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Hypolipidemic potentials of methanol extracts of *Vernonia colorata*

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ABSTRACT: This study was designed to evaluate the effect of concomitant feeding of high fat diet (HFD) and administration of methanol extracts of *Vernonia colorata* (MEVC) on lipid profile and body weight changes in Wistar albino rats. Thirty male rats aged between 10-12 weeks were used for this study. Animals received food and water *ad libitum*. Graded doses of the extract were dissolved in Dimethyl sulphoxide (DMSO) and administered orally on a daily basis. Body weight was measured weekly while plasma lipid components were measured at the end of the study which lasted for 10 weeks. The findings of this study revealed that concomitant feeding of high fat diet and administration of methanol extracts of *Vernonia colorata* resulted in significantly *($p<0.05$)* lower plasma triacylglycerol, cholesterol and LDL-cholesterol but higher HDL-cholesterol concentrations when compared to the high fat diet only and high fat diet + DMSO groups. The lipid lowering effects of methanol extracts of *Vernonia colorata* was similar to Orlistat. Administration of *Vernonia colorata* also resulted in dose-dependent decrease of 22.2% and 15.8% in body weight gain relative to 12.5% decrease in the Orlistat group. The findings of this study provide convincing evidence for the hypolipidemic and anti-obesity potentials of methanol extracts of *Vernonia colorata*. This suggests that the plant may be useful in weight loss regimen, attenuating dietary obesity and also serve as a potential drug lead in the search for natural products for the treatment of diseases associated with dyslipidemia.

Keywords: *Vernonia colorata*, high fat diet, obesity, dyslipidemia

Introduction

Obesity is gradually becoming a disease of global concern as higher values of body mass index (BMI) correlates with increasing prevalence of cardiovascular diseases, hypertension and diabetes.[1] Since genetic predisposition and the consumption of energy rich foods are the commonest pathogenetic factors, many therapies will therefore target to achieve weight reduction through dietary modulation.[1,2] Dyslipidemia is a major risk factor for several non-communicable diseases as it significantly increases the risk of obesity,

atherosclerosis, cardiovascular diseases, cerebrovascular diseases, hypertension and type 2 diabetes mellitus.[3] Dyslipidemia describes a group of metabolic anomalies characterized by any or all of the following: elevated total cholesterol, elevated low-density lipoprotein cholesterol (LDL-c), elevated triacylglycerol and decreased high-density lipoprotein cholesterol (HDL-c).[4,5,6] Expectedly, it contributes significantly to increased risk of atherosclerotic cardiovascular disease in diabetes [7] thereby constituting a significant public health challenge.

The high cost which results in its unaffordability by majority of people in sub Saharan Africa in addition to its associated undesirable side effects constitute a double jeopardy for currently available lipid-lowering synthetic drugs. This has led to the continued dependence on plants with folkloric use in ethnomedicine as alternatives to these synthetic drugs.[8] Moreover, there is a global dependence on medicinal plants in healthcare.[9] This implies that there is an urgent need to evaluate the scientific basis of the efficacy of these plants in certain health conditions. Correspondingly, there has been an upsurge in scientific reports demonstrating positive biological activity, thus providing scientific evidence for their efficacy. This has resulted in a steady increase in the use of plant extracts for the treatment of a wide variety of diseases.[10] Medicinal plants synthesize and store up secondary metabolites such as alkaloids, sterols, terpenes, flavonoids, saponins, glycosides, cyanogenics, tannins, resins, lactones, quinines and volatile oils. These secondary metabolites and other chemical constituents present in medicinal plants account for their medicinal potency and efficacy.[11,12]

Vernonia colorata is a medicinal plant, belonging to the Asteraceae family [13] and *Vernonia* genus. Other common members of that genus include *V. amygdalina* and *V. calvoana*. They are all eaten locally as leafy vegetables. *Vernonia colorata* is a perennial shrub of 3.5-8m high that is found throughout Central and West Africa. *Vernonia colorata* is similar to *Vernonia amygdalina* in appearance and nutrient content, but has broader, wildly hairy leaves and it is less bitter-tasting than *Vernonia amygdalina*.[14] Hence, it can be described as sweet bitter leaf due to its characteristic non-bitter taste.[15] The leaves can be eaten fresh or in semi-processed form and they are used as accompaniment to various indigenous staples or as a spice in food.[16] There are available reports on the effects of various extracts of *Vernonia* species on blood lipids [17] although reports on concomitant feeding of high fat diet and administration of *V. colorata* are not readily available. This present study is therefore aimed at evaluating the possible effects of concomitant feeding of high fat diet and administration of *V. colorata* on lipid profile and body weight changes.

Materials and Methods

Collection and Identification of Plant

Fresh mature leaves of *Vernonia colorata* (Figure 1) were harvested from the farms in Forestry Research Institute, Ahiaeke, Abia State. The leaves were identified and authenticated by Mr. Ibe Ndukwe, a Taxonomist at the Herbarium unit of the Department of Forestry and Environmental Management, Michael Okpara University of Agriculture, Umudike, Abia State. A voucher specimen (FHI 4873-*Vernonia colorata*) was deposited at the herbarium.

Preparation of Methanol Extracts of *Vernonia colorata*

The fresh leaves were plucked, sorted, rinsed in clean tap water and air dried in the absence of sun light at room temperature. The plant was pulverised and stored in air-tight containers. The pulverised *Vernonia colorata* (500g) was packed into a glass column and was saturated with methanol. This was allowed to stand for 48 hours to allow for complete extraction of methanol-soluble phytochemicals. The tap was opened at the end of the extraction process and the extract collected into pre-weighed beakers. The methanol was removed by evaporation to recover the crude extract.

Figure 1: *Vernonia colorata* showing its leaves and fruits.

Standard Drug

The standard drug Orlistat (Xenical Pharmaceuticals, Japan) was obtained from Blessed Pharmacy, 30 Lagos Street, Umuahia, Nigeria. It was reconstituted in distilled water and administered orally at a dose of 5.14 mg/kg body weight which was simulated from the human regimen.[2]

Feed Formulation

The basal diet and high fat diet was prepared with basic feed materials, following standard protocol. The components of the diet is presented in Table 1. The high fat diet was formulated such that 35 % of the total energy in the diet came from fats according to the method of Egedigwe *et al.*[18] All materials used for diet formulation were purchased from reputable vendors in Umuahia, Abia State, Nigeria. The components of the diet were weighed out into a bowl and thoroughly mixed to obtain a dough-like consistency. They were thereafter made into pellets by extrusion through an improvised device. The pellets were oven-dried at 40^{o}C and stored in air-tight containers to avoid rancidity.

Experimental Animals

Thirty male albino rats of the Wistar strain aged between 10-12 weeks obtained from the Animal Breeding Unit of the Faculty of Veterinary Medicine, University of Nigeria, Nsukka were used for this study. The rats were transported to the Animal House of the College of Natural Sciences, Michael Okpara University of Agriculture Umudike, Abia State where the study was carried out under controlled temperature (25-28 ℃) and 12 hour light/dark cycle. The animals were allowed to acclimatize, receiving food and water *ad libitum*. The study protocol followed the guidelines of the National Research Council for the care and use of laboratory animals.[19]

Table 1: Composition of Diets

	Concentration (g/1000 g of feed)	
Item	**High Fat Diet**	**Basal Diet**
Maize	388.80	388.80
Egg yolk powder	58.39	0.00
Groundnut cake	133.93	133.93
Cray fish	21.58	21.58
Vitamin mix	3.97	3.97
Mineral mix	15.89	15.89
Bone meal	19.87	19.87
Cellulose	3.97	3.97
Palm kernel oil	69.53	0.00
Palm oil	69.53	69.53
Corn starch	214.54	214.54

Experimental Design

The rats were randomized into six groups of 5 rats each. The rats in the six groups had similar average body weights at the onset of the study. The six groups received feed and extract as follows:

Group I: Basal Diet
Group II: High Fat Diet + 1000 mg/kg *V. colorata*
Group III: High Fat Diet + 200 mg/kg *V. colorata*
Group IV: High Fat Diet + DMSO
Group V: High Fat Diet + Orlistat (5.14 mg/kg body weight)
Group VI: High Fat Diet only

A portion of the methanol extract of *Vernonia colorata* (10 g) was dissolved in 10 ml of 5 % Dimethyl sulphoxide to give 100 mg/ml and was administered at 1000 and 200 mg/kg body weight. Oral administration of the extract was done daily and the rats were fed the formulated diet for 10 weeks. In order to avoid spoilage, fresh diet was compounded for each group on a weekly basis. Following an overnight fast, the rats were euthanized at the end of 10 weeks of treatment and whole blood was collected by cardiac puncture with sterile needle into heparinized tubes and centrifuged at 5,000 rpm for 10 minutes. Plasma was separated and stored at a temperature of -4 ℃ until required.

Biochemical Analysis

The enzymatic colorimetric endpoint methods were used to determine the plasma concentrations of total cholesterol, triacylglycerols and high density lipoprotein (HDL)-cholesterol.[20,21,22] Low density lipoprotein (LDL)-cholesterol concentration was estimated by difference according to the Friedewald equation.[23]

Body Weight Measurement

Body weight was determined on a weekly basis based on the daily measurements and increases or decreases in body weight was obtained for each group. This was calculated as the difference in animal body weight on the day of measurement and weight at onset of the study ($Day_{measurement} - Day_0$) according to the method of Zhou *et al*.[24]

Statistical Analysis

Descriptive statistics was carried out on the data generated and results were expressed as the Mean ± Standard Deviation. Significant differences between groups were separated by one way ANOVA and Duncan's multiple comparison test. Data analysis was done using *Statistical Package for the Social Sciences* (SPSS) version 20.0 (SPSS Inc Chicago IL) while body weight curve was plotted using Microsoft Excel 2007 (Microsoft Corporation US). A *p*-value < *0.05* was considered statistically significant.

Results

Effect of concomitant feeding of high fat diet and administration of methanol extract of *Vernonia colorata* on lipid profile:

Effect on Total Cholesterol concentration:

The administration of methanol extract of *Vernonia colorata* to high fat diet fed albino rats resulted in a dose dependent significant *(p<0.05)* decrease in the mean total cholesterol of the animals in the treated group relative to the group that received only high fat diet (Figure 2).

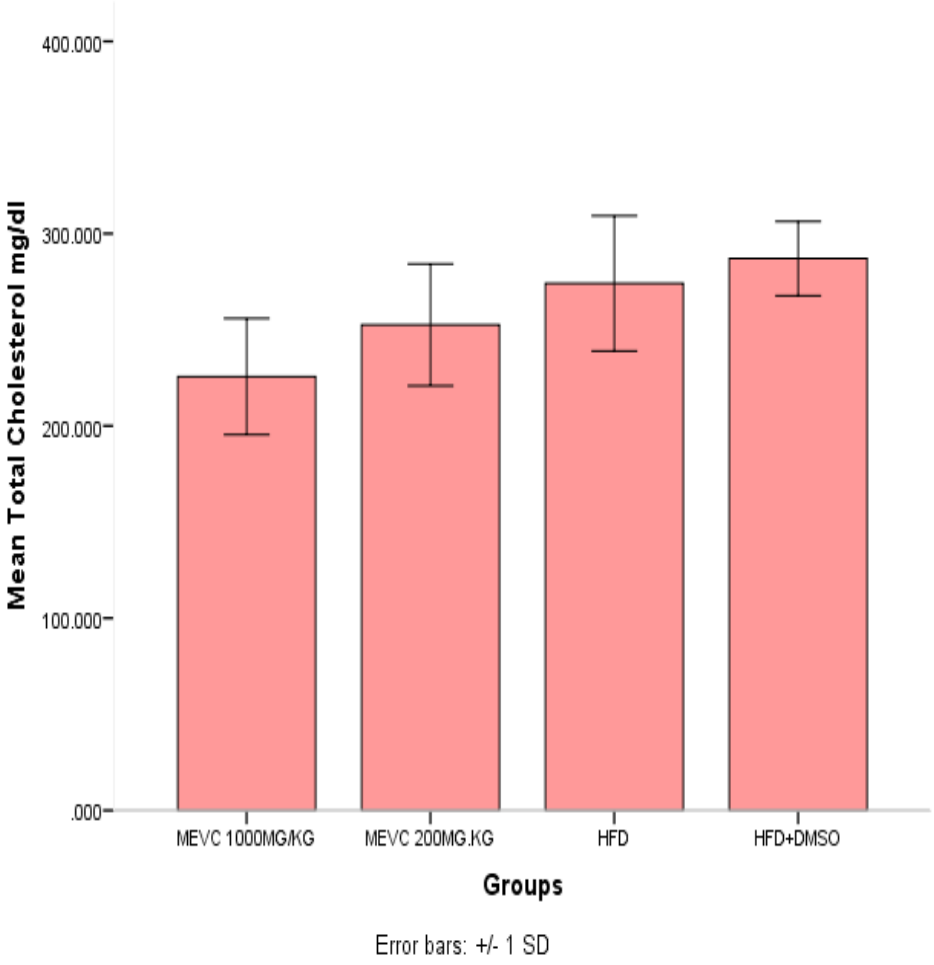

Figure 2: Total cholesterol concentration of rats concomitantly fed high fat diet and administered methanol extract of *Vernonia colorata*.

Effect on Triacylglycerol concentration:

Figure 3 shows that the administration of methanol extract of *Vernonia colorata* resulted in a significant *($p<0.05$)* decrease in triacylglycerol concentration of animals that received extracts relative to the high fat diet control group.

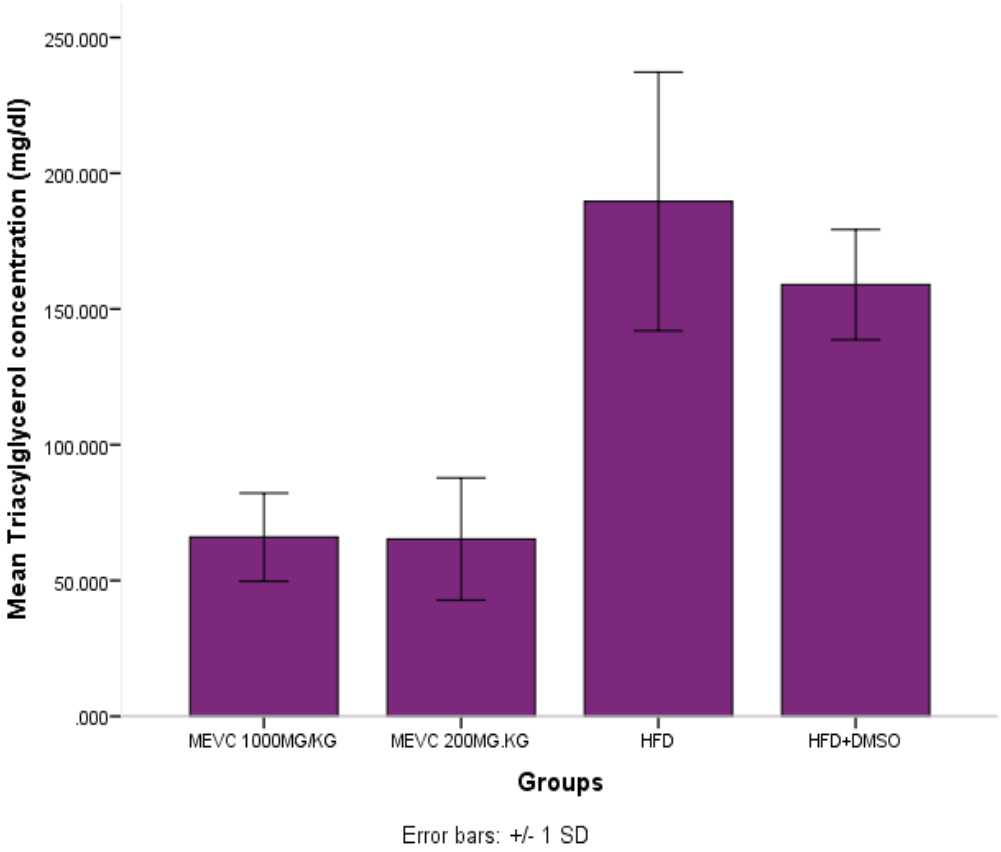

Figure 3: Triacylglycerol concentration of rats concomitantly fed high fat diet and administered methanol extract of *Vernonia colorata*.

Effect on HDL cholesterol concentration:

The administration of methanol extract of *Vernonia colorata* caused significant *($p<0.05$)* increase in HDL cholesterol concentration in the treated groups relative to the control group (Figure 4).

Effect on LDL cholesterol concentration:

There was a significant *($p<0.05$)* decrease in LDL cholesterol concentration upon concomitant feeding of high fat diet and administration of methanol extract of *Vernonia colorata* relative to the high fat diet control group (Figure 5).

Effect of concomitant feeding of high fat diet and administration of methanol extract of *Vernonia colorata* on body weight changes:

Concomitant feeding of high fat diet and administration of methanol extracts of *Vernonia colorata* resulted in respective decrease of 22.2 % and 15.8 % in body weight of animals that were administered 1000 mg/kg and 200 mg/kg of methanol extract of *Vernonia colorata* relative to 12.5 % decrease in the Orlistat group (Figure 6).

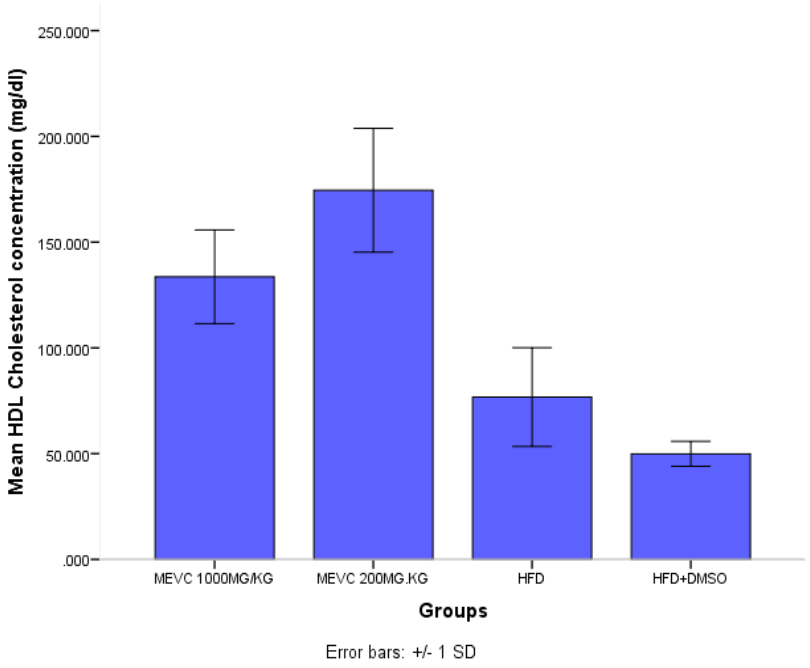

Figure 4: HDL cholesterol concentration in rats concomitantly fed high fat diet and administered methanol extract of *Vernonia colorata*.

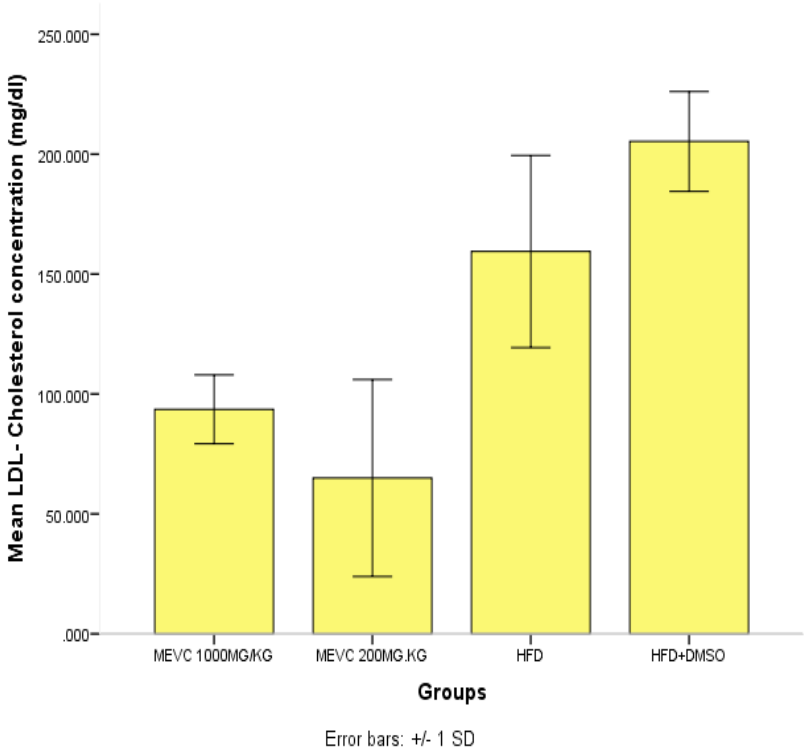

Figure 5: LDL cholesterol concentration in rats concomitantly fed high fat diet and administered methanol extract of *Vernonia colorata*.

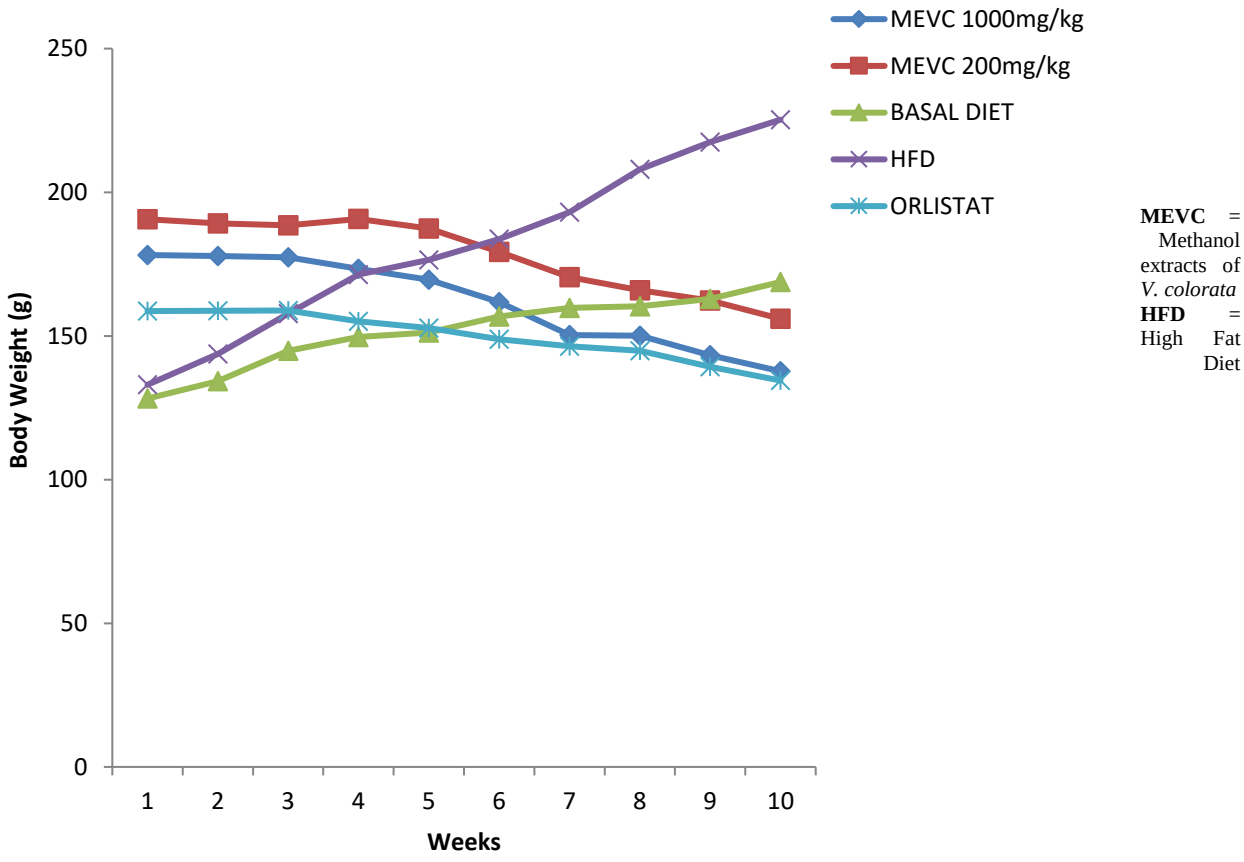

Figure 6: Effect of concomitant feeding of high fat diet and administration of methanol extracts of *Vernonia colorata* on body weight of rats.

Discussion

Increased consumption of high fat foods rich in cholesterol and triacylglycerols has been implicated in the elevation of plasma cholesterol level and has emerged as a strong risk factor for metabolic diseases such as, obesity, diabetes and cardiovascular diseases.[25,3] The use of diet to regulate plasma lipid level is now gaining an enormous acceptance worldwide. This underscores the recent focus on medicinal plants in many dietary studies as scientists seek to explore the wide range of therapeutic benefits of such medicinal plants.[1,2]

In this present study, it was observed that there was a decrease in plasma cholesterol level in rats administered methanol extract of *Vernonia colorat*a when compared with rats that were fed high fat diet only. The dose dependent decrease in cholesterol levels were observed in both groups concomitantly fed and administered methanol extracts of *Vernonia colorata* at both 1000 mg/kg and 200 mg/kg doses. This finding which is in agreement with previous findings [26,17] may be attributed to the repertoire of phytochemicals present in the methanol extract of *Vernonia colorata* leaves.[18] Such phytochemicals include but are not limited to saponins, sesquiterpenes, lactones and flavonoids.[27]

Rats that were fed only high fat diet showed higher plasma levels of triacyglycerols than rats that were treated with extracts of *Vernonia colorata*. The findings of this study indicated that the administration of *Vernonia colorata* to experimental animals effectively reduced triacylglycerol levels. These findings agree

with previous findings that have reported hypolipidemic effects of extracts of *Vernonia* species such as *V. amygdalina* in normolipidemic rats.[26,17]

The prevention and control of dyslipidemia is important in achieving a reduction in the burden of cardiovascular diseases.[5] Research evidence has shown that polyphenols and saponins contribute to the prevention of diseases related to lipid derangements.[28] The phytochemical analysis of *Vernonia colorata* has revealed the presence of dietary fiber, saponins and polyphenols.[29] Antioxidants prevent oxidative stress caused by free radicals that are produced *via* lipid peroxidation. Therefore, by terminating chain reactions triggered by these free radicals, polyphenols and saponins contained in *Vernonia colorata* may prevent the onset of cardiovascular diseases. Fundamentally, polyphenols and saponins contained in *Vernonia colorata* are known to stimulate cholesterol lowering activity by eliciting resin-like action, thereby reducing the enterohepatic circulation of bile acids. This process ensures the conversion of more cholesterol into bile acids, thus resulting in the reduction of total plasma cholesterol level due to increased excretion.[18]

In this present study, the administration of graded doses of methanol extract of *Vernonia colorata* caused significant elevation of HDL cholesterol in the treated groups relative to the control group. This finding is consistent with a previous report.[30] The findings of a dose-dependent elevation of HDL-cholesterol in this study is indicative of a positive modulation of lipid profile in high fat diet fed rats. This is because of the anti-atherogenic role which HDL cholesterol plays in promoting reverse cholesterol transport and ensuring that cholesterol from peripheral tissues return to the liver to be either excreted or reused.[31] Therapeutic interventions that raise HDL-cholesterol are highly encouraged, as higher levels have been correlated with reduced risk of conditions such as metabolic syndrome and future risk of cardiovascular diseases.[32]

Elevated LDL cholesterol in the blood plasma is considered detrimental, as their increased concentration can potentiate glucose intolerance [33] and cause lipid accumulation in the arteries. This leads to an increase in the adherence of circulating monocytes to the arterial endothelial cells where they differentiate to form macrophages and become converted to foam cells which can exacerbate atherosclerosis.[34] The findings of this present study revealed that administration of methanol extracts of *Vernonia colorata* administered at both 1000 mg/kg and 200 mg/kg significantly lowered LDL cholesterol concentrations of rats that were concomitantly fed a high fat diet. This finding is consistent with an earlier study which reported that incorporation of *Vernonia colorata* in the diets of normolipidemic rats had a lowering effect on serum low density lipoprotein (LDL) cholesterol concentration at both 5 % and 10 % dietary incorporation levels.[15]

Administration of methanol extract of *Vernonia colorata* at 1000 mg/kg and 200 mg/kg significantly *($p<0.05$)* down regulated weight gain in animals fed high fat diet relative to the untreated group that was fed high fat diet only. This is consistent with previous reports.[35,2] The dose dependent steady loss in weight could have been as a result of the presence of anti-nutritional factors such as saponins in *Vernonia colorata* which may reduce lipid distribution and nutrient availability by delaying the absorption of fats in the intestines.[36,30]

Conclusion

This study showed that the administration of methanol extracts of *Vernonia colorata* to animals fed a high fat diet had beneficial effects on the plasma lipid profile and body weight changes of the animals. The findings of this study therefore provide convincing evidence for the hypolipidemic and anti-obesity properties of methanol extracts of *Vernonia colorata*. This suggests that the plant may be useful in weight loss regimen, attenuating dietary obesity and also serve as a potential drug lead in the search for natural products for the treatment of diseases associated with dyslipidemia.

References

1. Thompson, A.K., Minihane, A.M. and William, C.M. (2011). Trans fatty acids and weight gain. *Int. J. Obes.* **35:** 315-324.
2. Atangwho, I.J., Edet, E.E., Uti, D.E., Obi, A.U., Asmawi, M.Z. and Ahmad, M. (2012). Biochemical and histological impact of *Vernonia amygdalina* supplemented diet in obese rats. *Saudi J. Biol. Sci*. **19**(3): 385-392. https://doi.org/10.1016/j.sjbs.2012.05.003.
3. Duca, F.A., Sakar, Y. and Covasa, M. (2013). The Modulatory Role of High Fat Feeding on Gastrointestinal signals in Obesity. *J. Nutri. Biochem.* **24**: 1663-1677.
4. Musunuru, K. (2010). Atherogenic dyslipidemia: cardiovascular risk and dietary intervention. *Lipids*. **45**(10): 907-914.
5. Opoku, S., Gan, Y., Fu, W., Chen, D., Addo-Yobo, E., Trofimovitch, D., Yue, W., Yan, F., Wang, Z. and Lu, Z. (2019). Prevalence and risk factors for dyslipidemia among adults in rural and urban China: findings from the China National Stroke Screening and prevention project (CNSSPP). *BMC Public Health*. **19:** 1500. https://doi.org/10.1186/s12889-019-7827-5.
6. Nnamudi, A.C., Orhue, N.E.J., Ijeh, I.I., Etim, O.E. and Eke-Ogaranya, I.N. (2020). Obesity, Metabolic Abnormalities, Metabolic Syndrome and BMI-Metabolic-Risk Sub-Phenotypes among Young Adult Nigerians. *Haya Saudi J. Life Sci.* **5**(6): 90-97. DOI: 10.36348/sjls.2020.v05i06.001.
7. Lan, N.S.R., Fegan, P.G., Yeap, B.B., Bell, D.A. and Watts, G.F. (2019). Dyslipidaemia in adults with type 1 diabetes-when to treat? *Diabetes Metab. Res. Rev.* **35**(1): e3090. DOI: 10.1002/dmrr.3090.
8. Ghosh, S., Parihar, V.S., More, P., Dhavale, D.D. and Chopade, B.A. (2015). Phytochemistry and therapeutic potential of medicinal plant: *Dioscorea bulbifera*. *Med. Chem.* **5**: 160-172.
9. Nnamudi, A.C., Onyeche, V.O., Ebohon, O. and Eke-Ogaranya, I.N. (2020). Nigerian Medicinal Plants for the Management of Liver Diseases: A Review. *European J. Med. Plants.* **31**(12): 29-51. DOI: 10.9734/EJMP/2020/v31i1230302.
10. Willcox, M.L. (1999). A clinical trial of 'AM', a Ugandan herbal remedy for malaria. *J. Public Health Med*. **21**(3): 318–324.
11. Okwu, D.E. (2007). Nigerian Medicinal Plants I. *Med. Aromat. Plant Sci. Biotechnol.* **1**(1): 90-96.
12. Paniagua-Zambrana, N.Y., Bussmann, R.W., Hart, R.E., Moya-Huanca, A.L., Ortiz-Soria, G., Ortiz-Vaca, M., Ortiz-Alvarez, D., Soria-Morán, J., Soria-Morán, M., Chávez, S., Chávez-Moreno, B., Chávez-Moreno, G., Roca, O. and Siripi, E. (2018). Who should conduct ethnobotanical studies? Effects of different interviewers in the case of the Chácobo Ethnobotany project, Beni, Bolivia. *J. Ethnobiol. Ethnomed.* **14**(1): 9. DOI: 10.1186/s13002-018-0210-2.
13. Burkill, H.M. (2000). The Useful Plants of West Tropical Africa. (2nd Ed.) Vol. 5. Royal Botanic Gardens, Kew. United Kingdom; pp 686.
14. Igile, G.O., Olesezk, W., Burda, S. and Jurzusta, M. (1995). Nutritional Assesment of *Vernonia amygdalina* Leaves in Growing Mice. *J. Agric. Food Chem.* **43**(8): 2162-2166.
15. Egedigwe, C.A. and Ijeh, I.I. (2010). Body and organ weight changes following dietary incorporation of *Vernonia colorata* and *Vernonia amygdalina* Del in Albino rats. *Niger. J. Nutri. Sci*. **31**(2): 58-61
16. Golly, K.J., Siaka, S., Guessennd, N., Soro, Y., Djama, A.J. and Dosso, M. (2012). Phytochemical assessment and antimicrobial activity of leaves extract of *Vernonia colorata* (Wild.) Drake on Resistant Germs of *Staphylococcus aureus* and *Pseudomonas aeruginosa*. *J. Chem. Pharm. Res.* **4**(5): 2490-2494
17. Ijeh, I.I. and Egedigwe, A.C. (2010). Effect of dietary incorporation of *Vernonia colorata* (Wild) leaves on blood lipid profile of albino rats. *Int. J. Biol. Chem. Sci.* **4**(1): 100-106.
18. Egedigwe, C.A., Ijeh, I.I., Okafor, P.N. and Ejike, C.E.C.C. (2016). Aqueous and methanol extracts of *Vernonia amygdalina* leaves exert their anti-obesity effects through the modulation of appetite-regulatory hormones. *Pharm. Biol*. **54**(12): 3232-3236. DOI: 10.1080/13880209.2016.1216135.
19. National Research Council. (2011). Guide for the Care and Use of Laboratory Animals: Eighth Edition. Washington, DC: The National Academies Press. https://doi.org/10.17226/12910.
20. Allain, C.C., Roon, L.S., Chan, C.S.G., Richard, W. and Fu, P.C. (1974). Enzymatic determination of total serum cholesterol. *Clin. Chem.* **20**:470-475.
21. Tietz, N.W. (1990). Clinical guide to laboratory tests. 3rd ed. Philadelphia. W.B. Saunders, pp. 555-556.
22. Lopes-Virella, M.F., Stone, P., Ellis, S. and Colwell, J.A. (1977). Cholesterol determination in high-density lipoproteins separated by three different methods. *Clin. Chem*. **23**(5): 882-884.

23. Friedewald, W.T., Levy, R.I. and Frederickson, D.S. (1972). Estimation of the concentration of LDL cholesterol in plasma, without the use of the preparative ultra-centrifuge. *Clin. Chem*. **18**: 499-502.
24. Zhou, J., Keenan, M.J., Losso, J.N., Raggio, A.M., Shen, L., McCutcheon, K.L., Tulley, R.T., Blackman, M.R. and Martin, R.J. (2011). Dietary whey protein decreases food intake and body fat in rats. *Obesity (Silver Spring)*. **19**(8): 1568-1573. doi: 10.1038/oby.2011.14.
25. Gosain, S., Ircchiaya, R., Sharma, P.C., Suresh, T., Kalra, A., Deep, A. and Bhardwaj, T.R. (2010). Hypolipidemic effect of ethanolic extract from the leaves of *Hibiscus sabdariffa* L. in hyperlipidemic rats. *Acta. Pol. Pharm*. **67**(2): 179-184.
26. Adaramoye, O.A., Akintayo, O., Achem, J. and Fafunso, M.A. (2008). Lipid-lowering effect of methanolic extract of *Vernonia amygdalina* leaves in rats fed on high cholesterol diet. *Vasc. Health Risk Manag*. **4**:235-241.
27. Ijeh, I.I. and Ejike, C.E.C.C. (2011). Current perspectives on the Medicinal Potentials of *Vernonia amygdalina* Del. *J. Med. Plant. Res*. **5**(7): 1051-1061.
28. Moradi-Afrapoli, F., Asghari, B., Saeidnia, S., Ajani, Y., Mirjani, M., Malmir, M., Bazaz, R.D., Hadjiakhoondi, A., Salehi, P., Hamburger, M. and Yassa, N. (2012). *In vitro* α-glucosidase inhibitory activity of phenolic constituents from aerial parts of *Polygonum hyrcanicum. Daru.* **20**(1): 37.
29. Ejoh, A.R., Nkonga, V.D., Innocent, G. and Moses, C.M. (2007). Nutritional components of some Non-Conventional Leafy Vegetables consumed in Cameroun. *Parkistan J. Nutr*. **6**(6): 712-717.
30. Ijeh, I.I., Egedigwe, A.C., Inyang, E.I. and Emmanuel, G. (2014). Hypolipidaemic effects and body weight changes of male Wistar rats administered crude saponin extracts from *Vernonia amygdalina* and *Vernonia colorata* leaves. *Niger. J. Biochem. Molecular Biol.* **29**(1): 34-43.
31. Stapleton, P.A., Goodwill, A.G., James, M.E., Brock, R.W. and Frisbee, J.C. (2010). Hypercholesterolemia and microvascular dysfunction: interventional strategies. *J. Inflamm.* **7**:54.
32. Chiesa, S.T. and Charakida, M. (2019). High-Density Lipoprotein Function and Dysfunction in Health and Disease. *Cardiovasc. Drugs Ther.* **33:** 207-219. https://doi.org/10.1007/s10557-018-06846-w.
33. Singh, A.K., Singh, N.K., Agrawal, N. and Gopal, K. (2011). Obesity and dyslipidemia. *Int. J. Biol. Med. Res.* **3**(2): 824-828.
34. Asem, N. and Asem, B.B. (2009). The Many Faces of Cholesterol: How Modifications in LDL and HDL Alter Their Potential to Promote or Prevent Atherosclerosis. *The UCLA USJ*. **22**: 1-17
35. Atangwho, I.J., Ebong, P.E., Egbung, G.E., Eteng, M.U. and Eyong, E.U. (2007). Effect of *Vernonia amygdalina* Del on liver function in alloxan-induced hyperglycemic rats. *J. Pharm. Bioresour*. **4**: 25-31.
36. Ijeh, I.I. and Obidoa, O. (2001). Effect of dietary incorporation of two varieties of *Vernonia amygdalina* on mean relative organ weight of weanling rabbits. *Niger. J. Biochem. Molecular Biol*. **16**:13