water for 12hrs(1Kg/1L). The macerate was filtered

using Whatman filter paper (No 1) and the filtrate was concentrated using a water bath (60^{o}C) to obtain concentrated crude extract. The extract was stored in a freezer until further use.

2.2 Experimental animals

Thirty-six (36) wistar-albino rats weighing, 110g-130g was used for the study. The experiment lasted for a period of forty-two days. The animals were acclimatized for a period of 14 days before use. Following acclimatization, the animals were distributed randomly into six groups of six (6) animals each. Group 1 was the control and fed with growers feed only. Group II to group VI were induced with hyperlipidemia using high fat diet (10g of egg yolk per 40g of the feed) for a period of four weeks. At the third week of inducement of hyperlipidemia with high fat diet, treatment of the animals with the plant's aqueous extract and standard drug commenced for a period of two weeks. Group II was untreated, group III was treated with atorvastatin (0.2mg/Kg) which is a standard drug, while groups IV to VI received 400mg/kg, 800mg/kg and 1200mg/Kg of the plant's aqueous extract respectively. At the end of the study, the animals were sacrificed and their blood samples was collected for biochemical and haematological study.

2.3 Biochemical studies

Total Cholesterol (TC), Triglycerides, (TG), Low Density Lipoprotein Cholesterol (LDL), High Density Lipoprotein Cholesterol (HDL) using randox kits (sigma aldrich).

Plasma aspartate aminotransferase (ASAT), alanine aminotransferase (ALAT) and Alkaline phosphatase (ALP) activities was estimated using colorimetric methods (Reitmann and Frankel, 1957: Beyegue *et. al.*, 2012). Also, bilirubin using Jendrassik and Groff (1938) method and albumin concentrations was analysed using bromocresol green method in order to evaluate the liver function.

2.4 Determination of Haematological parameters

Haemoglobin, white blood cell, Packed cell volume, red blood cell, neutrophils and lymphocytes levels was determined.

2.5 Statistical Analysis

Data was represented as mean ± SEM and subjected to One-way Analysis of Variance (ANOVA) using Statistical software SPSS. A level of $p \leq 0.05$ was considered as statistically significant.

3.0 RESULTS

Table I. Results of the aqueous extract of *Sorghum vulgare* leaf sheath on lipid profile of high fat diet induced dyslipidemic rats.

Groups	TC(mmol/l)	TG (mmol/l)	HDL(mmol/l)	LDL(mmol/l)	VLDL(mmol/l)
Control	3.60^{a} ±0.14	2.05^{a}±0.06	0.56^{a}±0.07	1.77^{a}±0.28	0.41^{a}±0.01
HFD	3.95^{a} ±0.17	2.90^{b}±0.17	0.79^{b}±0.06	0.52^{b}±0.30	0.58^{b}±0.03
HFDATV	4.22^{a} ±0.18	2.30^{a}±0.25	0.67^{b}±0.01	1.62^{a}±0.11	0.46^{a}±0.05
HFD400	4.55^{a} ±0.80	2.15^{a}±0.06	0.81^{b}±0.07	1.80^{a}±0.17	0.43^{a}±0.01
HFD800	3.62^{a} ±0.35	2.15^{a}±0.17	0.99^{b}±0.09	0.84^{b}±0.44	0.43^{a}±0.03
HFD1200	4.02^{a} ±0.11	2.22^{a}±0.06	0.98^{b}±0.06	0.81^{b}±0.12	0.44^{a}±0.01

Values are expressed as Mean ± SEM. Values in a column with the same alphabetical superscript do not differ significantly ($p \leq 0.05$). Values in a column with different alphabetical superscript differ significantly ($p \leq 0.05$).

Table II. Results of the aqueous extract of *Sorghum vulgare* leaf sheath on white blood cell and differentiated white blood cell of dyslipidemic rats.

Groups	WBC (x 10^{9})	N(%)	L(%)
Control	6.40^{a}±0.41	27.25^{a}±3.68	68.25^{a}± 4.97
HFD	9.62^{b}±1.62	30.50^{a}±3.32	68.25^{a}±2.83
HFDATV	6.23^{a}±0.88	31.00^{a}±1.47	66.00^{a}±1.47
HFD400	6.50^{b}±1.02	38.33^{a}±2.32	60.00^{a}± 2.04
HFD800	4.20^{ab}±0.51	29.50^{a}±4.21	69.25^{a}±4.88
HFD1200	7.67^{b}±1.23	30.00^{a}±2.19	66.75^{a}±1.97

Values are expressed as Mean ± SEM. Values in a column with the same alphabetical superscript do not differ significantly ($p \leq 0.05$). Values in a column with different alphabetical superscript differ significantly ($p \leq 0.05$).

Table III. Results of the aqueous extract of *Sorghum vulgare leaf* sheath on erythropoietic parameters of dyslipidemic rats.

Groups	PCV(%)	Hb(mg/dL)	RBC(x10^{12})
Control	40.25^{a}±0.75	13.42^{a}±0.25	6.02^{a}±0.10
HFD	43.75^{a}±1.43	14.57^{a}±0.48	6.62^{a}±0.14
HFDATV	33.00^{c}±0.40	11.00^{c}±0.12	4.00^{c}±0.81
HFD400	42.33^{a}±2.39	14.10^{a}±0.75	6.10^{a}±0.52
HFD800	43.00^{a}±1.35	14.35^{a}±0.45	6.37^{a}± 0.21
HFD1200	41.75^{a}±2.71	13.90^{a}±0.90	5.85^{a}± 0.41

Values are expressed as Mean ± SEM. Values in a column with the same alphabetical superscript do not differ significantly (p≤ 0.05). Values in a column with different alphabetical superscript differ significantly (p≤ 0.05).

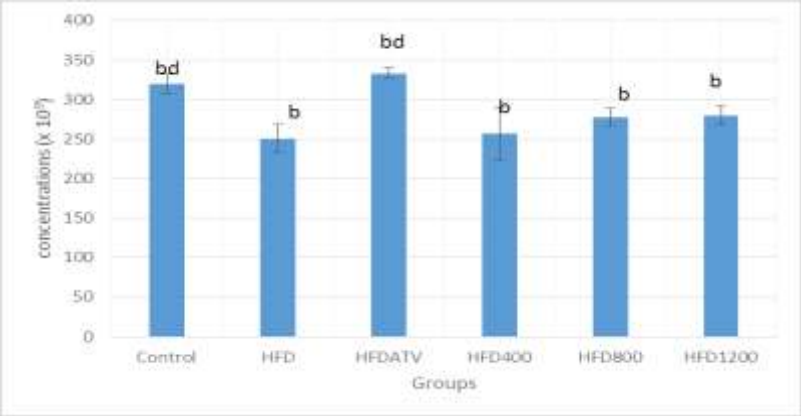

Figure 1: Concentration of blood platelets on dyslipidemic wistar-albino rats treated with aqueous extract of *Sorghum vulgare* leaf sheath.

Values are expressed as Mean ± SEM. Values in a column with the same alphabetical superscript do not differ significantly (p≤ 0.05). Values in a column with different alphabetical superscript differ significantly (p≤ 0.05).

Table IV. Hepatic Enzymes Activity of dyslipidaemic witar-albino rats, treated with aqueous extract of *Sorghum vulgare* leaf sheath on dyslipidemic rats.

Groups	AST(U/L)	ALT(U/L)	ALP(U/L)
Control	172.25^{bc}± 2.25	40.10^{a}±4.91	35.50^{a}±2.95
HFD	190.05^{b}± 3.22	50.00^{b}±2.16	32.00±^{b}.70
HFDATV	203.50^{b} ±2.25	55.50^{b}±2.66	29.25^{b}±1.25
HFD400	177.50 ^{bc}± 4.29	38.00^{d}±0.81	29.50^{b}±1.32
HFD800	167.00^{bc}± 2.04	31.25^{d}±1.18	34.25^{b}±1.84
HFD1200	191.75^{b} ± 3.14	48.50^{b}±2.32	31.75^{b}±0.47

Values are expressed as Mean±SEM. Values in a column with the same alphabetical superscript do not differ significantly (p≤0.05). Values in a column with different alphabetical superscript differ significantly (p≤ 0.05).

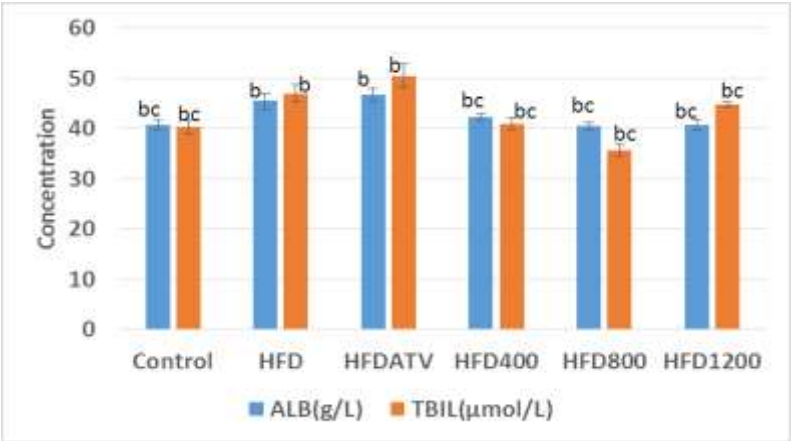

Figure 2: Concentration of liver function markers on dyslipidemic wistar-albino rats, treated with aqueous extract of *Sorghum vulgare* leaf sheath.

Values are expressed as Mean±SEM. Values in a column with the same alphabetical superscript do not differ significantly (p≤ 0.05). Values in a column with different alphabetical superscript differ significantly (p≤ 0.05).

4.0 DISCUSSION

Dyslipidemia, in which hypertriglyceridemia is one of it, is one of the major factors for type 2 diabetes, atherosclerosis, stroke and cardiovascular diseases development (Li *et. al.*, 2015). Table I shows the result of aqueous extract of *Sorghum vulgare* leaf sheath on lipid profile of high fat diet induced dyslipidemic rats. There was significant reduction (p ≤0.05) in triglyceride and very low density lipoproteins concentration of all treated groups when compared to the hyperlipidemic untreated group (group II). There was no significant difference in HDL concentration, Total cholesterol and LDL concentrations (p ≤0.05).

There is increasing evidence that atherosclerosis is accompanied by inflammation. White blood cell (WBC) count, fibrinogen and C-reactive proteins are all positively associated with increased cardiovascular mortality, mainly from coronary heart disease and ischemic stroke (Peter, 2004). The results in Table II showed significant reduction (p≤0.05) in the concentration of white blood cell of hyperlipidemic rats treated with atorvastatin (0.2mg/kg) and 800mg/kg of aqueous extract of *Sorghum vulgare* leaf sheath when compared to the hyperlipidemic untreated animals.

Low Red blood cell (RBC), packed cell volume (PCV) and haemoglobin (Hb) concentrations could pose anemia and elevated concentrations may result from increased erythropoietin production due to chronic hypoemia or in-appropriate erythropiotein secretion especially in lungs or renal disorder, liver disease or certain forms of heart disease (Arika *et. al*., 2016). Anemia due to low haemoglobin causes tissue hypoxia and changes in blood flow patterns which may play an atherogenic role (Ioana, 2015). The results on Table III shows no significant difference(p≤0.05) in the RBC, Hb and PCV concentrations.

Platelets are used in blood clotting. Few platelets result in uncontrolled bleeding while many platelets could cause clot formation in the blood vessels in cases like hardening of the arteries during atherosclerosis. Elevated platelets may suggest that the extract has stimulating effect on thrombopoietin and can be used to manage haemophilia. Hypertriglyceridemia can lead to triglyceride-rich very low density lipoprotein (VLDL) that potentiates platelet activity leading to hardening of the arteries, an effect mediated partly through apolipoprotein E and an interaction with the platelet LDL receptor (Kakouro *et. al.*, 2011). Figure 1 shows the Concentration of blood platelets on dyslipidemic wistar-albino rats treated with aqueous extract of *Sorghum vulgare* leaf sheath. There was no significant difference (p≤0.05) of the platelet count of the animals treated with the plant extract when compared to hyperlipidemic untreated animals. Although there was significant decrease (p≤0.05) in the platelet count of the animals treated with 400mg/kg of aqueous extract of *Sorghum vulgare* leaf when compared to the control and hyperlipidemic animals treated with atorvastatin (0.2mg/kg). The resultssuggests that the extract does not pose platelet hyperactivity, thus does not contribute to arteries hardening.

Transaminases are markers for hepatocyte integrity. Aspartate aminotransferase (AST) leaks when the liver, heart, skeletal muscle or erythrocytes are injured. Alanine aminotransferase (ALT), is another liver enzyme which increases specifically for liver injury. Alkaline phosphatase (ALP), is a biomarker for poor bile flow. ALP is induced in the liver cells and appears whenever there is back up of bile in any part of the biliary tree (Sheriff, 2004). The results of Hepatic Enzymes Activity on dyslipidemic witar-albino rats, treated with aqueous extract of *Sorghum vulgare* leaf sheath dyslipidemic rats is shown on Table IV. There was significant reduction (p ≤0.05) in AST activity and ALT activity of hyperlipidemic animals treated with 400mg/Kg and 800mg/kg of aqueous extract of *Sorghum vulgare* leaf sheath when compared to the hyperlipidemic untreated animals (group II) and the hyperlipidemic animals treated with 0.2mg/kg of atorvastatin (group III). There was no significant difference (p ≤0.05) in the ALP activity of the treated animals. The result suggests that the aqueous extract of *Sorghum vulgare* leaf sheath have hepatoprotective effect at 400mg/Kg and 800mg/kg of the plant extract.

Figure 2., shows the concentrations of liver function markers (total bilirubin and albumin) on dyslipidemic wistar-albino rats, treated with aqueous extract of *Sorghum vulgare* leaf sheath. There was significant reduction (p ≤0.05) in the albumin concentration of all groups treated with the aqueous extract of *Sorghum vulgare* leaf sheath, although they were within the normal range; 37-58 g/L or 3.7-5.8g/dL (Charles and Gikins, 2008) when compared to the hyperlipidemic untreated animals (group II) and the hyperlipidemic animals treated with 0.2mg/kg of atorvastatin (group III). The total bilirubin concentration decreased significantly (p ≤0.05) of all treated groups when compared to the hyperlipidemic untreated animals (group II) and the hyperlipidemic animals treated with 0.2mg/kg of atorvastatin (group III). This also, suggests that aqueous extract of have hepato-protective effects.

5.0 CONCLUSION

The results suggest that Aqueous extract of *Sorghum vulgare* leaf sheath has anti-hypertriglyceridemic effect and hepatoprotective properties in murine models.

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