Vol. 12(2), pp. 19-27, April-June 2020 DOI: 10.5897/JPP2016.0436 Article Number: D534B0A63364 ISSN: 2141-2502 Copyright©2020 Author(s) retain the copyright of this article http://www.academicjournals.org/JPP



Journal of Pharmacognosy and Phytotherapy

Full Length Research Paper

Phytochemical profiling, body weight effect and antihypercholesterolemia potentials of *Cnidoscolus aconitifolius* leaf extracts in male albino rat

¹Chukwu, Ezinne.C.*, ²Osuocha, Kelechi U., ³Iwueke, Adaku V

¹Department of Biochemistry, Federal University of Lafia, Nasarawa State, Nigeria. ²Biochemistry Unit, Department of Science Laboratory Technology, Federal University of Lafia, Nasarawa State, Nigeria. ³Department of Biochemistry, PAMO University of Medical Sciences, Rivers State, Nigeria.

Received 23 October, 2016; Accepted 16 November, 2016

The present study investigated the phytochemical constituents of *Cnidoscolus aconitifolius* leaves extracts and its anti-hypercholesterolemia potentials using standard analytical methods. Forty five male albino rats weighing (115-121 g), divided into nine groups of five rats were used. Group I served as the control while the other groups were administered 200, 400, 600 and 800 mg/kg body weight of aqueous and ethanol leaf extracts. GC-MS analysis showed 3,7,1,5-tetramethyl-2-hexadecen-1-ol, farnesyl bromide, β -sitosterol, squalene, β -amyrin, 1-heptatriacotanol, hexadecanoic acid, methyl ester, 2-pentadecanone, 6,10,14-trimethyl-, n-hexadecanoic acid, 9,12-octadecadienoyl chloride, (Z,Z, δ -tocopherol, Ergosta-5,22-dien-3-ol acetate, (3 β ,22E)-, 9,10-secocholesta-5,7,10(19)-triene-3,24,25-triol, (3 β ,5Z,7E)-acetamide, N-methyl-N-[4-(3-hydroxypyrrolidinyl)-2-butynyl]-, 1-gala-I-ido-octose, 10-methyl-E-11-tridecen-1-ol propionate, dodecanoic acid, 2-(acetyloxy)-1-[(acetyloxy)methyl]ethyl ester, 11,14-octadecadienoic acid, methyl ester, cyclopentaneundecanoic acid and methyl ester. Lipid profile showed significant reduction in TC, LDL and TG with increase in HDL in dose dependent ratio. This shows that extracts of this plant could be useful in treatment of coronary heart diseases.

Key words: Phytochemicals, hypercholesterolemia, Cnidoscolus aconitifolius, potentials.

INTRODUCTION

The use of medicinal plants in disease treatment has attracted the interest of man as these plants serve as potential sources of natural compounds with biological activities. Especially in developing countries, man has tried to lessen pain or treat diseases using plants with medicinal properties (Sakpa and Okhimamhe, 2014; Hussein et al., 2016). Dhanalakshmi and Manavalan (2014) posited that plants owing to its medicinal efficacy have continued to play a dominant role in the maintenance of human health. Plants derived medicine has been a part of the evolution of human healthcare for thousands of years (Sowmya et al., 2015). According to Ebeye et al. (2015), many people have for centuries developed various herbal medicines using locally available plants as remedy to their health problems. Oluwatosin et al. (2011) reported that herbal medicines

*Corresponding author. E-mail: callychuksu@gmail.com.

Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> License 4.0 International License



Figure 1. Cnidoscolus aconitifolius . Source: Mordi et al., (2013)

derived from plants extracts are being utilized increasingly to treat a wide variety of diseases and these plants are good sources of bioactive compounds and may boost the endogenous antioxidant defense system. Cnidoscolus aconitifolius belongs to the family of Euphorbiaceae. It is an evergreen, drought deciduous shrubs up to 6 m in height with alternate palmate lobed leaves, milky sap and small flowers on dichotomously branched cymes (Ovevemi and Ajani, 2014). It is a widely distributed annual plant, ranging from temperate to tropical zones and has a long history of use as both medicine and edible plant and due to its ease of cultivation, potential productivity and substantial nutritional value, the plant has spread all over the world including the tropics (Omotoso et al., 2014a). Ross-Ibarra and Molina-Cruz (2002) opined that due to its tolerance to poor growth conditions and resistance to pests and diseases, the plant has remained a valuable potential crop that could medically benefit people of different regions. C. aconitifolius leaves has been claimed traditionally to possess medicinal properties and efficacy such as darkening of gray hair, treating alcoholism, insomnia, gout, scorpion sting, brain and vision improvement (Mordi et al., 2013). In view of all the reputed medicinal efficacy of this plant leaves, the present study was aimed at evaluating the bioactive constituents of this plant leaves in order to ensure holistic utilization of this plant in disease treatments Figure 1.

MATERIALS AND METHODS

C. aconitifolius leaves were harvested from Acha in Isuikwuato L.G.A Abia State Nigeria. The leaves were separated from stem and washed with clean water and dried at room temperature. The dried plant leaves were ground into powder form using blender which was transferred into an air tight container stored at room temperature.

Preliminary phytochemical screening

The methods described by Odebiyi and sofowora (1978) were used to test for the presence of saponins, tannins, phenolics and alkaloids. Lieberman Burchad reaction as described by Herburne (1973 and Sofowara, 1993) was used for steroids and glycosides determination.

Phytochemicals analysis of *C. aconitifolius* leaves using GC-MS

The samples for GC-MS analysis were prepared by dissolving 3 g of extracted powder in methanol solvent. For the analysis, GC-MS-QP 2010 SHIMADZU instrument was used. To analyze the sample, the column oven and Injector temperature was set at 800 and 200°C respectively. The flow control mode was maintained in linear velocity with a split injection mode split ratio of 20. The column flow was 1.46 ml/min with a helium carrier gas of 99.9995% purity. The column oven temperature program was set as follows: The temperature was set at 80°C with 2 min hold time at the rate of 10. The temperature was 300°C with 10 min hold time. The column at 5 min was used with a length of 30 mm and diameter of 0.25 mm and its film thickness was 0.25 μ m. The ion source temperature for MS condition was 200°C and interface temperature was 240°C. Starting m/z (mass to charge) ratio was 40 and ending with m/z ratio of 700. (40-700 m/z).

Identification of components

Interpretation of mass spectrum GC-MS was conducted using the NIST Database. The spectrums of the unknown components were compared with spectrum of known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained.

Preparation of aqueous leaf extract

Exactly 200 g of the powdered plant were measured into a conical flask and 500 ml of water was added and left at room temperature for 48 h. The extracts were filtered. The filtrate was evaporated to dryness on a water bath to give a crude extract. The extraction efficiency was quantified by determining the weight of the extract. The dried extract was stored in desiccators until required for use. The extract was dissolved in appropriate volume of distilled water to the desired concentration (Gidado et al., 2005).

Preparation of ethanol leaf extract

Exactly 200 g of the powdered plant were measured into a conical flask and 500 ml of 70% ethanol were added and left at room temperature for 48 h. The extract was filtered. The filtrate was evaporated to dryness on a water bath (50°C) to give the crude extract, whose the mass was determined.

Experimental design

Forty five male albino rats aged 9 weeks weighing 115 - 121g were used for this study. The animals were randomly divided into 9 groups of 5 rats each for biochemical assessment of the effect of aqueous and ethanol extracts of *C. aconitifolius* leaves. Group I served as control, Group II received 200 mg of aqueous extract, Group III received 400 mg of aqueous extract per kg body weight, Group IV received 600 mg of aqueous extract and Group V received 800 mg of aqueous extract. Group VI received 200 mg of ethanol extract while Group VII received 400 mg of ethanol extract per kg body weight. Group VIII received 600 mg/kg while Group IX received 800 mg/kg body weight of ethanol extract. Each group of animals were housed in a standard rat cage and allowed to acclima-

Compounds	Aqueous extract	Ethanolic extract	
Alkaloid	+	++	
Tannin	+	++	
Saponins	+	+ +	
Flavonoids	-	+	
Oxalate	+	+	
Cyanogenic glycosides	+	-	
Phenol	+	++	

 Table 1. Preliminary phytochemical analysis of Cnidoscolus aconitifolius leaf extracts.

+ Present; - absent.

tize to laboratory condition for one week prior to commencement of feeding experiments. The extracts were administered once on daily basis. All animals were allowed free access to water and feed *ad libitum.* The method of administration of the extracts was by oral gavages which lasted for 28 days.

Determination of lipid profile

The method of Fossati and Prencipe (1982) was employed in the determination of lipid profile.

Statistical analysis

The statistical analysis of results was done using students package for social sciences (SPSS) version 20 computer software and data collected were analyzed using Analysis of Variance (ANOVA). Means were separated using Least Significant Difference (LSD).

RESULTS

The preliminary phytochemical results above shows the presence of alkaloids, tannins, saponins, flavoniods, oxalate and in aqueous and ethanol leaf extracts while ethanol leaf extract shows the absence of cyanogenic glycosides which was present in aqueous leaf extract (Table 1).

Results of the GC-MS analysis of C. aconitifolius leaf extract showed 19 compounds. The fragmentation pattern of the major compound was 3,7,11,15tetramethyl-2-hexadecen-1-ol with percentage peak of 22.733% and retention time of 14.736. Farnesyl bromide was also detected with percentage area peak of 19.457% and a retention time of 31,499 was also detected. β – Sitosterol with area peak of 16.513% and retention time of 31.841 was detected. Squalene with area peak of 14.469% and retention time 24.476 was found. β-Amyrin with area peak of 5.168% and retention time of 34.697 and 1-Heptatriacotanol with peak area of 4.232% and time of 32.168 were retention also detected. Hexadecanoic acid, methyl ester was found with area peak of 2.705 with retention time of 12.436. 2-Pentadecanone, 6,10,14-trimethyl- with area peak of 2.556 and retention time of 11.490 was detected. nHexadecanoic acid with area peak of 2.115% with retention time of 12.880; 9,12-octadecadienovl chloride, (Z,Z)-with area peak of 1.885% and retention time of 14.608; δ-tocopherol with area peak of 1.454% and retention time of 26.166; ergosta-5.22-dien-3-ol,acetate, $(3\beta, 22E)$ - with area peak of 1.039% and retention time of 30.600; 9,10-Secocholesta-5,7,10(19)-triene-3,24,25-triol, (3β,5Z,7E)- with area peak of 0.912% and retention time of 29.616 were detected (Figure 2). Acetamide, N-methyl-N-[4-(3-hydroxypyrrolidinyl)-2-butynyl]- with area peak of 0.852% with retention time of 10.987; I-Gala-I-ido-octose with area peak of 0.843% and retention time of 4.462; 10methyl-E-11-tridecen-1-ol propionate with area peak of 0.822% and retention time of 11.436 (Figure 2) were also Dodecanoic 2-(acetyloxy)-1detected. acid, [(acetyloxy)methyl]ethyl ester with area peak of 0.776% with retention time of 11.688; 11,14-octadecadienoic acid, methyl ester with area peak of 0.758 and retention time of 14.528 and cyclopentaneundecanoic acid, methyl ester with area peak of 0.709% with retention time of 14.961 were also detected.

Results of lipid profile presented in Table 4 shows that total cholesterol ranged from 83.56 mg/dl in the control to 64.23mg/dl in the treated group. Results indicate a gradual reduction in total cholesterol level in treated groups compared to control (p<0.05). Results of triglyceride show a range of 69.84 to 91.10mg/dl. Results show that the control had the highest triglyceride value of 91.10 mg/dl while group IX administered 800 mg/kg body weight of C. aconitifolius ethanol leaf extract had the least value of 69.84 mg/dl. High density lipoprotein result shows that the control had the lowest value of 30.32 ma/dl while aroup IX had the highest value of 48.78 mg/dl. Results suggest a gradual increase in high density lipoprotein level with increasing dosage of C. aconitifolius leaf extracts. Results of low density lipoprotein indicate that the control had the highest value of 71.46 mg/dl while group IX recorded the least value of 29.41 mg/dl.

DISCUSSION

Phytochemical constituents are responsible for most

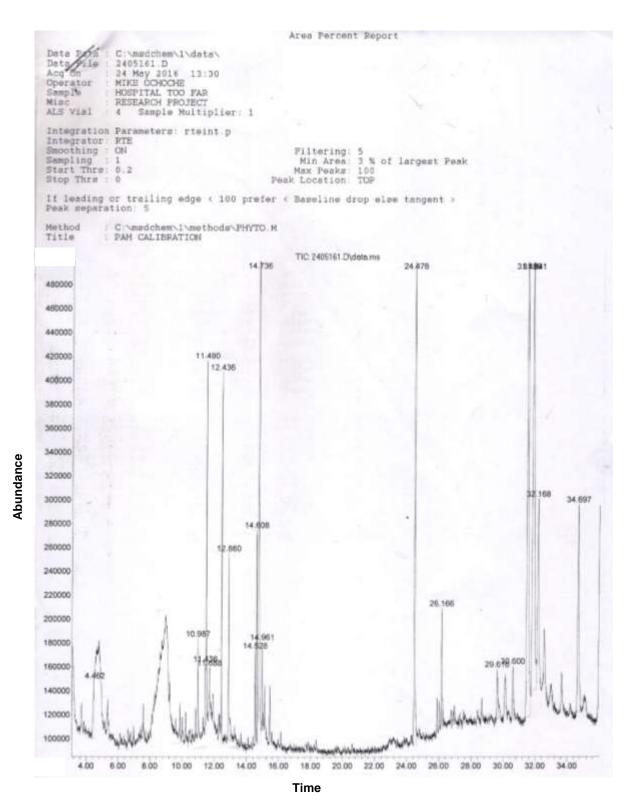


Figure 2. GC-MS chromatogram of methanol leaf extract of Cnidoscolus aconitifolius.

medicinal activities attributed to medicinal plant species (Obichi et al., 2015). The analysis of these phytochemicals is paramount as World Health Organization (WHO) has specified the need to determine the composition of biologically active substances considered for nutritional and medicinal purposes (WHO, 2012). The preliminary phytochemical analysis carried out on aqueous and ethanol extracts of C. aconitifolius leaves showed the presence of some bioactive compounds in the plant as presented in (Table 1). Results showed the presence of tannins, saponins, phenol, oxalate, phytate and alkaloids in aqueous and ethanol leaf extract. Flavoniods were absent in aqueous leaf extract while cyanogenic glycosides was absent in the ethanol leaf extract. This finding is in consonance with previous reports of Awoyinka et al. (2007) and Oluwatosin et al. (2011) who reported absence of cyanogenic glycosides in C. aconitifolius ethanolic leaf extract. but contrary to the findings of Adaramove and (2011) who reported presence of cyanogenic Aluko glycoside in ethanol leaf extract of C. aconitifolius. This could be adduced to be as a result of environmental factors or method of extraction. Analysis of tannins in the two extracts was positive but higher colour intensity was observed in the ethanol extract than the aqueous leaf extract. This could be attributed to the ethanol extracting potentials; more than water. Tannins are complex moiety with wide pharmacological activities and are produced by majority of plants as protective substance (Yakubu et al., 2008). The presence of tannins suggests the ability of this plant to play a major role as antihaemorrhagic agent (Araujo et al., 2008). Parekh and Chanda (2007) posited that tannins are known to react with proteins to provide the typical tanning effects which are important for the treatment of inflamed or ulcerated tissues. Araujo et al. (2008) also opined that the healing and anti-inflammatory activities popularly attributed to C. aconitifolius may be strongly associated with its tannin content.

Analysis of saponins shows positive results for both aqueous and ethanol leaf extracts. However, intense persistent frosting was observed in the ethanol extract than the aqueous extract. This could also be attributed to the extracting potentials of ethanol. Studies have shown that saponin play immense role as antihyperglycaemia, antihypercholesterolemia, cardiac depressant and dietary source saponins have been implicated of in chemopreventive strategy in lowering the risk of human cancer (Olaleve, 2007). Results of alkaloids analysis show that alkaloids were present in both extracts but more intense in ethanol extract than the aqueous extracts. The main reason that can be adduced for this is the solvent of extraction. Alkaloid is one of the largest groups of phytochemicals in plants and has been reported to be effective in the treatment of intestinal infections and hypertension (Akinpelu and Onakoya, 2006). This could also further justify the local use of C.aconitifolius leaf extracts in the management of cardiac related issues.

Cardiac glycoside was also detected in aqueous extract but absent in ethanolic extract. This is in concomitant with the reports of Mordi and Akanji (2012) who also reported absence of cardiac glycoside in ethanol leaf extract of *C. aconitifolius.* Olayinka et al. (1992) posited that cardiac glycosides have been used for over two centuries as stimulants in cases of cardiac failure. This perhaps may justify the already locally established function of the plant in the treatment and management of hypertension.

Flavonoids were absent in aqueous extract but present in ethanol extract of C. aconitifolius leaf. Similar findings have been reported by Awoyinka et al. (2007) and Mordi and Akanji (2012) who reported absence of flavonoids in aqueous leaf extract of C. aconitifolius. Sofowora (1993) opined that flavonoids contribute to the brilliant multi colour for most plants. Hence, the absence of flavonoid in the aqueous leaf extract may substantiate the sole greenish appearance of this leafy vegetable. Flavonoids have been reported to exhibit anti-oxidative properties through several mechanisms, such as scavenging of free radicals, chelation of metal ions such as iron and copper, inhibition of hydrolytic and oxidative enzymes and also act as anti-inflammatory agent (Kessler et al., 2003). On this premise, it may be advisable to extract the leaf of C. aconitifolius with ethanol in an attempt to exploit its antioxidative and anti-inflammatory properties since flavonoids are known to be effective for these purposes.

Phytate was obtained from this study in both aqueous and ethanolic extracts and its presence has been linked to the prevention of kidney stone, dental decay and calcification of blood vessels (Dutta and Chaudhuri, 2015). Aberoumand and Deokule (2009) opined that phytate may have beneficial roles as an antioxidant and antcarcinogen. Phenols were observed in both aqueous and ethanol leaf extracts. This is an indication that the plant might play an important role as dietary antioxidants as phenol have been known to prevent oxidative damage in living systems (Zee-cheng, 1997). Findings from this study show that the bioactive constituents of *C. aconitifolius* could be better extracted using ethanol as solvent of extraction.

Gas chromatography-mass spectroscopy analyses of compounds are presented in Table 2. The GC-MS analysis of C. aconitifolius leaves revealed a total of 19 prevailing compounds in methanol extract of C. aconitifolius leaf that could contribute to the medicinal relevance of the plant (Figure 2). The phyto-components with their respective retention time (RT), molecular formula, molecular weight (Mw) and relative percentages (peak areas %) are presented in Table 2. The major phytoconstituents present are I-Gala-I-ido-octose, N-methyl-N-[4-(3-hydroxypyrrolidinyl)-2acetamide, butynyl]-,10-methyl-E-11-tridecen-1-ol propionate, 2pentadecanone, 6,10,14-trimethyl-, dodecanoic acid, 2-(acetyloxy)-1-[(acetyloxy)methyl]ethyl ester. hexadecanoic acid, methyl ester, n-hexadecanoic acid, 11.14-octadecadienoic acid. methyl ester. 9.12octadecadienovl chloride, (Z.Z)-.3,7,11,15-tetramethyl-2hexadecen-1-ol, cyclopentaneundecanoic acid, methyl ester, squalene, δ- tocopherol, 9,10-secocholesta-5,7,10(19)-triene-3,24,25-triol, (3β,5Z,7E)-,ergosta-5,22dien-3-ol,acetate, (3β,22E)-, farnesyl bromide, β –

S/N	Retention time	Compound name	Formula	Mw	Area (%)
1	4.462	I-Gala-I-ido-octose	$C_8H_{16}O_8$	240	0.843
2	10.987	Acetamide, N-methyl-N-[4-(3-hydroxypyrrolidinyl)-2-butynyl]-	$C_{11}H_{18}N_2O_2$	210	0.852
3	11.436	10-Methyl-E-11-tridecen-1-ol propionate	$C_{17}H_{32}O_2$	268	0.822
4	11.490	2-Pentadecanone, 6,10,14-trimethyl-	C ₁₈ H ₃₆ O	268	2.556
5	11.688	Dodecanoic acid, 2-(acetyloxy)-1-[(acetyloxy)methyl]ethyl ester	$C_{19}H_{34}O_6$	358	0.776
6	12.436	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	2.705
7	12.880	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	2.115
8	14.528	11,14-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294	0.758
9	14.608	9,12-Octadecadienoyl chloride, (Z,Z)-	C ₁₈ H ₃₁ CIO	298	1.885
10	14.736	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296	22.733
11	14.961	Cyclopentaneundecanoic acid, methyl ester	$C_{17}H_{32}O_2$	268	0.709
12	24.476	Squalene	$C_{30}H_{50}$	410	14.469
13	26.166	δ- Tocopherol	C ₂₇ H ₄₆ O ₂	402	1.454
14	29.616	9,10-Secocholesta-5,7,10(19)-triene-3,24,25-triol, (3β,5Ζ,7Ε)-	C ₂₇ H ₄₄ O ₃	416	0.912
15	30.600	Ergosta-5,22-dien-3-ol,acetate, (3β,22E)-	C ₃₀ H ₄₈ O ₂	440	1.039
16	31.499	Farnesyl bromide	$C_{15}H_{25}Br$	284	19.457
17	31.841	β–Sitosterol	$C_{29}H_{50}O$	414	16.513
18	32.168	1-Heptatriacotanol	C ₃₇ H ₇₆ O	536	4.232
19	34.697	β-Amyrin	C ₃₀ H ₅₀ O	426	5.168

Table 2. GC-MS Phytochemical analysis of Cnidoscolus aconitifolius leaves.

sitosterol, 1-heptatriacotanol and β-amyrin as shown in Table 2. The I-gala-I-ido-octose has been reported to be used for the synthesis of higher sugar which are necessary for the production of drugs used to specifically facilitate learning or memory, particularly to prevent cognitive deficits associated with dementias (Jun et al., 2015). Another phytoconstituent identified is 10-methyl-E-11-tridecen-1-ol propionate, an alcoholic compound and have been reported to have antimicrobial properties (Mina et al., 2015). The presence of n- hexadecanioc acid shows that the plant can exhibit antioxidant, lubricant, nematicid, pesticide, hemolytic and 5-alpha reductase inhibition, flavor and antiandrogenic properties. Rency et al. (2015) also reported similar bioactivity on n-Hexadecanioc acid. δ - Tocopherol was also detected in the plant extract. Urooj et al. (2016) posited that the presence of vitamin-E in plant is an indication that the plant leaves have strong anti-oxidant and neuroprotective activities. 5- Tocopherol has been reported to exhibit antiageing, analgesic, anti-diabetic, anti -inflammatory, antioxidant and anti-leukemic activity (Prabhadevi et al., 2012). Squalene obtained from this study is suggested to be a triterpene having antibacterial, antioxidant, pesticide, antitumor, immune-stimulant and chemo preventive potentials (Kala et al., 2011; Dhanalakshmi and Manavalan, 2014). The compound squalene has also been reported to play a role in the synthesis of cholesterol, steroid hormones and vitamin D in human body (Rency et al., 2015). The phyto-component 9octadecenoic acid (z) have also been reported to

possess some biological activity such as antiinflammatory, anti-alopecic, haemolytic $5-\alpha$ reductase inhibitor, lubricant, antitumor, immune-stimulant, diuretic, antiandrogenic, antibacterial, antifungal, and lipoxygenase inhibitor activities (Ogunlesi et al., 2010; Omotoso et al., 2014a).

The mean body weight changes of rats administered aqueous and ethanol leaf extracts of C. aconitifolius is presented in Table 3. There was significant percentage weight reduction in rats administered 400, 600 and 800 mg/kg body weight of aqueous leaf extract of C. aconitifolius in a dose dependent manner. Chinyere et al. (2015) reported that weight reduction in experimental animal may be due to toxicity of the fed diet, unacceptability of diet by animals, indigestion and presence of non-nutritional factors in the diet. Result suggest that rats administered ethanol extract had significant percentage weight gain compared to the control (p<0.05). The significant weight gain observed with the rats administered ethanol leaf extract of C. aconitifolius suggest that the extract may not be toxic and the solvent may have extracted the bioactive ingredients more. This finding is in accordance with the reports of Kim et al. (2006) and Ebeye et al. (2015) who demonstrated significant increase in body weight of albino rats fed with C. aconitifolius ethanol leaf extract. The significant increase in body weight observed from this study could also be due to the fact that C. aconitifolius leaves contain valuable and viable nutritional constituents that are needed for growth, body repair and

Groups	Initial Body weight(g)	Final Body Weight(g)	Percentage Change in body weight (%)
Group I	120.78±1.48	135.95±1.42	12.56
Group II	121.13±1.23	134.78±0.69	11.26
Group III	115.45±1.04	125.70±0.69	8.87
Group IV	118.38±2.13	125.15±1.76	5.71
Group V	120.13±0.69	125.50±1.31	4.09
Group VI	119.95±0.95	132.50±0.75	10.46
Group VII	118.10±1.04	133.40±0.78	12.95
Group VIII	121.95±1.63	141.50±2.34	16.03
Group IX	117.18±0.48	141.93±2.33	21.12
LSD	1.8486	2.1542	

Table 3. Body weight effect of Cnidoscolus aconitifolius leaf extracts on body weight male albino rats(g).

n=5.Results represent mean of triplicate determinations ± standard deviation.

Table 4. Lipid profile of rats administered aqueous and ethanol leaf extracts of Cnidoscolus aconitifolius.

Group	Total cholesterol (mg/dl)	Triglyceride (mg/dl)	High density lipoprotein (mg/dl)	Low density lipoprotein (mg/dl)
Group I	83.56±0.66	91.10±0.74	30.32±0.99	71.46±0.89
Group II	77.82±0.14*	87.79±0.15*	35.86±0.05*	60.29±0.07*
Group III	76.96±0.61*	88.32±0.06*	35.08±0.10*	58.76±0.16*
Group IV	76.79±0.29*	84.16±0.62*	37.92±0.60*	54.96±1.31*
Group V	71.05±0.58*	83.77±1.13*	39.66±0.11*	48.88±0.62*
Group VI	68.81±0.31*	79.15±0.17*	41.13±0.00*	43.51±0.73*
Group VII	67.16±0.03*	79.02±0.09*	44.21±0.56*	38.75±0.37*
Group VIII	65.95±0.11*	76.11±0.01*	48.03±0.13*	33.14±0.78*
Group IX	64.23±0.35*	69.84±0.12*	48.78±0.21*	29.41±0.16*
LSD	0.018	0.013	0.144	0.032

Results represent mean of triplicate determinations ± standard deviation; n=5.

maintenance which may increase appetite resulting in increased food intake which ultimately may lead to increase in the body weights observed. This shows that *C. aconitifolius* ethanol extract could be effective in improving body weight loss. This report is in accordance with the findings of Mordi (2012) and Odokuma (2012) who posited increase in body weight of rats fed with *C. aconitifolius* leaves. This study revealed significant increase in body weight in the groups fed with ethanol extract compared to those fed with aqueous extracts. This could also be as a result of the absence toxic constituents in the ethanol extract which may have led credence to the observed weight gain.

Lipids have been reported to play important role in cardiovascular diseases not only by way of hyperlipidaemia and development of atherosclerosis but also by modifying the composition, structure and stability of cellular membrane (Chinyere et al., 2015). According to Mathew (2000), excessive lipids in the blood are considered accelerate the development of to atherosclerosis and are the major risk factor in

myocardial infarction. Anofi and Mutiu (2015) also posited that alterations in the concentration of lipids like total cholesterol, high density lipoprotein cholesterol and triglycerides can provide information on the status of lipid metabolism as well as predisposition of the animals to atherosclerosis. Cholesterol is an essential substance involved in many cellular functions, including the maintenance of membrane fluidity, production of vitamin D on the surface of the skin, production of hormones and possibly helping cell connections in the brain (Ugwu et al., 2011). Mohale et al. (2008) also noted that high levels of circulating cholesterol and its accumulation in heart tissue are well associated with cardiovascular damage. Results of total cholesterol showed significant decrease in total cholesterol level of rats administered C. aconitifolius leaf extract compared to control (p<0.05). This decrease may be attributed to bioactive constituents of C. aconitifolius leaf which may have promoted the actions of high density lipoprotein responsible for transporting cholesterol out of the blood. This may justify its local usage in management of heart related diseases.

Triglycerides accumulation in serum has been reported as one of the risk factors in coronary heart diseases (Mohale et al., 2008). Results also showed significant decrease in the level of triglycerides in rats administered *C. aconitifolius* leaf extracts compared to control (p<0.05). This suggests that *C. aconitifolius* leaf extracts may play important role in management of coronary heart diseases.

High density lipoprotein is an antiatherogenic factor which is important in the transport of cholesterol from cells to the liver where it is catabolised (Lacko et al., 2000). High density lipoprotein (HDL) results shows significant increase in the groups treated with C. aconitifolius leaf extracts compared to control (p<0.05). This may suggest that there was continuous export of excess cholesterol to the liver for excretion into the bile which may reduce the risk of atherosclerosis or coronary diseases. This increase in HDL may be attributed to the bioactive constituents of the plant and may substantiate the cardio-protective effects of C. aconitifolius leaf extracts. Low density lipoprotein is a known triggering factor for coronary occlusion (Ezekwe et al., 2014). Findings from the present study indicate marked reduction in low density lipoprotein level in rats administered C. aconitifolius leaf extracts compared to control (p<0.05). The decrease in serum low density lipoprotein may be understandable since according to Yakubu et al. (2008) a decrease in serum total cholesterol should normally result in a decrease in low density lipoprotein. This decrease in low density lipoprotein may be one of the prominent beneficial effects of consuming C. aconitifolius leaf extracts as low serum low density lipoprotein (LDL) have been reported to signify less risk to coronary heart diseases (Mohale et al., 2008). However, a more significant reduction in total cholesterol, triglyceride, low density lipoprotein and increase in high density lipoprotein was recorded from the groups administered ethanol extract relative to the groups administered aqueous leaf extract. This may further suggest the use of ethanol in extraction of the bioactive constituents of C. aconitifolius leaf in order to fully utilize its potential medicinal properties in management of diseases.

Conclusion

This study has revealed the bioactive components in *C. aconitifolius* leaf extracts and its potentials in reduction of cholesterol level. It is therefore suggested that ethanol may be a better solvent of extraction and adequate use of this plant leaf should be encouraged in order to maximize its medicinal efficacies.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

- Aberoumand A, Deokule SS (2009).Studies on nutritional values of some wild edible plants from Iran and India. Pakistan Journal of Nutrition 8(1):26-31.
- Akinpelu DA, Onakoya TM (2006). Antimicrobial activities of medicinal plants used in folklore remedies in South- West. African Journal of Biotechnology 5:1078-1081.
- Anofi OT, Mutiu IK (2015).Toxicopathological evaluation of hydroethanol extract of *Dianthus basuticus* in wistar rats. J. Evi-Bas. Complementary and Alternative Medicine 10(3):87-99.
- Araujo TAS, Alencar NL, Amorim ELC, Albuquerque UP (2008). A new approach to study medicinal plants with tannins and flavonoids contents from the local knowledge. Journal of Ethnopharmacology 120:72-80.
- Awoyinka OA, Balogun IO, Ogunnowo AA (2007). Phytochemical screening and *in vitro* bioactivity of *Cnidoscolus aconitifolius*. Journal of Medicinal Plant Research 1(3):63-65.
- Chinyere GC, Ikpoh JC, Osuocha KU (2015). Serum lipid profile and liver enzymes of rats fed *Lageneria sphaerica* (wild bottle gourd) supplemented diet. International Research Journal of Biochemistry and Biotechnology 2(2):28-36.
- Dhanalakshmi R, Manavalan R (2014). Bioactive Compounds in Leaves of *Corchorus trilocularis* L. by GC-MS Analysis. International Journal of PharmTech Research 6(7):1991-1998.
- Ebeye OA, Ekundina VO, Ekele CM, Eboh DEO (2015). The histological effect of *Cnidoscolus aconitifolius* aqueous leaf extracts on the archetecture of the ovary, testis and sperm cells of adult wistar rats. International Journal of Herbs and Pharmacological Research 4(1):7-17.
- Ezekwe AS, Elekwa I, Osuocha KU (2014). Hypoglycemic, hypolipidemic and body weight effects of unripe pulp of *Carica papaya* using diabetic albino rat model. Journal of Pharmacognosy and Phytochemistry 2(6):109-114.
- Fossati P, Prencipe L (1982). Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. Journal of Clinical Chemistry 28(10):2077-80.
- Hussein JH, Mohammed YH, Imad HH (2016). Study of chemical composition of *Foeniculum vulgare* using Fourier transform infrared spectrophotometer and gas chromatography-mass spectrometry. Journal of Pharmacognosy and Phytotherapy 8(3):60-89.
- Jun L, Stefan W, Chunlin X (2015). A review of bioactive plant polysaccharides: Biological activities, functionalization and biomedical application. Journal Bioactive Carbohydrates and Dietary Fibre 5(1):31-61.
- Kala SMU, Balasubramanian T, Tresina S, Mohan VR (2011). GC-MS determination of bioactive components of *Eugenia singampattiana Bedd.* International Journal of Chemical Technology Research 3:1534-1537.
- Lacko AG, Barter P, Ehnholm C, Van-Tol A (2000). International symposium on basic aspects of HDL metabolism and disease prevention. Journal of Lipid Research 41(10):1695-1699.
- Mathew JP (2000). Liver enzymes. In principles of health. White man publications. New York pp. 130-136.
- Mina M, Farida AB, Amal R, Abdel AE, Hassane M, Nourdinne B, Mohammed L (2015). Study on chemical components of ethanol extractives of *Dipcadis erotinum* (Hyacinthaceae) By GC/MS. Intern. International Journal of Current Research and Academic Review 3(9):134-139.
- Mohale DS, Dewani AP, Saoji AN, Khadse CD (2008). Antihyperlipidemic activity of isolated constituents *Lageneria* species in albino rats. International Journal of Green Pharmacy 2:817-827.
- Mordi JC, Akanji MA (2012). Phytochemical screening of the dried leaf extract of *Cnidoscolus aconitifolius* and associated changes in liver enzymes induced by its administration in Wistar Rats. Current Research Journal of Biological Sciences 4(2): 153-158.
- Mordi J, Uzuegbu U, Nwangwa E, Okunima A (2013). Effect of the dry aqueous leaf extract of *Cnidoscolus aconitifolius* on blood alcohol clearance in rabbits. Journal of Natural Sciences Research 3(5):91-95.
- Obichi EA, Monago CC,Belonwu DC (2015).Effect of *Cnidoscolus* aconitifolius (Family *Euphorbiaceae*) Aqueous Leaf Extract on Some

Antioxidant Enzymes and Haematological Parameters of High Fat Diet and Streptozotocin Induced Diabetic Wistar Albino Rats. Journal of Applied Sciences and Environmental Management 19(1):201-209.

- Odokuma EI (2012). Histological effects of alcoholic extract of Cnidoscolus aconitifolius on bone marrow biopsy in adult male Wistar Rats. Basic Sciences of Medicine 1(1):6-8.
- Olaleye MT (2007). Cytotoxicity and antibacterial activity of methanolic extract of *Hibiscus sabdariffa*. Journal of Medicinal Plants Research 1(1):9-13.
- Olayinka AO, Onoruvwe O, Lot TY (1992). Cardiovascular effects of the methanolic extracts of the stem bark of *Khaya senegalensis*. Phytotherapy Research 6(5):282-284.
- Oluwatosin AA, Adekunbi A, Ademola AO (2011). *Cnidoscolus aconitifolius* leaf extract protects against hepatic damage induced by chronic ethanol administration in wistar rats. Alcohol and Alcoholism 46(4):451-458.
- Omotoso AE, Eseyin OO, Suleiman M (2014a). Phytochemical analysis of *Cnidoscolus aconitifolius* (Euphorbiaceae) leaf with Spectrometric Techniques. Nigerian Journal of Pharmaceutical and Applied Science Research 3(1):38-49.
- Oyeyemi MO, Ajani OS (2014). Haematological parameters and serum testosterone of West African dwarf rams treated with aqueous extract of *Cnidoscolus aconitifolius* (Chaya). Journal of Medicinal Plants Research-Academic Journals 8(14):571-575.
- Prabhadevi V, Sathish S, Johnson M, Venkatramani B, Janakiraman N (2012). Phytochemical studies on Allamanda cathartica L. using GC-MS. Asian Pacific Journal of Tropical Biomedicine 2(2):550-554.
- Rency RC, Vasantha K, Maruthasalam A (2015). Identification of bioactive compounds from ethanolic leaf extracts of premna serratifolia L. using GC-MS. Journal of Biosciences 6(2):96-101.

- Ross-Ibarra J, Molina-Cruz A (2002). The Ethnobotany of Chaya (*Cnidoscolus aconitifolius*): A Nutritious Maya Vegetable. Economic Botany 56:350-364.
- Sofowora A (1993). Medicinal Plants and Traditional Medicines in Africa. Chichester John Wiley and Sons New York pp. 97-145.
- Sakpa CL, Okhimamhe AF (2014). Effects of aqueous leaf extract of Chaya (*Cnidoscolus aconitifolius*) on pituitary-gonadal axis hormones of male Wistar rats. Journal of Experimental and Clinical Anatomy 13(2):34-39.
- Sowmya S, Gopalakrishnan VK, Poornima K (2015). Enzymatic and non enzymatic antioxidant activity of Tabernaemontana divaricate R.Br. against DEN and Fe-NTA induced renal damage in Wistar Albino Rats. Journal of Applied Pharmaceutical Science 5(5):33-37.
- Ugwu CE, Olajide JE, Alumana EO, Ezeanyik LUS (2011). Comparative effects of the leaves of *Vernonia amygdalina* and *Telfairia occidentalis* incorporated diets on the lipid profile of rats. African Journal of Biochemistry Research Academic Journals 5(1):28-32.
- Urooj A, Fazal S, Muhammad S, Shehla A, Nisar A, Gowhar A, Khwaja F, Robert DES (2016). Passiflora incarnate attenuation of neuropathic allodynia and vulvodynia apropos GABA-ergic and opioidergic antinociceptive and behavioural mechanisms. Complementary and Alternative Medicine 16:77-83.
- Yakubu MT, Akanji MA, Oladiji AT, Olatinwo AO, Adesokan AA, Oyenike M, Yakubu RN, Owoyele BV, Sunmonu TO, Ajao SM (2008). Effect of *Cnidoscolus aconitifolius* leaf extract on reproductive hormones of female rats. Iranian Journal of Reproductive Medicine. 6:149-155.