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Antiobesity Effects of Pulp Extract *Tamarindus indica* in Albino Rat

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Abstract: The pulp extract of *Tamarindus indica* is used by traditional herbalists as a purgative, drug vehicle and antiobesity agent. The effect of 28 days administration of *Tamarindus indica* pulp extract on the body weight and lipid profile of the rat was evaluated. There was a significant increase in the weight of the control compared to the treated which significantly decreased ($p<0.05$) especially the rats given higher doses (2700 to 4500 mg kg^{-1} body weight). Serum cholesterol and Low Density Lipoprotein (LDL) revealed a significant decrease ($p<0.05$) while High Density Lipoprotein (HDL) and triglycerides increased in the controlled group compared to the control. Xenical treated group was not significantly different ($p<0.05$) from the control. Triglycerides significantly increased ($p<0.05$) and LDL significantly decreased ($p<0.05$) in the pulp extract treated group as compared to xenical treated group.

Key words: *Tamarindus indica*, antiobesity, lipid profile, cholesterol, high density lipoprotein, low density lipoprotein, triglyceride and xenical

INTRODUCTION

Obesity is by far the most prevalent type of body weight imbalance, which has been recognized as a medical problem of growing concern. According to specialists at the first international obesity conference in Africa which focused on the growing number of obese people across the developing world, the global obesity epidemic is completely out of control, with more than 300 million overweight adults worldwide. Obesity rates are rising exponentially with overweight adults across the world suffering from weight-related illnesses like diabetes, heart disease and sleeping disorders. In Nigeria between 6-8% of people are obese (Genevieve, 2004). The obesity epidemic in the world today, is an unintended consequence of the economic, social and technological advances realized during the past several decades.

With recognition of this epidemic, obesity has an increasing awareness of the need to improve the quality and effectiveness of available treatments. The current core of treatment for obesity includes behavior therapy aimed at modifying eating-related activities, exercise to increase caloric expenditure and diets to lower calorie and fat intake. Pharmacological treatments are generally considered as an adjunct to this core therapy (Bray and Greenway, 1999).

Traditional medicine has been with us for ages but there has been a renewed interest in the subject in the recent past. This may be attributed to the down turn of the economy, as traditional medicine is perceived to be a cheaper means of treatment (Baiyewu, 2001). Even in the developing countries, the popularity of crude herbal products is on the increase. In these technologically advanced society, consumers preference is shifting from purely synthetic drug to nature based drugs (Wambebe, 1998). Factors responsible for this shift are high cost of conventional drugs, relatively high incidence of

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toxicity and side effect, unavailability of orthodox drugs in many rural areas and clinical limitation especially in the management of some chronic disease. Based on the above factors, traditional medicine has some advantages over orthodox medicine (Wambebe, 1998).

The resolution of the 31st World Health Organisation assembly took formal interest in herbal remedies has asked for a proper identification, sustainable exploitation, scientific development and evaluation of efficiency, safety, appropriate utilization and standardization of medicinal plants (WHO, 1991). This call is a follow up of the interest of World Health Organisation in traditional medicine in developing world (Wambebe, 1998).

In Nigeria and Africa at large, there are a lot of such plant waiting exploration, scientific validation of claims and standardization. *Tamarindus indica* is one of such plant. It has an antiobesity claim amongst the young female folk in the northern part of Nigeria. It is also used in traditional medicine for the treatment of cold, fever, stomach disorder, diarrhea and jaundice and as skin cleanser (Doughari, 2006). The pulp is composed of tartaric acid, citric and malic acids, potassium bitartarate, pectin, gum, water and parenchymatous fiber. It is used to allay thirst, is nutritive and forms useful drinks given to persons recovering from sickness to keep their bowels regular (Morton, 1987). It has been used extensively in the general traditional medicinal practice in northern Nigeria as a drug conveyor (in combination with other herbal drugs) and slimming extract for obese individuals particularly among the adult female folk. This study is therefore to ascertain the antiobesity effects of aqueous extract of *Tamarindus indica* pulp on Albino rat.

MATERIALS AND METHODS

Collection of Plant Material

Tamarindus indica pulp was obtained from the wild of Sokoto north local government area, Sokoto- Nigeria. The voucher specimen was deposited in the department of botany of the Usmanu Danfodio University, Sokoto-Nigeria, were it was botanically identified by the herbarium as *Tamarindus indica* pulp locally known as *Tsamiya* in Hausa as recommended by Kumar *et al*. (2000).

Preparation of Extract

The aqueous pulp extract of *Tamarindus indica* was obtained using the hot water extraction technique in order to stimulate the local procedure as described by Akinyole and Olerede (2000). Four hundred gram of *T. indica* pulp was soaked in 2 L of distilled water and boiled for 5 min. This was shaken for 10 min and allowed to cool then filtered using Whatman filter paper No. 1 into a measuring cylinder. The filtrate was then evaporated to a residue in a drying cabinet and stored at 4°C until used. The percentage yield was 25.2% w/w.

Experimental Animal Model

Adult Albino rats weighing 201.6±65 g body weight (b.wt.) were obtained from the animal farmhouse of the Zoology Department of Usmanu Danfodiyo University, Sokoto, Nigeria. They were housed in the experimental animal laboratory of Usmanu Danfodiyo University, Sokoto. The animals were allowed acclimatize for seven days having access to water and food *ad libitum*. They were randomized to one of the following experimental groups, with five animals per group:

Group 1: Normal rats chow and distilled water only (control group).
Group 2: Normal rats chow and 900 mg kg^{-1} b.wt. of the extract.
Group 3: Normal rats chow and 1800 mg kg^{-1} b.wt. of the extract.
Group 4: Normal rats chow and 2700 mg kg^{-1} b.wt. of the extract.
Group 5: Normal rats chow and 3600 mg kg^{-1} b.wt. of the extract.

Group 6: Normal rats chow and 4500 mg kg^{-1} b.wt. of the extract.
Group 7: Normal rats chow and xenical (slimming drug).

These daily oral administration continued for 28 days and weights of the rats were measured every week. At the end of the experiment, the animals were fasted overnight and the final weights recorded. Blood samples were collected by individually euthanizing the rats using chloroform. This research was conducted in June/July 2006 at the small animal laboratory of the Faculty of Veterinary Medicine of Usmanu Danfodiyo University, Sokoto, Nigeria.

Weight Measurement

The weight of each rat was measured on a weekly basis using a standard weighing machine with the net weight gain as:

$$\text{Net weight gain} = \text{Initial weight } (W_0) - \text{New weight } (W_1)$$

Blood Sample Collection

Blood samples of 2.5 mL each were collected from each of the rat into two plain labeled dry blood sample containers for lipid profile and fasting blood glucose analysis.

Lipid Profile Determination

Total Cholesterol Determination

This was determined according to the modified enzymatic method of Trinder (1969). The method is based on the ability of all cholesterol ester present in plasma to quantitatively hydrolyze into free cholesterol and fatty acids by cholesterol esterase. In the presence of oxygen, free cholesterol is then oxidized by cholesterol oxidase to cholest-4-ene-3-one and hydrogen peroxide (H_2O_2). The H_2O_2 reacts with phenol and 4-aminophenazone in the presence of peroxidase to form an o-quinoneimine dye. The intensity of the color form is proportional to the cholesterol concentration in the sample with maximum absorption at 500 nm.

Total Triglyceride Determination

Triglyceride estimation was according to the enzymatic method by Folch *et al.* (1957) as modified by Tiez (1976). The method employs the enzymatic hydrolysis of triglycerides by lipase to form glycerol and free fatty acids. The glycerol produced was assayed by enzyme-coupling to form glycerol-3-phosphate. The final measured product of subsequent coupled reaction is proportional to the amount of glycerol, which is in turn proportional to the amount of triglycerides in the sample. Absorbance was taken at 490 nm.

High-Density Lipoprotein Cholesterol (HDL-c) Determination

HDL-c estimation was according to the method of Tiez (1976). The method utilizes the ability to isolate HDL-c from other major classes of plasma lipoproteins by the formation of insoluble complexes of lipoproteins, poly anions and divalent cat ions. In the presence of Mn^{2+} and heparin, chylomicrons, VLDL and LDL are selectively precipitated, leaving only HDL-c in solution. The precipitated lipoproteins are sedimented by centrifugation and the clear HDL-c -containing supernatant is recovered for cholesterol analysis.

Low-Density Lipoprotein-Cholesterol (LDL-c) Determination

LDL determination was as described by Friedewald *et al.* (1972) using the Friedewald formula:

$$\text{LDL-c} = \frac{\text{Total cholesterol} - \text{HDL-c} - \text{Triglyceride}}{5}$$

This formula is based on the assumption that VLDL-c is present in a concentration equal to one-fifth of the triglyceride concentration.

Blood Sugar Determination

This was determined by the enzymatic method of glucose oxidase as described by Trinder (1969). This is based on the ability of glucose to undergo enzymatic oxidation in the presence of glucose oxidase to form gluconolactone and hydrogen peroxide. The hydrogen peroxide formed reacts, under catalysis with phenol and 4-aminophenazone to form a red-violet quinoneimine dye as indicator that was measured with maximum absorbance at 490 nm. The intensity of the color is proportional to the concentration of glucose in the sample.

Statistics

The data collected in the study was subjected to analysis of variance (ANOVA) using the Dunnette multiple comparison test for test of significance. Values are expressed as Means±SEM.

RESULTS

Effects of Aqueous Pulp Extract of *T. indica* on Body Weight of Rat

The weekly body weight changes of the rats following oral administration of aqueous pulp extract of *Tamarindus indica* is shown in Table 1, 2 and Fig. 1. The pulp extract was found to significantly decrease ($p<0.05$) the body weight of the animals in a dose dependent manner. The control on the other hand increased in weight in the first 2 weeks followed by a decrease in the third and then increase in the fourth weeks. There was also a slight increase in body weight in the xenical treated group between the first and second week, this was later lost in the third and fourth week.

Effects of Aqueous Pulp Extract of *T. indica* on Antiobesity Parameters

The result of the antiobesity effects of aqueous pulp extract of *Tamarindus indica* is shown in Table 3. Serum cholesterol and LDL revealed a significant decrease at 3600 mg kg^{-1} ($p<0.05$) and 4500 mg kg^{-1} ($p<0.01$) as compared to that of the control, respectively while significant decrease in

Table 1: Antiobesity effect of *T. indica* pulp extract on the mean body weight of rat administered with aqueous pulp extract of *Tamarindus indica*

		900	1800	2700	3600	4500	
Weeks	Control	----------	----------	(mg kg^{-1})	----------	----------	Xenical
0	270.0±37.6	166.2±05.8	181.6±11.0	177.1±4.9	242.9±15.5	195.6±22.7	177.9±12.8
1	272.9±41.2	166.4±08.5	177.4±14.1	177.3±6.1	246.9±17.2	188.7±17.6	182.6±9.3
2	287.5±43.5	187.8±11.1	193.7±13.1	178.8±8.1	242.5±11.7	187.8±21.3	173.4±9.0
3	283.2±40.6	194.8±17.0	198.5±20.3	174.3±5.6	240.2±17.4	179.1±22.2	170.9±9.9
4	270.4±42.0	188.1±04.4	180.3±13.7	169.5±4.4	236.6±15.3	176.4±21.7	167.9±4.0

Table 2: Antiobesity effect of *T. indica* pulp extract on the weekly mean body weight change total weekly mean body weight change from the original weight of rat administered with aqueous pulp extract of *Tamarindus indica*

		900	1800	2700	3600	4500	
Weeks	Control	----------	----------	(mg kg^{-1})	----------	----------	Xenical
1	-2.9	+0.2	-4.2	+0.2	+4.0	-6.9	+4.7
2	+7.5	+21.6	+2.1	+1.7	+0.4	-7.8	-4.5
3	+3.2	+28.6	+6.9	-2.8	-2.7	-16.5	-7.0
4	+20.0	+21.9	-21.9	-7.6	-6.3	-19.2	-10.0

-: Represent weight loss (g); +: Represents weight gain (g); 1 represents week (0-1); 2 represents week (0-2); 3 represents week (0-3); 4 represents week (0-4) where 0 is the original weight of the rat at start of the experiment

Table 3: Effects of *T. indica* pulp extract on lipid profile

Doses	Total cholesterol (mg dL^{-1})	Triglyceride (mg dL^{-1})	High density lipoprotein (mg dL^{-1})	Low density lipoprotein (mg dL^{-1})	Glucose (m mol L^{-1})
Control	85.5±9.9	142.3±16.1	10.5±2.0	45.8±8.2	5.3±0.7
Treated (mg kg^{-1})					
900	63.7±4.2	157.2±21.3	9.7±0.1	21.6±7.1	7.2±2.0
1800	55.5±1.4*	165.4±22.1	9.7±1.9	25.1±7.3	7.5±1.7
2700	68.4±8.8	140.9±32.2	9.7±0.2	39.1±7.8	6.0±0.8
3600	61.9±2.5*	222.3±6.5•*	15.3±3.6	15.3±3.6*	7.4±2.4
4500	52.6±5.6**	209.0±12.3•	17.2±2.9	9.6±2.5***•	5.1±1.0
Xenical treated	69.8±2.5	128.8±11.3	12.1±0.8	35.4±5.4	7.2±2.4

*: p>0.05 when compared with control; **: p>0.01 when compared with control; •: p>0.05 when compared with xenical

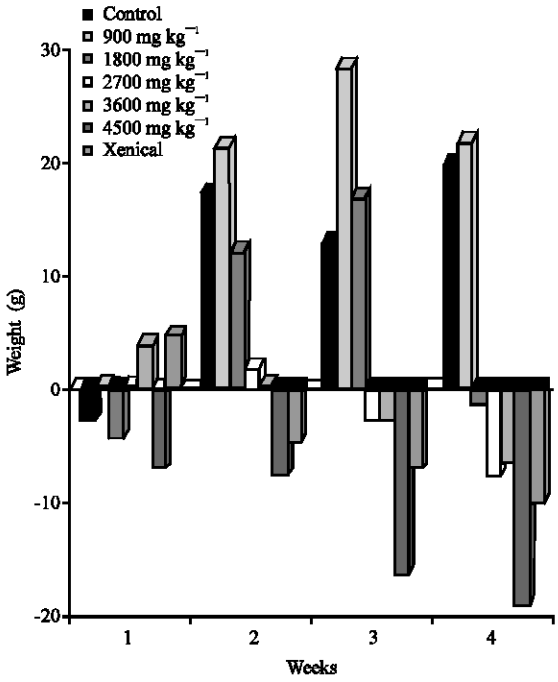

Fig. 1: Weekly body weight change after 28 days administration of aqueous pulp extract of *Tamarindus indica*

LDL at 4500 mg kg^{-1} ($p<0.05$) as compared to xenical treated group. There was no significant difference between the control and the xenical treated group in the entire lipid profile. HDL and Fasting blood glucose was not significantly different at all dose rate of the extract compared to that of xenical treated group and control group respectively. Significant increase in triglyceride was seen at 3600 mg kg^{-1} ($p<0.05$) as compared to control, ($p<0.05$) at 3600 and 4500 mg kg^{-1} as compared to xenical treated group.

DISCUSSION

The initial criteria for obesity treatment proposed after World War II were to estimate the amount of weight loss either in absolute terms or relative to initial weight (Bray and Greenway, 1999). The weekly body weight changes in the rats following oral administration of aqueous pulp extract of

T. indica resulted in a dose dependent decrease in body weight. The decrease in body weight may be attributed to the reduction in food and water intake caused by chemicals that affect brain centers involved in satiety and hunger or could have inhibited digestive enzymes or decreased bioavailability of nutrient caused by antinutritional factors present in plant extract (Moody *et al*., 2003). It could also be attributed to the presence of antinutritional factors like saponins in the extract. Though the rats were fed with diet with adequate protein, the plant extract might not have allowed proper absorption of protein which could account for the decreased body weight (Akinyole and Olerede, 2000).

Cardiovascular disease (CVD) is a major complication of obesity and cause of death in the world, mainly due to atherosclerosis (hardening of the arteries). Abnormal blood lipids are risk factors for CVD (NIH, 2000). Studies have illustrated the beneficial effects of saponins on blood cholesterol levels (Mc Donald *et al*., 2005). This desirable effect is achieved by the binding of bile acids and cholesterol by saponins. Bile acids form mixed micelles (molecular aggregates) with cholesterol, facilitating its absorption. Cholesterol is continually secreted into the intestine via the bile, with much of it subsequently reabsorbed. Saponins cause a depletion of body cholesterol by preventing its reabsorption, thus increasing its excretion, in a similar way as other cholesterol-lowering drugs, such as cholestyramine. Therefore the significant decrease ($p<0.05$) in serum cholesterol may be attributed to the effect of this phytochemicals present in the aqueous pulp extract of this plant. This is also supported by the work of Ray Schealian (2005).

LDL decreased significantly ($p<0.05$) in the pulp extract treated groups in a dose dependent manner and this may be due to significant increased ($p<0.05$) in HDL concentration in the treated groups, as acknowledged HDL major function is in the efflux of cholesterol from tissues thereby reducing the amount of cholesterol (Brown and Goldstein, 1984). Although the role of high triglycerides as an independent factor in the development of CVD remains controversial, data from several prospective studies suggest that triglycerides are probably an important risk factor. Hypertriglyceridaemia is often associated with increased plasminogen activator inhibitor levels and impaired fibrinolysis. Serum Triglycerides of this study significantly increased ($p<0.05$) compared to the control. Fasting blood glucose revealed no significant difference in the extract treated group as compared to the control in contrast to the work of Ray Schealian (2004). Xenical treated group showed significant difference ($p<0.05$) in the triglyceride and LDL as compared to the pulp extract treated group.

Conclusively, this anti-cholesterol activity if further investigated might prove to reduce the complications of obesity. The present research has shown that this is a promising plant and the result confirms its use in traditional medicine for the management and care of overweight and obesity with respect to its hypolipidemic activity although more research has to be carried out to validate this claim.

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