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Effects of *Aframomum melegueta* K. Schum. Leaf on Monosodium Glutamate and High Fat Diet-Induced Obesity in Wistar Rat

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ABSTRACT

Overweight and obesity are described as abnormal or excessive fat accumulation which presents a serious health risk. Natural products, particularly medicinal plants, are believed to possess potential anti-obesity agents. This study investigated the effects of aqueous leaf extract of *A. melegueta* on monosodium glutamate (MSG) and high fat diet (HFD)-induced obesity in Wistar rats with a view to employing the plant as a new class of anti-obesity agent. The Wistar rats (65) of both sexes were randomly grouped into 13 (n = 5). Thereafter, obesity was induced via subcutaneous injection of MSG (2 and 4 mg/g bwt) in the rat neonates and HFD using three models. The rats were then treated with *A. melegueta* extract (200 and 400 mg/kg bwt); and the concentrations of insulin, leptin, lipid (total triglycerides, total cholesterol, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, and very low-density lipoprotein cholesterol) were determined spectrophotometrically. Also lipase activity, cardiac, and atherogenic indices were determined. Results showed that the administration of MSG and HFD caused a significant ($p < 0.05$) increase in insulin-leptin levels and lipase activity. However, the parameters were significantly reduced when treated with the extract. The alterations in lipid profile, atherogenic, and cardiac indices in obese rats were restored to near normal following the administration of the extract. The study concluded that *A. melegueta* leaf possessed anti-obesity property; justifying its use in folk medicine.

Keywords: Obesity, Monosodium glutamate, *Aframomum melegueta*, High fat diet.

Introduction

Globally, the incidences of obesity are rising at alarming rates which is fast becoming a major public health concern with incalculable social costs. Obesity facilitates the development of metabolic disorders such as diabetes, hypertension, and cardiovascular diseases in addition to chronic diseases such as stroke, osteoarthritis, sleep apnea, some types of cancers, and inflammation-induced pathologies.[1] The etiology of obesity could be genetic, dietary, lifestyle, and environmental factors, which favor a chronic positive energy balance, and leads to increased body fat mass.[2] Obesity is more frequent in populations living in environments characterized by long-term positive energy imbalance due to sedentary lifestyle, low resting metabolic rate, or both.[3]

Drugs used in the management of obesity have been reported to possess several side effects; necessitating the need to search for new anti-obesity drugs with little or no side effects.[4] *Aframomum melegueta*, native to West African region, is a plant in ginger family, Zingiberaceae. The plant is obtained from ground seeds and imparts a pungent, peppery flavour with hints of citrus. It is commonly known as grains of paradise, melegueta pepper, alligator pepper, Guinea grains, ossame, fom wisa.

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Phytochemically, the seed contains 6-Gingerol, 6-Shogaol, 6-Paradol, 6-Gingeredione, {2-5-butylfuran-2-yl) ethyl}-2-methoxyphenol, and paradol (most active principle).[5] The plant has been reported to possess anti-inflammatory, anti-measles, anti-asthma, anti-diarrhoea, and energy enhancing properties.[6-8] In the Eastern part of Nigeria and among other cultures of West Africa, the seeds and leaf of *A. melegueta* are added to the diets of newly delivered women and pregnant women to prevent excessive weight gain and after-birth fat accumulation. There are numerous studies on the seeds of *A. melegueta*, however, there is little scientific information on the leaf of the plant. Therefore, this study was undertaken to scientifically investigate the possible anti-obesity effect of the aqueous leaf extract of *A. melegueta* to back up its use in traditional medicine.

Materials and Methods

Plant sample collection

Fresh leaf samples of *A. melegueta* were collected in November, 2019 from Babajakan village in Osun state in Nigeria. Identification and authentication were carried out at IFE Herbarium, Department of Botany, Obafemi Awolowo University, Ile-Ife, Nigeria, with the voucher number 17525.

Plant sample preparation

The leaves were sundried for 14 days and pulverized using electric blender. The aqueous leaf extract of *A. melegueta* was prepared following the modified method.[9] The powdered sample (500 g) was suspended in 6 L of distilled water for 48 h and filtered using muslin cloth and Whatman No 1 filter paper. The residue was re-extracted 5 more times until the filtrate became colourless. The filtrates were combined, allowed to settle, decanted and re-filtered. The combined filtrate was first concentrated *in vacuo* on a rotary evaporator to reduce the volume and then freeze-dried using lyophilizer. The extraction procedure yielded 19.28 g of aqueous extract of *A.*

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melegueta from 500 g starting material. The yield represented 3.86 % of the starting material.

Experimental animals
Adult female (20) and male Wistar rats (10) weighing between 120-150 g were purchased from the Animal House, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, Nigeria. The rats were housed in polyethylene cages and mated in ratio 1:2 (male: female) respectively at the Animal House Unit of the Department of Biochemistry, Adeleke university, Ede. The animals were kept under standard conditions (20–26°C) and were fed with food and water *ad libitum.* The animals were cared for following the guidelines specified in the principles of laboratory animal care (NIH publication no. 82-23, revised 1985). The ethical approval was obtained from the ethical committee of Adeleke University with the ethical approved number: AUERC/FOS/IND/03.

Experimental esign: Induction of obesity
Obesity was induced in neonates (4 days old) with monosodium glutamate (MSG) (2 mg/kg bwt) on post-natal days 4, 6, 8, and 10; and later MSG (4 mg/g bwt) on post-natal days 12, 14, 16 and 18 according to the slightly modified method of Campos-Sep´ulveda[10] and high fat diet (HFD) according to modified method of Penka.[11] The induction was carried out using 3 models, namely:

Model I: Induction of Obesity with Monsodium glutamate (MSG)
Newborn rats (neonates) received subcutaneous injection of MSG (2 mg/g bwt) on post-natal days 4, 6, 8, and 10; and later MSG (4 mg/g bwt) on post-natal days 12, 14, 16 and 18. After weaning (i.e. 3 weeks of birth), the rats were fed with normal standard diet until 16 weeks of age.

Model II: Induction of Obesity with Monosodium glutamate and High Fat Diet
Newborn rats received subcutaneous injection of MSG (2 mg/g bwt) on post-natal days 4, 6, 8, and 10; and later MSG (4 mg/g bwt) from post-natal days 12, 14, 16 and 18. After weaning, the rats were fed with HFD until 16 weeks of age.

Model III: Induction of Obesity using HFD Only
After weaning, the rats were fed with HFD until 16 weeks of age.

Animal grouping and treatments
The obese rats were divided into 13 groups and orally administered with *A. melegueta* extract and orlistat daily for 45 days as shown in Table 1.

Evaluation of obesity in the experimental animals
The obesity status in the experimental rats were determined before and after oral administration of *A. melegueta* extract and orlistat. Leptin and insulin concentrations were estimated in the plasma using ELISA Kits as described by Considine *et al*.[12] and Kullin *et al.*[13]

Lee Index (LI) determination in the experimental animals
Each rat was weighed (g) and the length (cm) was measured from the nose-tip to the anus using a tape rule; and LI was calculated as described.[14]

$$\mathrm{LI} = \frac{\sqrt[3]{\mathrm{Wt(g)}}}{\mathrm{L(cm)}}$$

where Wt(g) is the weight of animal in gram and L is the length of animal in cm. Rats with LI ≥ 0.3 were considered as obese.

Collection and preparation of blood pplasma
Blood plasma was prepared according to the method described by Bode and Oyedapo.[15] The blood samples were collected into anticoagulant (tri-sodium citrate 3.8% w/v) and centrifuged on Bench Centrifuge (Model 90-2 Searchtech Instrument England, UK.) at 3000 rpm for 10 min. Supernatant (plasma) was collected into sterile bottles, labeled and stored in freezer at -20^{o}C for biochemical analyses.

Biochemical analyses
Estimation of leptin and insulin concentrations
Leptin and insulin concentrations were estimated in the plasma using ELISA Kits.[12-13]

Lipid pprofile determination
Lipid profile assay was carried out using Randox Diagnostic kits: total cholesterol (TC)[16]; total triglycerides (TG)[17]; high-density lipoprotein cholesterol (HDL-c)[18]; low-density lipoprotein cholesterol (LDL-c) and very low-density lipoprotein cholesterol (LDL-c).[18]

Atherogenic and coronary risk indices
The atherogenic and coronary risk indices were calculated from the expression.[19-20]

$$\text{Atherogenic Index} = \frac{\mathrm{LDL-c}}{\mathrm{HDL-c}}$$

$$\text{Coronary Risk Index (CRI)} = \frac{\mathrm{TC}}{\mathrm{HDL-c}}$$

Lipase activity assay
The lipase assay was carried out spectrophotometrically with p-nitrophenyllaurate as substrate.[21] The assay mixture consisted of *p*-nitrophenyllaurate (0.7 ml) and plasma (0.3 ml) inside a cuvette. The solution was mixed and changes in absorbance at 410 nm was monitored over a period of 3 min at 15 secs interval. One unit (U) of lipase activity was defined as the amount of enzyme that liberated 1 mmol/ml equivalent of *p*-nitrophenol per min under assay conditions. Lipase activity was calculated as shown:

$$\text{Activity} = \frac{\text{Change in absorbance X Total assay volume}}{€ \text{ X } V_E \text{ (Enzyme volume)}}$$

Where € = Molar extinction coefficient of *p*-nitrophenol at pH 8 = 15,000 $M^{-1}cm^{-1}$

Table 1: Grouping and Treatment of Experimental Animals

	Group	Treatment
Model I	I	Control received distilled water
	II	MSG only
	III	MSG + 50 mg orlistat/kg bwt
	IV	MSG + 200mg extract/kg bwt
	V	MSG + 400 mg extract/kg bwt
Model II	VI	MSG + HFD (High Fat Diet) only
	VII	MSG +HFD + 50 mg orlistat/kg bwt
	VIII	MSG + HFD + 200 mg extract/kg bwt
	IX	MSG + HFD + 400 mg extract/kg bwt
Model III	X	HFD (High Fat Diet) only
	XI	HFD + 50 mg orlistat/kg bwt
	XII	HFD + 200 mg extract/kg bwt
	XIII	HFD + 400 mg extract/kg bwt

Statistical analysis
Data were expressed as Mean ± SEM. Differences between the mean values of the control and treated groups were determined by one-way analysis of variance with a Dunnett Post Hoc test using the Graph Pad Prism 5. Significant difference was considered if $p < 0.05$.

Results and Discussion

Effects of A. melegueta on body weight and Lee index of MSG, MSG + HFD and HFD induced obesity
The present study investigated the effects of *A. melegueta* extract on high fat diet-induced obesity in Wistar rat model with a view to using the plant for prevention, management and treatment of obesity. The

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results recorded a significant increase in percentage weight gain in MSG only; MSG + HFD and HFD only as compared to the normal diet-fed group; highest percentage weight increase was observed in MSG-HFD treated group (Table 2). Previous studies had shown that destruction of arcuate nucleus in hypothalamus indirectly caused an increase in adiposity and body mass measured in rats and humans, and facilitated leptin resistance.[22] Also studies also showed that rats treated with MSG had a significant increase in body weight leading to obesity.[23] Meanwhile, the mechanism by which MSG causes obesity was through the alteration in the regulation of fatty acid metabolism.[24] That is, continuous ingestion of HFD produces positive energy imbalance and a significant increase in body weight gain that leads to obesity. Previous studies demonstrated that fat-rich diet induces obesity by increasing energy intake. Fat is the most energy dense nutrient; therefore, further addition of fat to food causes an increase in calories and energy density.[25] In the three models of obesity tested, treatment with *A. melegueta* aqueous extract for 45 days caused marked reduction in the percentage body weight gain at 200 and 400 mg/kg body weight as compared with the control groups. The study observed that the highest body weight decrease was recorded in MSG-treated group. The reduction in weight gain following the administration of *A. melegueta* extract, might be due to the presence of saponins and flavonoids which have been reported to possess appetite-suppressing property. After these phytochemicals are absorbed in the gastrointestinal tract, they can cross the blood brain barrier into the brain, to amplify the signaling in basal hypothalamus that regulate the energy sensing function.[26] In addition, flavonoids also activate β-adrenergic receptors for fat oxidation.[27] The Lee index is a fast, simple and accurate way to evaluate obesity in rats.[14] It involves dividing the cube root of the weight in grams and nasoanal length in centimeter.[14] Values <0.30 are considered normal while values >0.30 are taken as index of obesity.

Table 2: Effects of *A. melegueta* on Body Weight and Lee Index of MSG, MSG + HFD and HFD induced Obesity

Group	Initial Body Weight (g)	Final Body Weigh (g)	% Change in Weight	Initial Lee Index	Final Lee Index
Control	163.80 ± 1.66	179.20 ± 7.70	9.40	0.28 ± 0.001	0.28 ± 0.002
MSG only	219.40 ± 5.80[a]	250.80 ± 9.10[a]	14.31	0.32 ± 0.001[a]	0.33 ± 0.001[a]
MSG + orlistat 50 (mg/kg bwt)	210.80 ± 7.98[a]	213.30 ± 2.84[b]	1.19	0.33 ± 0.001[a]	0.29 ± 0.001
MSG + AM 200 (mg/kg bwt)	215.80 ± 9.99[a]	227.00 ± 2.55[b]	5.19	0.32 ± 0.001[a]	0.29 ± 0.001
MSG + AM 400 (mg/kg bwt)	215.00 ± 8.63[a]	225.30 ± 3.79[b]	4.79	0.32 ± 0.001[a]	0.28 ± 0.001
MSG + HFD only	187.30 ± 1.93	274.80 ± 2.28[a]	46.72	0.34 ± 0.001[a]	0.35 ± 0.001[a]
MSG + HFD + orlistat 50 (mg/kg bwt)	184.00 ± 2.48	199.00 ± 2.88[b]	8.15	0.34 ± 0.001[a]	0.29 ± 0.001
MSG + HFD + AM 200 (mg/kg bwt)	176.30 ± 3.66	216.70 ± 1.20[b]	22.91	0.34 ± 0.001[a]	0.29 ± 0.001
MSG + HFD + AM 400 (mg/kg bwt)	180.30 ± 4.42	204.30 ± 9.11[b]	13.31	0.32 ± 0.001[a]	0.29 ± 0.001
HFD only	162.80 ± 2.21	192.00 ± 3.06	17.94	0.32 ± 0.001[a]	0.32 ± 0.001[a]
HFD+ orlistat 50 (mg/kg bwt)	164.30 ± 0.63	168.00 ± 2.31[b]	2.25	0.32 ± 0.001[a]	0.29 ± 0.001
HFD+ 200 AM (mg/kg bwt)	166.60 ± 1.33	186.70 ± 1.33	12.06	0.32 ± 0.001[a]	0.29 ± 0.001
HFD+ 400 AM (mg/kg bwt)	160.40 ± 1.66	167.30 ± 6.45[b]	4.30	0.32 ± 0.001[a]	0.29 ± 0.001

Each value represents Mean ± SEM, (n = 5). Value was significant if $p < 0.05$. The values across column with superscript ([a]) implied significant difference from control while ([b]) implied significant difference from MSG, MSG + HFD and HFD groups.
MSG:– Monosodium glutamate, HFD: High Fat Diet, AM: *A. melegueta*

Table 3: Effects of *A. melegueta* on blood glucose level in obese rats

	Initial Blood Glucose (mg/dL)	Final Blood Glucose (mg/dL)	% Change
Control	70.50 ± 4.50	62.67 ± 1.67	11.11
MSG only	97.67 ± 3.93[a]	96.00 ± 1.00[a]	1.71
MSG + orlistat 50 (mg/kg bwt)	92.67 ± 2.20[a]	83.00 ± 1.20[b]	10.51
MSG + AM 200 (mg/kg bwt)	94.67 ± 2.91[a]	89.00 ± 1.00	5.99
MSG + AM 400 (mg/kg bwt)	96.33 ± 4.06[a]	85.67 ± 1.76[b]	11.07
MSG + HFD only	94.00 ± 1.00[a]	96.67 ± 0.88[a]	-2.76
MSG + HFD+ orlistat 50 (mg/kg bwt)	94.50 ± 1.50[a]	94.33 ± 3.71	0.18
MSG + HFD+ AM 200 (mg/kg bwt)	90.00 ± 4.00[a]	89.67 ± 6.89	0.37
MSG + HFD+ AM 400 (mg/kg bwt)	87.00 ± 2.00[a]	89.33 ± 3.71	-2.68
HFD only	92.50 ± 2.50[a]	96.67 ± 1.20[a]	-4.51
HFD+ orlistat	101.00 ± 1.00[a]	93.00 ± 3.22	7.92
HFD+ AM 200 (mg/kg bwt)	93.67 ± 1.45[a]	76.67 ± 1.20[b]	18.15
HFD+ AM 400 (mg/kg bwt)	93.00 ± 1.00[a]	73.67 ± 2.91[b]	20.78

Each value represents Mean ± SEM, (n = 5). Value was significant if $p < 0.05$. The values across column with superscript ([a]) implied significant difference from control while ([b]) implied significant difference from MSG, MSG + HFD and HFD groups.
MSG: Monosodium glutamate, HFD: High Fat Diet, AM: *A. melegueta*

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It has been established that injections of monosodium glutamate to new born rats caused lesion in the ventromedial region of the hypothalamus, leading to obesity due to loss of control in absorption and expenditure of energy.[28] Also destruction of hypothalamic ventromedial nucleus and arcuate nucleus in rat neonates with monosodium glutamate caused an increase in the Lee index.[29] The increase in Lee index was attributed to high fat absorption in the ingested diets.[30-31] The results from this study revealed that at 16 weeks of age, rats administered with MSG only, MSG +HFD only and HFD only became obese as Lee index > 0.30 were observed (Table 2).

Effects of A. melegueta and Orlistat on plasma lipid profiles of obese rats

The hallmark of dyslipidemia in obesity is characterized by post-prandial triglycerides (TG) accumulation, high level of low-density lipoprotein (LDL) and low high-density lipoprotein (HDL-c). Hypertriglyceridemia could be the major cause of other lipid abnormalities since it leads to delayed clearance of TG-rich lipoproteins and formation of small dense LDL-c.[32] In this study, dyslipidemia was significantly apparent across all the induced groups compared with control. In the absence of increased exogenous dietary lipid, MSG significantly elevated plasma and hepatic TG, plasma total cholesterol (TC), LDL and very low-density lipoprotein (VLDL-c). Similar results were obtained in HFD-only group however; the largest effect on lipid homeostasis was observed in MSG + HFD group; suggesting that MSG potentiated the effects of HFD (Table 4).

Lipid that has been absorbed via GIT is converted into free fatty acids and glycerol and re-synthesized as triglycerides. The occurrence of hypertriglyceridemia, hypercholestrolemia was probably due to the increase in hepatic VLDL-c synthesis induced by hyperinsulinemia. The decreasing level of HDL-c in obesity may be a result of an increased triglyceride load in the HDL-c particle that is acted upon by hepatic lipase, which hydrolyzes the triglycerides. The reduction of the level of triglyceride results in the generation of a small HDL-c particle that is filtered by the kidney, resulting in a decrease in apolipoprotein A and HDL-c concentrations.[33]

The reduction in total cholesterol by *A. melegueta* may be due to the presence of phytoactive constituents capable of inhibiting the activity of 3-hydroxy-3-methyl-glutaryl-coenzyme A reductase (HMG-CoA reductase) an enzyme that plays prominent role in cholesterol biosynthesis[38] by catalyzing the conversion of HMG-CoA to mevalonic acid.[39] Studies have shown that flavonoids are able to improve the lipid profile in normal rats[40] and also inhibit the activity of HMG-CoA reductase[39, 41].

In diet-induced obesity, tea flavonoids increased fecal excretion of cholesterol, bile acid and reduced cholesterol absorption from intestine.[42] Increase in HDL cholesterol is associated with decreases in cardiovascular risks. Drugs that decrease total cholesterol also increase HDL-c.[43] *A. melegueta* extract significantly improved HDL-c level in blood plasma when compared with the obese control; which has potential advantage in hypercholesterolemia treatment.[44] *A. melegueta* antioxidant constituents (flavonoids) were shown to cause a decrease in oxysterol levels in bile acid metabolism and blocked lipid absorption into the liver.[45-46]

Effects of A. melegueta Extract and Orlistat on Atherogenic and Plasma Indices of Obese Rats

Atherogenic index of plasma (AIP) is a novel index composed of TG and HDL-c. It has been employed to quantify blood lipid levels and commonly used as optimal indicator of dyslipidemia and associated diseases.[20, 34-35] The atherogenic and coronary heart indices were found to increase significantly in all the obese rats and most apparent in MSG+HFD group when compared with the control (Figures 1-6). The atherogenic and coronary artery risk indices are indicators of cholesterol deposition in rat tissues.[36-37] These parameters were significantly decreased when treated with *A. melegueta* extract; suggesting that the plant extract possessed hypolipidemic property.

Effects of A. melegueta and Orlistat on Plasma Insulin, Leptin Levels

Adipose tissue plays a major role in fat storage. Accumulation of fat in adipocytes is the underlying phenomenon in obesity. Adipose tissue secretes leptin which plays key roles in energy regulation.[47-48] Leptin secretion is in direct proportion to the level of triglyceride stored in adipocytes.[49] Obesity induces hyperleptinemia and hyperinsulinemia which are correlated with body weight and visceral fat. Reduction in body weight is possible by means of decrease in plasma leptin levels.[50] In this study, significant increase in plasma leptin level was observed in all the obese groups as compared with the control (Table 5). In this study, obese rats developed a hyperglycemic state associated with hyperinsulinemia and high blood glucose level when compared with the control (Table 3) (Table 5). Similar results were found in other studies.[51-52] Treatment with *A. melegueta* extract caused a reduction in the leptin levels in two models tested but not in MSG+HFD group. Obesity is accompanied by decreased sensitivity of adipocytes to insulin. Long term exposure to MSG in experimental animals lead to hyperlipidemia, hyperglycemia and insulin resistance.[53]

Table 4: Effects of *A. melegueta* Extract and Orlistat on Plasma Lipid Profiles of Obese Rats

	TRG (mg/dL)	TC (mg/dL)	HDL-c (mg/dL)	LDL-c (mg/dL)	VLDL-c (mg/dL)
Control	58.58 ± 4.10	27.37 ± 0.57	7.61 ± 0.35	8.04 ± 0.71	11.72 ± 0.82
MSG only	81.67 ± 6.50[a]	42.62 ± 2.93[a]	4.73 ± 0.21[a]	21.56 ± 2.31[a]	16.33 ± 1.30[a]
MSG + orlistat 50 (mg/kg bwt)	40.29 ± 4.00[b]	27.26 ± 3.03[b]	6.87 ± 0.95	12.33 ± 2.07[b]	8.06 ± 0.80[b]
MSG + AM 200 (mg/kg bwt)	45.15 ± 5.14[b]	30.94 ± 5.97	4.96 ± 0.20	16.95 ± 3.36	9.03 ± 1.03[b]
MSG+AM 400 (mg/kg bwt)	37.40 ± 2.60[b]	23.36 ± 1.37[b]	7.91 ± 0.81[b]	7.97 ± 1.02[b]	7.48 ± 0.52[b]
MSG + HFD only	109.80 ±11.61[a]	47.78 ± 2.64[a]	5.17 ± 0.28[a]	20.65 ± 2.50[a]	21.97 ± 2.32[a]
MSG + HFD+ orlistat 50 (mg/kg bwt)	101.7 ± 10.94	41.36 ± 1.87[a]	5.71 ± 0.28	15.31 ± 1.82	20.34 ± 2.19
MSG + HFD + AM 200 (mg/kg bwt)	90.22 ± 5.62	31.68 ± 1.76[b]	5.58 ± 0.85	8.05 ± 1.64[b]	18.04 ± 1.13
MSG+ HFD + AM 400 (mg/kg bwt)	82.08 ± 9.97	36.70 ± 2.02[b]	5.55 ± 0.10	14.74 ± 3.14	16.42 ± 1.99
HFD only	94.08 ± 7.22[a]	89.19 ± 7.63[a]	4.17 ± 0.18[a]	66.21 ±7.32[a]	18.82 ± 1.45[a]
HFD + Orlistat 50 (mg/kg bwt)	54.55 ± 8.26[b]	30.13 ± 3.44[b]	7.03 ± 0.92[b]	12.19 ± 4.28[b]	10.91 ± 1.65[b]
HFD + AM 200 (mg/kg bwt)	67.20 ± 3.47	32.68 ± 6.15[b]	5.50 ± 0.17	13.75 ± 5.72[b]	13.44 ± 0.69
HFD + AM 400 (mg/kg bwt)	48.81 ± 4.20[b]	22.81 ± 1.19[b]	6.39 ± 0.31[b]	6.66 ± 1.81[b]	9.763 ± 0.84[b]

Each value represents Mean ± SEM, n = 5 readings. Value of $p < 0.05$ was considered significant. The values across column with superscript (a) implied significant difference from control while (b) implied significant difference from MSG, MSG+HFD and HFD groups. TRG – Triglycerides, TC- Total cholesterol, HDL-c – High-density lipoprotein cholesterol, LDL-c – Low-density lipoprotein cholesterol, VLDL-c – Very low-density lipoprotein cholesterol, MSG- Monosodium glutamate, HFD- High fat diet, AM- *A. melegueta*

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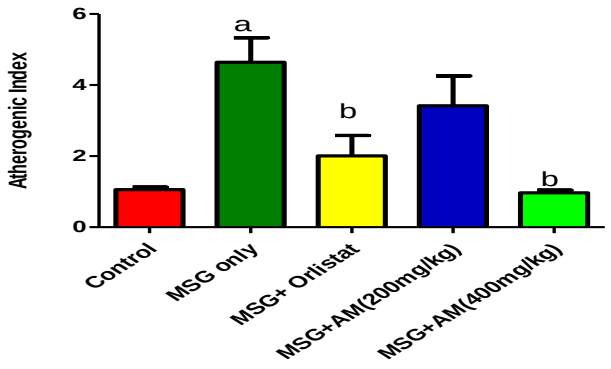

Figure 1: Effects of *A. melegueta* Extract and Orlistat on the Atherogenic Index of MSG-induced Obesity.
Bar with alphabet a is significantly different from control and b is significantly different from MSG only

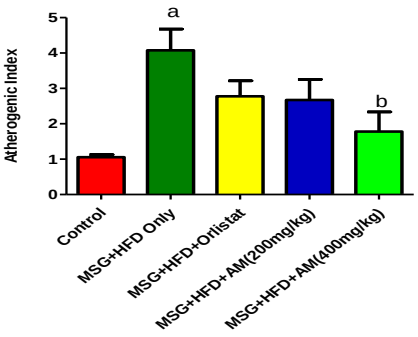

Figure 2: Effects of *A. melegueta* Extract and Orlistat on the Atherogenic Index of MSG + HFD-induced Obesity.
Bar with alphabet a is significantly different from control and b is significantly different from MSG + HFD only.

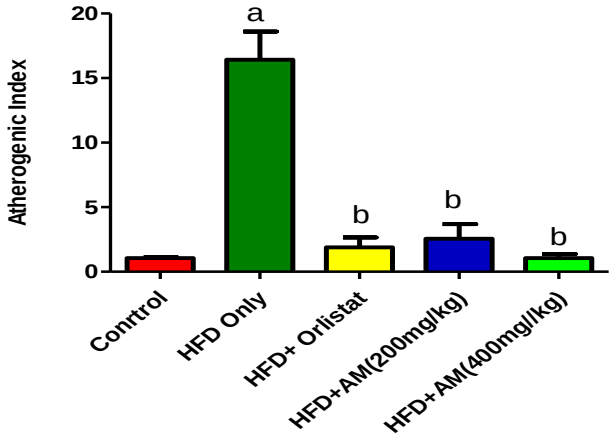

Figure 3: Effects of *A. melegueta* extract and orlistat on the atherogenic index of HFD-induced obesity.
Bar with alphabet a is significantly different from control and b is significantly different from MSG + HFD only.

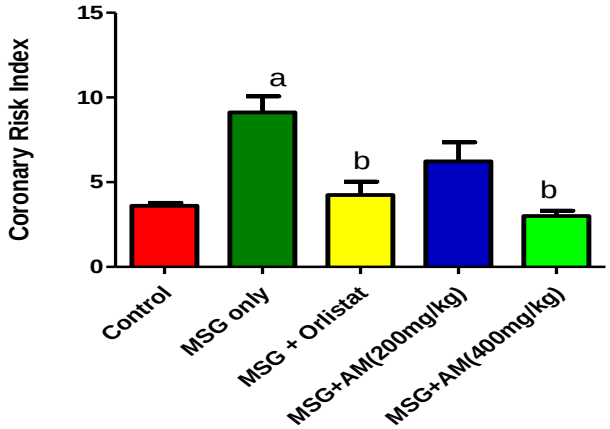

Figure 4: Effects of *A. melegueta* Extract and Orlistat on the Coronary Risk Index of MSG-induced Obesity.
Bar with alphabet a is significantly different from control and b is significantly different from MSG only.

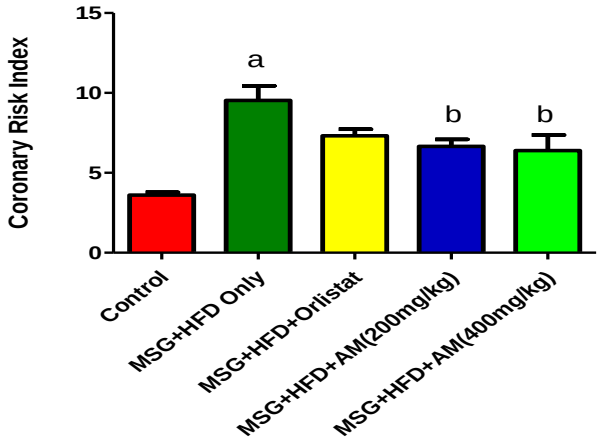

Figure 5: Effects of *A. melegueta* extract and orlistat on the coronary risk index of MSG+ HFD-induced obesity.
Bar with alphabet a is significantly different from control and b is significantly different from MSG + HFD only.

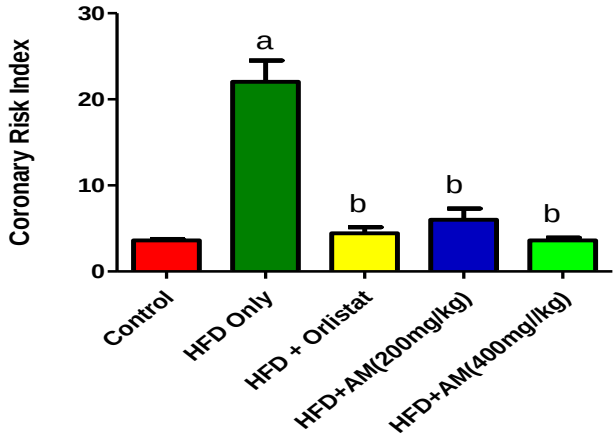

Figure 6: Effects of *A. melegueta* extract and orlistat on the coronary risk index of HFD-induced obesity.
Bar with alphabet a is significantly different from control and b is significantly different from HFD only.

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Table 5: Effects of *A. melegueta* Extract and Orlistat on Plasma Insulin and Leptin Concentrations of Obese Rats

	Initial Insulin Concentration (mU/L)	Final Insulin Concentration (mU/L)	Initial Leptin Concentration (µg/L)	Final Leptin Concentration (µg/L)
Control	1.55 ± 0.22	2.05 ± 0.25	2.22 ± 0.06	1.72 ± 0.30
MSG only	2.87 ± 0.39[a]	2.57 ± 0.02	4.74 ± 0.14[a]	5.96 ± 0.07[a]
MSG + orlistat 50 mg/kg bwt	3.26 ± 0.12[a]	2.42 ± 0.17	4.36 ± 0.33[a]	3.24 ± 0.36[b]
MSG + AM 200 mg/kg bwt	2.75 ± 0.33[a]	2.53 ± 0.05	4.85 ± 0.76[a]	5.82 ± 0.01
MSG + AM 400 mg/kg bwt	2.70 ± 0.06[a]	2.44 ± 0.11	4.56 ± 0.73[a]	2.69 ± 0.27[b]
MSG + HFD only	2.97 ± 0.04[a]	4.23 ± 0.38[a]	4.53 ± 0.14[a]	7.09 ± 0.07[a]
MSG + HFD+ orlistat 50 (mg/kg bwt)	3.36 ± 0.15[a]	3.88 ± 0.20	4.78 ± 0.65[a]	6.84 ± 0.03[b]
MSG + HFD + AM 200 (mg/kg bwt)	3.30 ± 0.59[a]	4.22 ± 0.40	5.08 ± 0.70[a]	7.04 ± 0.05
MSG + HFD + AM 400 (mg/kg bwt)	3.61 ± 0.29[a]	3.79 ± 0.43	4.94 ± 0.65[a]	7.00 ± 0.05
HFD only	3.13 ± 0.41[a]	3.31 ± 0.02[a]	5.04 ± 0.53[a]	6.36 ± 1.15[a]
HFD + Orlistat (50 mg/kg bwt)	3.26 ± 0.25[a]	2.55 ± 0.27[b]	5.04 ± 0.05[a]	2.18 ± 0.03[b]
HFD + AM 200 (mg/kg bwt)	3.43 ± 0.21[a]	2.64 ± 0.17[b]	5.01 ± 0.02[a]	1.94 ± 0.03[b]
HFD + AM 400 (mg/kg bwt)	3.13 ± 0.26[a]	2.71 ± 0.09[b]	5.55 ± 0.40[a]	1.84 ± 0.18[b]

Each value represented Mean ± SEM, n = 5 readings. Value of $p < 0.05$ was considered significant. The values across column with superscript (a) implied significant difference from control while (b) implied significant difference from MSG, MSG+HFD and HFD groups. MSG- Monosodium glutamate, HFD – High Fat Diet, AM- *A. melegueta*

Table 6: Effects of *A. melegueta* Extract and Orlistat on Plasma Lipase Activity of Obese Rats

Group	Lipase Activity (mU/L)	Activity (%)	% Inhibition
Control	0.50 ± 0.02	100	0
MSG only	0.44 ± 0.01	88	12
MSG + orlistat 50 (mg/kg bwt)	0.42 ± 0.01	84	16
MSG + AM 200 (mg/kg bwt)	0.48 ± 0.01	96	04
MSG +AM 400 (mg/kg bwt)	0.48 ± 0.04	96	04
MSG + HFD only	0.46 ± 0.03	92	08
MSG + HFD+ orlistat 50 (mg/kg bwt)	0.43 ± 0.01	86	14
MSG + HFD+ AM 200 (mg/kg bwt)	0.47 ± 0.01	94	06
MSG + HFD + AM 400 (mg/kg bwt)	0.47 ± 0.02	94	06
HFD only	0.45 ± 0.02	90	10
HFD + orlistat 50 (mg/kg bwt)	0.37 ± 0.07[b]	74	26
HFD + AM 200 (mg/kg bwt)	0.47 ± 0.04	94	06
HFD + AM 400 (mg/kg bwt)	0.50 ± 0.06	100	-

Each value represented Mean ± SEM, n = 5 readings. Value of $p < 0.05$ was considered significant. The values across column with superscript (a) implied significant difference from control while (b) in each model implied significant difference from MSG, MSG + HFD, HFD-induced obese rats respectively. MSG- Monosodium glutamate, HFD- High Fat Diet, AM- *A. melegueta*

In MSG-induced obesity, treatment with 400 mg/kg bwt extract caused a slight reduction in blood glucose level and insulin concentration. Administration of extract to MSG+ HFD treated group produced a nonsignificant decrease in insulin and blood glucose levels. However, *A. melegueta* caused a significant decrease in blood glucose level and improved insulin sensitivity in HFD rats. The ability of the extract to reduce insulin and blood glucose levels could be through the interference with the energy metabolism of the cells.[43]

Effects of A. melegueta and Orlistat on Lipase Activity

A different action mechanism was observed on the lipase activity, where the extract and orlistat groups exerted opposite effects. Induction of obesity caused a decrease in lipase activity due to body weight gain and energy storage. Orlistat is known to exert its effect against obesity and obesity-induced inflammation through blocking of fat absorption and lowering the triglycerides.[54] *A. melegueta* was able to enhance lipase activity showing that it exerted its effects on cellular fat storage by inducing lipolysis and elevating body energy metabolism. In contrast, orlistat blocks fat absorption into cells, thereby preventing fat storage.

In the MSG+HFD model, treatment with the extract significantly increased the activity of SOD. There was no significant change in the activities of the enzymes when treated with orlistat. In the HFD model, administration of extract and orlistat significantly increased the activities of the enzymes and reduced glutathione.

Conclusion

Administration of *A. melegueta* extract corrected the level of circulating lipids; reduced insulin and leptin levels, increased lipase activity and restored the body weight of the obese rats in a dose-dependent especially in HFD and MSG-induced obesity. These effects could be due to the presence of thermogenic and lipid-modulating compounds in *A. melegueta*. Also the positive effects of *A. melegueta* on atherogenic indices (predictors of coronary heart disease) and obesity-induced oxidative stress were strong indicators that the plant possessed robust anti-obesity property; and justified its use in traditional medicine.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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References

1. Mohammed BS. Absence of an effect of liposuction on insulin action and risk factors for coronary heart disease. New Engl J Med. 2015; 350:2549–2557.
2. Steven BH and Thomas AW. Mechanisms, pathophysiology, and management of obesity. New Engl J Med. 2017; 376:254-266.
3. Hill JO, Wyatt HR, Peters JC. Energy balance and obesity. Circul. 2012; 126(1):126-132.
4. Yanovski SZ and Yanovski JA. Long-term drug treatment for obesity: A systematic and clinical review. J Am Med Assoc. 2014; 311(1):74-86.
5. Sugita J, Yoneshiro T, Hatano T, Aita S, Ikemoto T, Uchiwa H, Iwanaga T, Kameya T, Kawai Y, Saito M. Grains of paradise (*Aframomum melegueta*) extract activates brown adipose tissue and increases whole-body energy expenditure in men. Br J Nutr. 2013; 110(4):733-738.
6. Doherty VF, Olaniran OO, Kanife UC. Antimicrobial activities of *Aframomum melegueta.* Int J Biol. 2010; 2:126-131.
7. Oladunmoye MK and Kehinde FY. Ethnobotanical survey of medicinal plants used in treating viral infections among Yoruba tribe of South Western Nigeria. Afr J Microbiol Res. 2011; 5:2991-3004.
8. Kokou I, Damintoti KS, Amegnona A, Yao A, Messanvi G. Effect of *Aframomum melegueta* on carbon tetrachloride induced liver injury. J Appl Pharm Sci. 2013; 3(9):98-102.
9. Oyedapo OO and Amos S. Further investigation into the activities of the root extract of *Plumbago zaylanica*. Phytother Res. 1997; 11:62-63.
10. Campos-Sep´ulveda AE, Ayala-Guerrero F, Rubio-Poo C, Rom´an-Ramos R. Neonatal monosodium glutamate increases ethanol susceptibility effect in adult mice. In: Proc Western Pharmacol Soc. 2002; 45:44–46.
11. Penka A, Nikolay B, Katerina G. Effects of the application of a high-fat-carbohydrate diet without additional cholesterol for the inducement of metabolic syndrome in rats. Am Int J Contemp Res. 2013; 3(9):1-9.
12. Considine RV, Sinha MK, Heiman ML, Kriaciunas A, Stephens TW, Nyce MR. Serum immunoreactive leptin concentration in normal-weight and obese humans. New Engl J Med. 1996; 334:292-295.
13. Kullin M, Li Z, Bondo HJ, Welsh N, Karlsson FA, Sandler S. Protection of rat pancreatic islets by potassium channel openers against alloxan, sodium nitroprusside and interleukin-1beta mediated suppression--possible involvement of the mitochondrial membrane potential. Diabetol. 2003; 46:80-88.
14. Lobato NS, Filgueira FP, Akamine EH, Davel AP, Rossoni, LV, Tostes RC. Obesity induced by neonatal treatment with monosodium glutamate impairs microvascular reactivity in adult rats: role of NO and prostanoids. Nutr Metabol Cardiovasc Dis. 2011; 21(10):808–16.
15. Bode SO and Oyedapo OO. Biological activities and phytoconstituents of the lower plant *Platycerium anglolense*, Welwex Hook. J Med Plants Res. 2011; 5:1321-1329.
16. Richmond N. Preparation and properties of a cholesterol oxidase from Nocadia sp. and its application to the enzymatic assay of total cholesterol in serum. J Chem. 1973; 1(9):135.
17. Tietz NW. Colorimetric method of triglyceride estimation. In Clinical guide to laboratory tests. 2nd ed. WB Saunders Company, Philadelphia, USA. 1990; 554-556p.
18. Afolabi IS, Akuiyibo SM, Rotimi SO, Adeyemi AO. *In vivo* evaluation of lipid and antioxidants qualities of *Carica papaya* seed oil. J Nat Prod. 2011; 4:125-135.
19. Ademuyiwa O, Ugbaja RN, Rotimi, SO. Plasma lipid profile, atherogenic and coronary risk indices in some residents of Abeokuta in south-western Nigeria. Biokem. 2008; 20(2):85-91.
20. Zhu X, Yu L, Zhon H, Ma Q, Zhou X, Lei T. Artherogenic index of plasma is a novel and better biomarker associated with obesity: a population-based cross-sectional study in China. Lipids in Healths Dis. 2018; 17:37-42.
21. Vorderwiilbecke T, Kieslich K, Erdmann H. Comparison of lipases by different assays. Enzyme Microb Tech. 1992; 14:631-639.
22. Pepino MY, Finkbeiner S, Beauchamp GK, Mennella JA. Obese women have lower monosodium glutamate taste sensitivity and prefer higher concentrations than do normal-weight women. Obes. 18:959-965.
23. Oluba OM, Onyeneke EC, Idonije BO, Eidangbe GO. Effect of soy protein on monosodium glutamate (msg)-induced obesity in rats. Asian J Pharm Biol Res. 2011; 1(1):8-14.
24. Insawang T, Selmi C, Cha'on U, et al. Monosodium glutamate (MSG) intake is associated with the prevalence of metabolic syndrome in a rural Thai population. Nutr Metab (Lond). 2012; 9(1):50.
25. Kretschmer BD, Schelling P, Beier N, Liebscher C, Treutel S, Krüger N. Modulatory role of food, feeding regime and physical exercise on body weight and insulin resistance. Life Sci. 2004; 76(14):1553-1573.
26. Chidrawar VR, Patel KN, Bothra SB, Shiromwar SS, Koli AR, Kalyankar GG. Anti-obesity effect of *Stellaria media* methanolic extract in the murine model of cafeteria diet induced obesity. Internat J Nutr Pharmacol Neurol Dis. 2012; 2:121-31.
27. Ohkoshi E, Miyazaki H, Shindo K, Watanabe H, Yoshida A, Yajima H. Constituents from the leaves of *Nelumbo nucifera* stimulate lipolysis in the white adipose tissue of mice. Planta Med. 2007; 73:1255-1259.
28. Dolnikoff M, Martin–Hidalgo A, Machado UF, Lima FB, Herrera E. Decreased lipolysis and enhanced glycerol and glucose utilization by adipose tissue prior to development of obesity in Monosodium glutamate (MSG) treated rats. Int J Obes Related Metabol Disord. 2001; 25:426-433.
29. Nakagawa T, Ukai K, Ohyama T, Gomita Y, Okamura H. Effects of chronic administration of sibutramine on body weight, food intake and motor activity in neonatally monosodium glutamate-treated obese female rats: relationship of antiobesity effect with monoamines. Experimental Animals. 2000; 49:239-249.
30. Rotimi OA, Olayiwola IO, Ademuyiwa O, Balogun AE. Effects of fibre-enriched diets on tissue lipid profiles of MSG obese rats. Food Chem Toxicol. 2012; 50:4062–4067.
31. Fernandes G, Arena A, Campos K, Volpato G, Anselmo-Franci J, Damasceno D, Kempinas W. Glutamate-induced obesity leads to decreased sperm reserves and acceleration of transit time in the epididymis of adult male rats. Reprod Biol Endocrinol. 2012; 10:105-110.
32. Klop B, Elte JWF, Cabezas MC. Dyslipidemia in obesity: mechanisms and potential targets. Nutr. 2013; 5:1218–1240.
33. Hamdy AM and Khaled AA. Modulatory effect of green tea on lipid metabolism and brain neurotransmitters of obese mice model. Clujul Med. 2012; 85:347-352.
34. Boran k, Pathak NS, Borah P, Hussain MI, Das D. Association of apolipoprotein A- I gene polymorphisms

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with cardiovascular disease risk factors and atherogenic indices in patient patients from Assam, Northeast India, Balikan. J Med Genet. 2017; 20:59-70.
35. Yang SH, Du Y, Li XL, Zhang Y, Li S, Xu RX. Triglyceride to high-density lipoprotein cholesterol ratio and cardiovascular events in diabetics with coronary artery disease. Am J Med Sci. 2017; 354:17-24.
36. Murray MT and Pizzorno J. Cholesterol. In: Encyclopedia of Nat Med 2nd ed. 1998; 347–400.
37. Lee CL, Wen JY, Hsu YW, Pan TM. Monascus-fermented yellow pigments monascin and ankaflavin showed antiobesity effect via the suppression of differentiation and lipogenesis in obese rats fed a high-fat diet. J Agric Food Chem. 2013; 61:1493-1500.
38. Reddy PV and Urooj A. Inhibition of 3-hydroxy-3-methylglutaryl coenzyme A reductase (*ex vivo*) by *Morus indica* (Mulberry). Chines J Biol. 2014; 1-5.
39. Rang HP, Dale MM, Ritter JM, Moore RK. Pharmacology 6th ed Edinburgh: Churchill livingstone; 2007; 333p.
40. Ito Y, Ichikawa T, Morohoshi Y, Nakamura T, Saegusa Y, Ishihara K. Effect of tea catechins on body fat accumulation in rats fed a normal diet. Biomed Res. 2008; 29:27-32.
41. Min SW and Kim DH. Kakkalide and irisolidone: HMG-CoA reductase inhibitors isolated from the flower of *Pueraria thunbergiana*. Biol Pharm Bull. 2007; 10:1965-1968.
42. Espinosa J, Medeiros LF, Souza A, Güntzel ARC, Rücker B, Casali EA, Ethur E, Wink MR, Torres IL. Ethanolic extract of *Casearia sylvestris Sw* exhibits *in vitro* antioxidant and antimicrobial activities and *in vivo* hypolipidemic effect in rats. Revista Brasileira de Plant Med. 2015; 17:305-315.
43. Jung UJ and Cho M. Obesity and its metabolic complications: the role of adipokines and the relationship between obesity, inflammation, insulin resistance, dyslipidemia and nonalcoholic fatty liver disease. Int J Mol Sci. 2014; 15:6184-6223.
44. Asdaq SMB. Antioxidant and hypolipidemic potential of aged garlic extract and its constituent, s-allyl cysteine, in rats. Evid-Based Compl Altern Med. 2015; 1:1-7.
45. Lotito SB and Frei B. Dietary flavonoids attenuate tumor necrosis factor alpha-induced adhesion molecule expression in human aortic endothelial cells. Structure-function relationships and activity after first pass metabolism. J Biol Chem. 2006; 281:37102-37110.
46. Crosignani A, Zuin M, Allocca M, Del Puppo, M. Oxysterols in bile acid metabolism. Clin Chim Acta. 2011; 412:2037-2045.
47. Brennan AM and Mantzoros CS. Drug insight: the role of leptin in human physiology and pathophysiology- emerging clinical applications. Nat Rev Endocrinol. 2006; 2:318-327.
48. Garg A. Adipose tissue dysfunction in obesity and lipodystrophy. Clinical Cornerstone. 2006; 8:S7-S13.
49. Staiger H and Haring HU. Adipocytokines: fat-derived humoral mediators of metabolic homeostasis. Exp Clin Endocrinol Diab. 2005; 113:67-79.
50. Van Dielen FM, Veer C, Buurman WA, Greve JWM. Leptin and soluble leptin receptor levels in obese and weight-losing individuals. J Clin Endocrinol Metabol. 2002; 87:1708-1716.
51. Jang IS, Hwang DY, Chae KR. Role of dietary fat type in the development of adiposity from dietary obesity-susceptible Sprague–Dawley rats. Br J Nutr. 2003; 89:429-437.
52. Jen KL, Buison A, Pellizzon M. Differential effects of fatty acids and exercise on body weight regulation and metabolism in female Wistar rats. Exp Biol Med. 2003; 228:843-849.
53. Lopez MJ, Diaz SM, Salas A. Oleoyl-estrone induces massive loss of body weight in Zucker fa/fa rats fed a high-energy hyperlipidemic diet. J Nutr Biochem. 2000; 11:530–535.
54. Karagozlu M, Kim M, Lee M. Citrus Peel Ethanol Extract Inhibits the Adipogenesis Caused from High Fat-Induced DIO Model. Food Nutr Sci 2016; 7:8-19.