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Effects of Datura Metel Leaves Extract on Blood Parameters and Lipid Profile Using Albino Rats as Model

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

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Original Research Article

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ABSTRACT

Datura metel is commonly found locally and it is readily available to the people, consequently, it is abused and deliberately used as poison. The aim of this study is to ascertain the effect of *Datural metel* through blood parameters and lipid profile when consumed through food intake using albino rat model. The leaves were extracted with ethanol and the phytochemical parameters determined. Different concentration of the extract were mixed with rat food and the food were fed to albino rats placed in four groups of five. The blood and lipid profile of rats were picked from each group determined at the end of week 1,2 and 3. Rats from each group was sacrificed and the blood was collected through cardiac puncture which was later analyzed for blood parameters and lipid profile. The results obtained from the first set of rats did not show any significant effect on the blood parameters and lipid profile, but the results obtained from subsequent sacrificed rats showed gradual increase in the values of the rate of haemoglobin (HB) 6.7 – 10G/DL, Red blood cells (RBC) $3.82 - 4.43 \times 10^6 \mu l$ and mean cell volume (MCV) 52.36 - 69.98FL. The results obtained for mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH) and high density lipoproteins (LDL) 27.2 - 20.2 mg/dl and cholesterol 79 -71 mg/dl. This study

suggests that the concentration of ethanolic extracts of *Datura metel* leaves have different active components that have diverse revamping effects on the blood parameters and lipid profile based on concentration of the extracts and duration of intake of the extract. This invariably portray that presence of *Datura metel* in food can enhance the quality of the food in the body and also maintain good cardiovascular wellbeing based on its concentration and timeline of consumption.

Keywords: Phytochemicals; lipid profile; datura metel; blood parameters; cardiovascular wellbeing.

1. INTRODUCTION

A lipid profile or lipid panel is a panel of blood tests that serves as an initial broad medical screening tool for abnormalities in lipids such as cholesterol and triglycerides. The results of this test can identify certain genetic diseases and can determine approximate risks for cardiovascular disease, certain forms of pancreatitis and other diseases [1]. The lipid profile typically includes low-density lipoproteins (LDL), High density lipoproteins (HDL), Triglycerides, Total cholesterol and the laboratory may also calculate very low density lipoproteins(VLDL) [2]. Quantitating lipoproteins is the best determinant of cardiovascular risk and thus particle concentrations are not only the best way to predict risk, but are also the best goals of therapy [3]). This definitely affirms the importance of lipid profile in confirming the presence and concurrently determining the toxic effects of foreign materials in the body system. Actually, the toxicity of Datura metel on the body system, precisely on the cardiovascular wellbeing can be obviously analyzed through the confirmation of the effectual changes instigated in the lipid profile by its substances.

Datura metel is an annual herb which belongs to the family of Solanaceae growing up to 3 ft. high. It is slightly furry, with dark violet shoots and oval leaves that are often violet as well. The seed capsule is covered with numerous conical humps and a few spines. There is evidence that Datura metel seeds have been used in ancient Indian medicine, modern Indian folk medicine, and Ayurvedic medical practices. The most common medicinal uses in these systems are for skin conditions, anxiety disorders and respiratory ailments, along with a litany of other conditions. The seeds are also sometimes used as a substitute for opium [4].

The preliminary phytochemical investigation performed on methanolic and hydroalcoholic extract of the plant dried seeds revealed the presence of alkaloids, tannins, cardiac glycosides, flavonoids, carbohydrates, amino acids and phenolic compounds [5]. Studies on chemical composition of the plant carried out by Indira, , [6] showed that it contained fat (14.72%), carbohydrate (20.73%). (51.22%), protein moisture (4.63%), ash content (5.14%), total sugar (5.63%), reducing sugar (2.65%), crude fibre (17.35%). Trace elements such as Calcium (174.0mg/100g), phosphorus (690.0mg/100gm), potassium 0.50, sodium 0.085, iron 16.8, zinc 2. 63, copper 6.9 and magnesium 390.0. Total saturated acids was 18.03% and total unsaturated fatty acids was 81.74%.

Datura toxins may be ingested accidentally by consumption of honey produced by several wasp species, including Brachygastra lecheguana, during the Datura blooming season [7]. From 1950 to 1965, the State Chemical Laboratories in Agra, India, investigated 2,778 deaths caused by ingesting Datura [8]. Typical findings in Datura poisoning are dryness of the mouth, thirst, flushing, fever, amnesia, urinary retention, decreased salivation. papillary dilation, hallucination, palpitation, delirium leading to coma, cardiac and respiratory arrest [9]. Study carried out by Freye, [10] showed that overwhelming majority of those who described their use of Datura found their experience extremely unpleasant both mentally and physically dangerous. However, Fuller [11] reported that anthropologists found some indigenous groups with a great deal of experience and knowledge of Datura use the plant for recreational purposes. In Nigeria, it was reported that the plant is abused by adding its decoction of leaves to drinks as a substitute for marijuana because it is relatively cheap and readily available [12]. The leaves and seeds are also locally used as hallucinogen, antispasmodic and bronchodilator [13,14].

The neuropsychological effects of aqueous extracts of leaves and seeds of the plant were studied by Abenna et al. [15]. They found that the plant has antidepressant activity at low doses.

Oral administration of the drug to dogs at a dose rate of 0.6, 1.2, 1.5, 2 and 2.4 g/kg respectively showed a graded dose response relationship. Das et al., [16] in their study, showed that all parts of the plant were poisonous because of the presence of toxic tropane anti-cholinergic which caused neural alkaloids toxicity. Histological evaluation of the organs showed decrease in organ weight, circulatory disturbance, necrotic changes in the liver architecture with increase of serum alkaline phosphatase, serum glutamic - oxaloacetic transaminase and glutamyl pyruvic transaminase in liver and heart [17].

2. MATERIALS AND METHODS

2.1 Collection and Preparation of Plant Samples

The plant was collected in Orogun area of Ibadan, Oyo State, Nigeria in March 2019 and authenticated at the Herbarium of Nigeria Natural Medicine Development Agency, Lagos. It was washed and air-dried for three weeks in the industrial Chemistry Laboratory section of the University of Ibadan. The dried sample was chopped into smaller pieces grinded in electric blender till it was transformed into powder which was stored in a clean, dry and air tight polythene bags.

2.2 Thin Layer Chromatography

This was performed on pre-coated silica gel G plates of size 10 x 10 (E Merck, Darmstadt, Germany) for characterization of the extracts in order to determine the number of components present in each of the extracts. The TLC plate was cut into different sizes of 6cm by 2cm for those requiring 2 spots and 6cm by 2.5cm for 3 spots. Several solvent mixtures were prepared for proper resolution of the components present which includes ethylacetate/hexane (6:4), ethylacetate/hexane (7:3). ethyacetate/hexane (8:4), methanol/hexane methanol/hexane petroleum (6:4), (1:1), ether/ethylacetate (7:3).

2.3 Phytochemical Analysis

Preliminary phytochemical analysis was carried out to determine the presence of alkaloids, glycosides, terpenoids, steroids, flavonoids, reducing sugars, triterpenes, phenolic compounds and tannins.

2.4 Fourier Transformation Infra-red Spectroscopy (FT-IR)

The FT-IR analysis was carried out by placing the extract in the sample chamber of the equipment and the spectra were recorded in the range of 3600 – 600cm⁻¹ on Nicolet Avatar 330 FT-IR Spectrophotometer.

2.5 Experimental Animals

Wistar rats [18] weighing between 80-100g were obtained from the Experimental Animal Unit of the Faculty of Veterinary Medicine of the University of Ibadan. They were kept in well ventilated rat cages with free access to water and feed and were left in this environment for two [2] weeks to acclimatize [19]. The feed administered to the rats were mixed with varied concentration of the ethanolic Datural metel extract ranging from 600mg/kg, 900mg/kg and 1200mg/kg. The rats were categorized into four groups of five rats in concordance with the varied concentration of the extract. A rat from each group were sacrificed weekly and the blood collected through cardiac puncture to determine the lipid profile and haematological examination carried out at the pathological laboratory of Faculty of Veterinary Medicine, University of Ibadan.

2.6 Haematological and Lipid Profile Examination

Haematological examination [20] involved placing the haemocymeter on a flat horizontal surface and applying firm pressure using both index finger. This is used to slide the coverglass into position over the ruled counting areas. RBC dilution was achieved by inversion 10 -12 times. The RBC was taken up in a capillary tube and the counting chamber was filled by holding the capillary or pipette at an angle of 45 degrees and the tip was slightly touched against the edge of the chamber and coverglass. Thereafter, the counting chamber was placed on the microscope stage and 2 minutes was allowed to elapse before the count commences so as to allow the cells to settle. A high dry objective was used either the X8 or X10 evepiece and the cells contained in 80 of the 400 small squares were counted. All cells touching the centreline bordering the bottom and left hand side of each group of 16 squares were included in the count.

Red blood cells calculation: Number of cells x depth x dilution x area.

2.7 Cholesterol Examination

This was carried out for the quantitative in-vitro determination of cholesterol in serum and plasma. Mixing and incubation was carried out for 10 minutes at +20 to $+25^{\circ}$ C and for 5minutes at 37° C. The specimen was inserted into the RX Monza flowcell holder and 'Read' was pressed within 60 minutes [3].

2.8 Tryglycerides Examination

This was carried out for the quantitative in-vitro determination of tryglycerides in serum and plasma. The same procedure for cholesterol determination was repeated for this process [3].

3. RESULTS AND DISCUSSION

3.1 Percentage Yield of Sample

The percentage yield was 9.24%. The extract obtained was oily and slurry paste with very dark green colour and a very good pleasant odour. The yield obtained was higher than the reported yield of 4.85% by Okwu and Igara, [21] and Alabri et al., [18] 6.21%. The better yield recorded may be due to the fact that the sample used was exhaustively extracted. Also, the geographical location of collection and time/season of collection as well as the purity may also be responsible for the higher yield.

3.2 Thin Layer Chromatography Elucidation

Five fractions were spotted on the pre-coated silica gel – G plates. The most prominent spotted TLC plates were developed in a tank that contained a mixture of ethylacetate/hexane (7:3). The Rf of the five spotted spots are as follows: 0.09, 0.3, 0.69, 0.75 and 0.8 respectively.

3.3 Phytochemical Elucidation

As shown in Table 1, alkaloids, flavonoids, triterpenoids, phenolic compounds, tannins, saponins, tripenoids and steroids were the phytochemicals detected in the medicinal plant. Evans [22], attested to the presence of alkaloids in the plant. In their own report, Siddiqui and Al-Mailay, [23] confirms the presence of glycosides, tripenoids and steroids when they worked on the

plant. Presence of tannins was ascertained by lyengar in [24].

Table 1. Results of phytochemical screening

Alkaloids	+
Tripenoids and steroids	+
Glycosides	+
Flavonoids	+
Reducing sugar	-
Triterpenoids	+
Phenolic compounds	+
Tannins	+
Saponins	+

+ = presence, - = absence

3.4 FT-IR analysis of Ethanolic Crude Extract

There were numerous IR signals which was recorded which tentatively gave the idea of the organic compounds that may be present in the crude extract. The IR within the range of 1069.60 - 1219.20cm⁻¹ suggests C-O_{str} which probably ether, 3433.60cm⁻¹ suggests N-H_{str} which may indicate presence of an amine, 3605.60cm suggest O - H_{str} which probably depicts the presence of alcohol, 2952.80cm⁻¹ which may suggest the presence alkene. 2870.40cm probably indicate the presence of $C - H_{str}$ of an alkane, while 2363.40cm⁻¹ may indicate the presence of a nitrile. The spectra also showed 2097.0 cm⁻¹ which suggests C=C_{str} of an alkyne. 1637.60 cm⁻¹ suggests C=C_{str} of alkene. 1219.20 cm⁻¹ suggests C-N_{str} of an amine while 653.60 cm⁻¹ suggests the presence of an alkyl halide (Table 2).

 Table 2. IR signals of ethanolic crude extract of Datura metel leaves

IR Wavenumber	Functional group
3605.60	O-H _{str}
3433.60	N-H _{str}
2952.80	$C-H_{str}(sp^2)$
2870.40	C-H _{str} (sp ³)
2362.40	C≡N _{str}
2097.60	C≡C _{str}
1637.60	C=C _{str}
1406.40	C=C _{bend}
1219.20	C-N(_{amine})
1069.60	C-O _{str}
653.60	C-CL _{str}

3.5 Haematological Analysis

It was observed from the results of the readings in Table 3 that the changes of the rate of haemoglobin (Hb) from control to group D did not follow markedly the gradual and successive

grps	PCV%	Hb g/dl	Rbc x10 ⁶ µl	Wbc x10 ³ µl	platelet	Lym %	Neat%	Max%	Eo%	MCV	МСНС	МСН
control	47±0.02	15.6±0.06	7.63±0.07	3700	90000	72±0.03	26±0.05	1±0.03	1±0.03	61.60±0.03	33.19±0.02	20.45±0.06
600 mg/kg	34±0.04	11.3±0.01	5.36±0.04	3600	85000	65±0.03	32±0.02	2±0.02	1±0.02	60.39±0.03	33.24±0.04	21.08±0.01
900 mg/kg	51±0.01	16.8±0.03	9.71±0.02	4000	76000	67±0.02	31±0.02	2±0.01	0±0.05	55.20±0.04	32.94±0.02	17.30±0.01
1200 mg/kg	41±0.03	13.5±0.02	6.77±0.01	3250	81000	63±0.04	32±0.02	2±0.02	3±0.02	60.56±0.01	32.93±0.04	19.94±0.02

Table 3. Haematological parameters of albino rats fed with feed mixed ethanolic extract of Datura metel leaves at 2 weeks

Each value is the mean of three measurement ± SD

Table 4. Haematological parameters of albino rats fed with feed mixed ethanolic extract of Datura metel leaves at 3 weeks

grps	PCV%	Hb g/dl	Rbc x10 ⁶ µl	Wbc x10 ³ µl	platelet	Lym %	Neat%	Max%	Eo%	MCV	МСНС	МСН
control	20±1.02	6.7±0.05	3.82±0.3	4450	120000	64±0.07	34±0.02	1±0.1	1±0.05	52.36±0.02	33.50±1.02	17.54±0.00
600 mg/kg	24±0.08	8.4±0.02	4.26±0.02	4000	12900	67±0.05	29±0.00	2±0.3	2±0.2	56.34±0.06	35.00±0.05	19.72±0.05
900 mg/kg	39±0.06	12.6±0.03	6.47±0.05	3800	11600	66±0.06	31±0.04	1±0.02	2±0.08	60.28±0.07	32.31±0.04	14.89±0.02
1200 mg/kg	31±0.2	10.0±0.02	4.43±0.01	4600	13000	74±0.05	24±0.03	3±0.04	3±0.03	69.98±0.03	32.58±1.02	22.80±0.01

Each value is the mean of three measurement ± SD

Table 5. Concentration of lipid profile parameters (mg/dl) of albino rats fed with feed mixed with ethanolic extract of Datura metel leaves at week 2

grps	Total protein g/dl	Alb g/dl	Glob g/dl	A.G Ratio	AST μΙ	ALT μl	ALΡ μl	Trig mg/dl	Chol mg/dl	HDL mg/dl	LDL mg/dl
control	7.0±1.2	3.1±0.3	3.9±0.2	0.7±0.1	46±0.2	33±1.1	120±0.5	50±1.5	63±0.5	28±1.5	25±1.05
600 mg/kg	6.1±0.5	2.4±0.1	3.7±0.1	0.7±0.2	45±0.2	33±0.4	125±0.2	43±1.2	65±0.2	32±1.2	24.4±0.5
900 mg/kg	7.2±0.4	3.1±0.2	4.1±0.2	0.7±0.1	41±0.1	30±0.2	106±0.2	34±0.6	40±1.5	20±0.4	13.2±1.5
1200 mg/kg	7.3±0.6	3.2±0.3	4.1±0.1	0.7±0.1	42±0.2	31±0.5	120±0.1	31±1.3	42±0.8	19±0.2	24.4±0.3

Each value is the mean of three measurement ± SD

grps	Total protein g/dl	Alb g/dl	Glob g/dl	A.G Ratio	AST μΙ	ALT μΙ	ALΡ μl	Trig mg/dl	Chol mg/dl	HDL mg/dl	LDL mg/dl
control	8.4±0.3	3.7±0.2	4.7±0.2	0.7±0.2	42±2.4	31±0.5	124±1.3	44±0.7	71±1.8	42±0.5	27.2±1.2
600 mg/kg	7.8±0.2	3.6±0.1	4.2±0.3	0.8±0.1	48±1.3	35±0.3	112±0.8	38±1.6	66±0.7	32±1.7	26.5±0.2
900 mg/kg	6.5±0.1	2.9±0.1	3.6±0.2	0.8±0.2	46±1.4	35±0.4	106±1.7	40±0.4	65±0.2	32±0.2	25±0.6
1200 mg/kg	7.7±0.1	3.2±0.2	4.5±0.1	0.7±0.1	42±0.5	31±0.6	124±1.3	44±1.2	71±1.5	42±0.2	20.2±1.9

Table 6. Concentration of lipid profile parameters (mg/dl) of albino rats fed with feed mixed with ethanolic extract of Datura metel leaves at week 3

Each value is the mean of three measurement ± SD

changes in value in respect to the gradual increase in concentration of the plant extract. However in Table 4, it was observed there was gradual and successive increase in the rate of haemoglobin (Hb) from control to the other groups in terms of ascending concentration of Datura metel leaves extract but this trend was negated in group D as the rate of haemoglobin was seen to have declined unexpectedly. This absurd difference is presumably sequel to the lower quantity of feed subconsciously eaten by the rats in the group which tends to invariably reduce the intake of the extract. Comparing the results obtained for changes in the red blood cells, it was observed in Table 3 that the results do not follow markedly gradual and successive changes in value in respect to the gradual increase in concentration of the plant extracts mixed with the feeds that are fed to the rats but in Table 4, there was gradual and successive increase in the values of RBC from control to the other groups in terms of ascending concentration of extract which was also negated in group D. The pattern of changes in the values of Mean Cell Volume (MVC) shows that it takes considerable duration of time before the extracts of the leaves of the plant could instil its effect on the MCV as there was no significant gradual and successive changes in week 2 of the experiment compared to what was obtained in week 3. The changes observed for the Mean Corpuscular Haemoglobin Concentration (MCHC) in Tables 3 and 4 were uncoordinated and irregular compared with successive gradual increase in concentration of the extracts of the plant. The Mean Corpuscular Haemoglobin (MCH) also followed the same trend observed for the previous parameters as shown in Tables 3 and 4.

3.6 Lipid Profile Parameters

As shown in Tables 5 and 6, the changes in high density lipoproteins (HDL) indicated decrease as the concentration of the feed was increased during two weeks of feeding but at three weeks of feeding, the values obtained for 600 mg/kg and 900mg/kg were the same which probably indicated that there was not much effect but there was marked increased at 1200 mg/kg. However, values obtained for Low density lipoproteins (LDL) at two weeks showed there was no marked changes when the feed concentration was increased but at three weeks (Table 6), there was gradual and successive decrease in LDL which indicated that it took considerable duration of time before the effect of the extract can be felt. Same trend was

observed for the cholesterol levels in table 5 where the values obtained indicated no clear cut changes in the cholesterol but in Table 6, there was gradual and successive decrease in the value from control to the other groups in terms of ascending concentrations of extracts. It was also observed that the values obtained for other parameters in the lipid profiling are the same.

4. CONCLUSION

It can be concluded that there was no significant and meaningful changes in the values obtained for blood parameters and lipid profile, this indicate that the effect of the extract of the plant is not instantaneous, rather it takes time before it can instil its effect on the body. However at three weeks, the effect was very visible and significant. It is very important to note that measurement of blood parameters indicate the health status which are diagnostic for certain diseases such as anemia, leukemia and detection of the presence of inflammation [25]. Decrease in the number of red blood cells may indicate anemia while absurd increase in red blood cells may indicate polycythaemia [25]. Cholesterol is thought to amplify accelerate atherosclerosis and ischaemic stroke. It has been proposed that cholesterol particularly LDL cholesterol which accounts for about 60% total cholesterol in the circulation, is taken up by macrophages [26] hence the consecutive decrease of cholesterol level by the extracts at different concentrations showed that the plant has no noxious effect in the body and that it possesses positive cardiovascular activity.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation, but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

ETHICAL APPROVAL

The experimental procedures carried out in this study were in compliance with University of Ibadan ethics committee for the care and use of laboratory animals.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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