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Nutritive properties and phytochemical analysis of Thorn Apple "Gegemu" (*Datura metel*).

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Abstract
This experiment reports the phytochemical properties of seeds of a non-leafy vegetable Datura metel "gegemu" obtained from a major dump site in Ekiti State. The samples were analyzed for proximate and antioxidant composition and the following results were obtained: Protein value (12.59%), Ash (7.23%), Moisture content (15.54%), carbohydrate (40.00%). This vital but non-edible egg plant was found to be rich in manganese, copper and zinc. Hence, the egg plant can be utilized for medicinal (traditional) purpose.
Keywords: Antioxidant, *Datura metel*, Proximate, Phytochemical Analysis

Introduction:

Non-leafy vegetable are very good source of nutrient in our food intake in Nigeria, although they are seasonal, they are either enjoyed cooked or raw as the case may be. *Datura* is a genius of nine species of poisonous vespertine flowering plants belonging to the family of solanace they are known as "angels trumpets" sometimes sharing that name with the closely related genus Brugmansia and commonly known as DATURAS (Wikipedia 2015). This plant is widely distributed in West Africa, North Africa Restricted in some advanced countries like United State, Mexico but the highest species diversity occurs in Tunisia in Africa.

The Neo-Latin name Datura is taken from Hindi dhatura Jimson weed also known as white thorn-apple. In Indian it is called Kanak and Unmatt. In Mexico it is called toloache.

In India *Datura metel* is attributed to a poisonous and aphrodisiac species often used in little quantity as medicine from the ancient times and also in rituals and prayers. (preissel *et al,* 2002). It is also used to treat rheumatic aches and pains.

Datura species can change in size of plant, leaf and flower depending on the location. This same species can grow in half-shady, damp arrears and develop into a flowering bush which can be as tall as half the height of a human being, it can also grow in a very day location into a thin plant not much more than an ankle-high with tiny flowers and few miniature leaves. (Lester R.N *et al* 2006). *Datura* species are usually planted annually from the seed produced in spiny pods, in containers or outside. The plants have the ability to reseed themselves when planted on large areas and can become invasive. They are susceptible to fungi in the root area hence, no need of manure or composite fertilizers.

Datura metel contain alkaloids in their seed and flowers, they have been used or poison for ages. Although, the level of toxins present in the plant may vary due to certain factors like, age of the plant, soil where it is grown and the local weather conditions. There have been case of modern users ingesting datura species which either resulted into their death or serious illness. (AHTP, 2006)

In history no other substance has received as many severely negative recreational experience report is datura has the vane majority of those who have used datura find their experience extremely unpleasant both mentally and often physically dangerous. (ATHP, 2006).

This egg plant is short – leaved perennial plant which is often cultivated as annual growing 40 to 150cm tall with large coarsely lobed leaves 10 to 20cm and 5cm broad. The flowers are white and purple with a three lobed corolla and yellow stamens (ATHP, 2006).

The fruit is greenish in colour but very torny and it is less than 4cm in diameter it is a wild plant. The fruits contain numerous small, soft seeds. (AHTP, 2006). In the carribean it is used as strong herbal bread given to animals. In Chumash it plays an essential role in initiation ritual upon reaching puberty of young boys and girls. A cold water extract of datura is given them is drink and this send them on a visionary, deep hallucinatory sleep. In Nigerian, and California serves as snake repellant, anesthetic for setting bones to treat bad bruises, wounds to freshen the blood and is treat hemorrhoids. (Africa herbal topical plants, 2006).

Materials and methods

Sample preparation

The gegemu used for this research work were freshly plucked from refuse dump Ado- Ekiti and Ilawe -Ekiti while some were plucked fresh from Ikere Ekiti near a refuse dump. They were washed, rinsed with distilled

water and air dried.

The sample were further chopped into smaller pieces oven dried at 50°C, An electrical blender that is both fat and free is used to grind the dryed sample to powder or a mortal and pestle was used where applicable.

The powder sample were sieved with a sieve 1 mm of moderate mesh size and later stored in a clean, dry and air tight polythene bags and taken to the laboratory for analysis.

Proximate Analysis

The proximate analysis is the most widely used method of analysis for expressing the overall nutrient quality. It measures the quantity of the nutrients present in a food rather than the quality. Moisture content, crude fat and crude fibre were determined in accordance with the official methods of the Association of official Analytical chemists (AOAC, 1999).

The analysis consists of Ash content, crude fibre, crude protein, moisture content and lipid (fat) content determinations. The carbohydrate portion may be determined by subtracting the total percentage of al the aforementioned five analyses from one hundred (100) hence, carbohydrate by difference is obtained.

Phytochemical analysis

A portion of each of the extracts used was subjected to standard chemical test for the detection of saponins, tannins, anthraquinones and combine anthraquinones using the methods described by Harbones (1973) , Van-burden and Robinson (1973), Odebiyi and sofowora (1978)

Moisture Content Determination.

Procedure: 5g of the sample was weighed into a previously weighed Petri dish W_1 and kept in the oven for 5hrs at 50^0-60^0c. The Petri dish containing the sample is removed from the oven and placed in the desiccators so as to cool down. There after, the weight is quickly, taken with minimum exposure to air W_2. The procedure is repeated until a constant weight W_3 is reached. (Odebiyi, A. and Sofowora, A. E. 1978).

Calculation: the loss of weight during drying is equal to the moisture content of the sample.

$$= \frac{W_2 - W_3}{W_2 - W_3} \times 100$$

Where W_1 = Weight of dried Petril dish

$W_2 = W_1$ + 5g of the sample after drying

W_3 = the constant weight reached after drying procedure was repeated.

Ash (MINERAL) and Organic Matter Determination.

Procedure: 1g of the sample is weighed into a crucible which has been previously weighed N.B weights are recorded to the nearness 0.001g. The sample in the crucible is then placed in a cool muffle furnace and the temperature ramp to 500° over a period of 4hrs. The sample is allowed to cool down and transfer into the desiccators before the final weighing. Odebiyi, A. and Sofowora, A. E. (1978

Calculation

$$\% \text{ Ash} = \frac{W_3 - W_2}{W_1} \times 100$$

WhereW_1 = Weight of sample

$W_2 = W_1$ + Weight of crucible before heating

$W_3 = W_1$ + Weight of crucible after heating

Lipid Content Determination.

Procedure: 5g of the dry sample is weighed into fat-free extractor thimble, which has been previously dried and weigh (W_1). The thimble is plug tightly with cotton wool and weighed again (W_2). The timble was placed in the extractor and the solvent, (methanol) was added into the round bottom flask up to about two third full.

The consider was then replaced and the joints were made air tight and the set-up was placed on the heating mantle. The reflux was allowed to take place until the extraction is complete.

There is a complete extraction if the tip of the tube becomes clear and the soxhlet apparatus is detached, the thimble and the cotton were rinsed into the flask, with the aid of a methanol solvent.

The thimble was finally transfers into a fat free beaker and the whole was placed in an oven at 50°c and dried untile a constant weight is obtained. The thimble was then cooled in a desicccators and weight (W_3)

Caculation

$$\% \text{ fat} = \frac{\text{Weight of sample loss}}{\text{Weight of sample}} \times \frac{100}{1}$$

Where W_1 = Weight of thimble dried in oven
W_2 = Weight of thimble that was plugged with cotton wool
W_3 = Weight of thimble cooled in a desiccators

Determination of Nitrogen and Phosphorus

3.5g of selenium powder was added to 1 litter of sulphuric acid inside a container (sturdy pyrex glass). Heat is applied with the use of a hot plate at 280°c until the solution is clear. After the selenium has dissolved the hot plate was turned off and allowed to cool. (AOAC, 1999).

Nitrogen/Phosphorus Stock Solution

4.714g $(NH_4)_2$ SO_4 was added into 100ml volumetric flask and 0.43g of KH_2PO_4 was poured, there after both chemicals is oven dried at 105°c before weighting them. The salts were diluted to the mark. This solution then contains 10,000ppmN and 1000ppmP.

(1) 0.0200g of dry sample was weighed into 50ml digestion tube and 2.5ml of the H_2SO_4/FE mixture to each tube to s blanks to be used for standards.

An aluminum block is placed on the hot plate and heat at approximately 20°c until the samples fumes.

Secondly, the tubes are removed from the hot plates and allowed to cool down for 10mins. Aml of 30% H_2O_2 was carefully added to the samples and standardized. After the reaction as subsidize an additional 2ml H_2O_2 is added.

Thirdly, the tubes were replaced on the plate and a heavy 15ml glass vial on top of each tube and heated to 33°c. the tube was left until a clear solution is observed usually for about 2hrs. The yellow tint of the samples appears when the digest as completed.

Lastly, the samples were cooled and 0.200, 0.400, 0.600 and 0.800ml of the N/P stock solution was added to 5 standard solutions respectively. The samples and standard were diluted to 50ml mark. (AOAC, 1999).

Determination of Nitrogen and Phosphorus.

(1) Murphy – Riley (M – R) stock solution: 140ml of concentrated H_2SO_4 was slowly added to one liter of distilled water, thereafter, 12.0g of ammonium molybdate and 0.290 of antimony potassium tartarate. Dilution of the entire mixture was done distilled water to 2 liters. (AOAC,1999)

Crude Protein Determination.

Appartus: kjeldal digestion flask, pipette (25mls and 5mls). Burette (50ml), conical flask (100ml) micro-kjedahl distillation unit, volumetric flask (250ml) and beakers. (AOAC, 1999)
Reagent used: kjedahl tablet, conc. H_2SO_4 and 0.1M HCL

Preparation of reagents

50% NAOH (1.25M)
50g of sodium hydroxide pellets was accurately weighted and dissolved in distilled water and was further diluted to 100ml with distilled water
0.1M HCL
1M solution of HCL contains 36.5g in 1000cm^3 of distiller water.
0.1M solution will contain $\frac{36.5 \times 0.1}{4}$ in 1000cm^3 of distilled water
0.1M solution of HCL= 36.5 x 0.1 in 250cm^3 of distilled H_2O
Vol of conc. HCL needed for 0.1M in 250cm^3

$$= \frac{36.5 \times 100 \times 1.0}{4 \times 36.5 \times 1.18}$$

= 2.119mls
=36.5% purity
Density of conc. HCL = 1.18glcm^3

Procedure:

2g of the samples were weighed into a macro-kjeldahal flask with a tablet of kjeldahl catalyst and added to 40ml to nitrogen free conc. H_2SO_4. The mixture was heated until a clear solution is obtained. The digest was cooled thoroughly and was transferred to 250ml volumetric flask and made up to the mark with distilled water. The

distillation procedure involved the liberation of NH_3 by NaOH 40% w/w . The NH_3 was trapped in boric acid solution and titrated with standardized HCL.

Calculation

The nitrogen content of the sample was calculated by weigh from the expression.

$$\% \text{ of crude } N_2 = \frac{1.4 \times 10^{-4} \times \text{vol. of flask} \times 100 \times \text{Ave. Titer}}{\text{Dil. Factor} \times \text{mass of digested sample}}$$

To calculates the amount of protein in the sample.
The amount of Nitrogen was multiplied by the factor 6.25
Therefore,

$$\% \text{ protein} = N \times 6.25$$

Determination of Metals

Atomic Absorption Spectrophotometry

A sample of 4g was weighed and ashed in a muffle furnace. The standard stock solution of 0ppm- 10ppm was obtained; and a relationship was obtained between the concentration and absorbance. The sample was injected and automatically prints out the final output in part per million (ppm). That is the absorbance has been converted to ppm by using the already stored equation for each element. (Asaolu, 1995)

$$\text{Calculate of } \% M = \frac{PPM \times \text{Dil. Vol.} \times 0.0001}{\text{Weigh of sample (g)}}$$

PPM is in Mg/ml
Dil. Volume is 33ml
Weight of sample is 4g

Total Phenolic compound Determination.

Reagents: diethylether, ammonium hydroxide, aryl alcohol and distilled water.

Procedure: the sample was fast defatted. The free fat sample was boiled with 50ml of either for 15mm, 5ml of the extraction was pipette into a 50ml flask. Their 10ml of distilled water was added into a 50ml flask. Then 210ml of distilled water was added 2ml of ammonium hydroxide solution and 5ml of conc. Arylalcohol were also added. The samples were made up to the mark and left to react for 30mins. The colors charges develop, and this was measured at 505nm. Odebiyi, A. and Sofowora, A. E. (1978

Cacium, Magnesium and Potassium Determination. (AOAC 1999).

1. **LA/C^2 dilution solution:** dissolving 8.0g of $Lacl_3\,7H_2o$ and 4.4ml CsCL in 2 liters deionized or distilled water.
2. **Mixed stock solution:** in a 100ml volumetric flask, 1000ppm stock solution was added k, 16.0ml, Ca, 4.000ml and Mg 2.00ml then dilute to the mark with La/Cs dilution solution
3. **Standards:** To 525ml vials, add 2ml of the zero blank (step four in the digest procedure). Then dilution with 18ml of la/Cs solution from the respective vials was made and 0, 0.500 and 2.000ml of the mixed stock solution was subtracted. AOAC, 1995
4. **Sample:** the diluted 1.00ml of each sample, each with 9.00ml of La/Cs solution was made and the samples were read and standards too on the AAS (Asaolu, 1995).
   Calculation: Standard represents simples with the following concentration.
   Calcium 0, 0.250, 0.500, 0.750 and 1.00%
   Potassium 0, 1.00, 2.00, 3.00 and 4.00%
   Magnesium 0, 0.125, 0.250, 0.375 and 0.500%

Phytochemical Analysis.

Samples were screened for alkaloids, saponin, tannins, and combine anthroquinones. Odebiyi, A. and Sofowora, A. E. (1978).

Extracts preparation

5g of dried samples was boiled with about 100ml of distilled water for about 1 hour. This was filtered and the filtrate was used for the analysis.

Water extract

5g of dried sample was boiled with about 100ml of distilled water for about 1 hour. This was filtered and the filtrate was used for the analysis.

Test for Alkaloids
2ml of each of the water extracts was used for each of the following test; mayner's, wagner's, draggerdloff's piuric acid and tannic acid tests. Turbidity or precipitation indicated the presence of alkaloids. Odebiyi, A. and Sofowora, A. E. (1978).

Test of Saponins
1g of dried sample was shaken vigorously with water. Frothing was taken as preliminary evidence for presence of saponin. Odebiyi, A. and Sofowora, A. E. (1978).

Test for Tannin
2ml of 5% fed in water was added to 1ml of the water extract. A blue/black coloration was taken an evidence for the presence of tannin. Odebiyi, A. and Sofowora, A. E. (1978).

Test for Anthroquinones
A free anthroquinones 5g of each sample was extracted with 20ml of benzene and concentrated to 5ml of 10% NH_4OH solution was added to the filtrate and the mixture shaken vigorously. The presence of a pink-red color in the ammoniacal phase indicated the presence of free anthroquinones. Odebiyi, A. and Sofowora, A. E. (1978).

Combined Anthroquinones
5g of each sample was extracted with 20ml of 1% HCL for 3 hours, filtered hot and concentrated to 5ml. the filtrate was shaken with 5ml of benzene 3ml of NH_4OH was added to the benzene layer. A pink/red color in the ammoniacal phase indicated presence of anthroquinone derivatives. Odebiyi, A. and Sofowora, A. E. (1978).
Where v = titre value

5g of the sample was blend with 100ml of distilled water (free from lumps). The mixture was filtered.

10ml of the filtrate was taken into 100ml standard flask was made up to the mark.

10ml of the resultants solution was pipette into a conical flask and titrated with the standard indophenols solution (Y).

10ml of sample solution = $\frac{Y \times 0.05}{V}$

100ml of sample contained 'K' mg of ascorbic acid.
100g of sample contained 20 'K' mg of ascorbic acid.
In 100g sample =20 'K' mg ascorbic acid.

$$K\ (mg) = Y \times \frac{0.05}{V} \times \frac{100}{10} \times \frac{100}{W}\ mg/100g$$

Result and discussion
Proximate composition of dried *Datura* using "oguntona 1998". All values are reported in percentage and values are reported in triplicates, but the mean of these values were used.

The below shows the proximate composition of dried sample of Datura Species in triplicate

Table 1. Proximate composite in percentage.

S/N	Sample	Ash	Moisture	Protein	fat
1	Datura mete	7.23	15.54	12.59	2.35

Table 2 : proximate composition in percentage

S/N	Parameters	Ash	S.T.D	CV
1.	Total Ash	7.23	8.16×10^{-3}	7.00×10^{-1}
2.	Moisture content	15.54	8.16×10^{-3}	5.25×10^{-2}
3.	Protein content	12.59	8.16	6.48
4.	Fat	2.35	3.47	8.16×10^{-1}
5.	Crude fibre	22.29	9.70	3.37
6.	Carbohydrate	40.00	0.59	5.99

Table 3: Mineral composite of *Datura mete* in part per million (ppm)

S/N	Sample	Na	Mn	Cu	Fe	Pb	Ni	Zn	Cr
1	*Datura mete*	432.29	19.47	1.11	7.89	0.07	0.06	4.69	0.03

S/N	Sample	Co	Ca	Mg	K
1	*Datura mete*	0.41	2.80	2.50	1.56

Table 4: Result for Phytochemical Composition

S/N	Sample	Combine Anthroquinine	Alkaloid	Saponin	Tannin	Anthroquinone
1	*Datura mete*	–ve	+ve	+ve	+ve	-ve

Key: +ve means present
-ve means absent

Table 5: Percentage Phenol in Mg/100g of Samples

S/N	Sample	Ml used	ABS	Grade reading	% phenol	Phenol mg/100g
1	*Datura mete*	50	0.01	3.81	0.04	0.08

Table 6: Vitamin C Composition

S/N	Sample	Vitamin C	S.T.D	CV
1	*Datura mete*	7.49	0.57	7.16

Discussion

The result of the proximate analysis shows that the percentage Ash of *Datura* is 7.23%. For moisture content *Datura* has 15.54% which is very high. *Datura* percentage protein is 12.59%, while it has 2.35% fat.

Datura mete has 2.80% Calcium, 2.5% magnesium and 1.56% potassium. *Datura mete* it dried samples has sodium content of 432.29 ppm, Mn to be 19.47 ppm, Copper 1.11ppm, Iron 7.89 ppm & Pb 0.07 ppm, Nickel 0.06ppm, Zn 4.69 ppm, chromium 0.03 ppm and lastly, cobalt to be 0.41ppm. Appreciable amounts of metals like potassium, sodium, calcium are vital in regulation of water - electrolyte and acid-base balance in the body, provides both man and animal with rigidity and support , essentials for bone and teeth formation and development, blood clotting and normal functioning of the heart, nervous systems and the muscles respectively. (Omidiji, M. *et al,* 2005) Although, this amount are negligible, continuous ingestion of *Datura mete* will be disastrous (Dalziel, J.M., 1999 and Busson F, 1995). The phytochemical analysis of *Datura mete* shows that it posses alkaloids and saponin, which can be extracted or isolated for dietary supplements and nutraceuticals. Saponins can be widely used because of their effects on ammonia emission in animal feeding. Saponins are known to possess both microbial and anti-inflammatory activities. (Fawusi *et al,* 2000). Studies have reported the beneficial effects of saponins on blood cholesterol levels and stimulation of the immune system. (Imungi, J.G. and Gbile Z.O 1999). Alkaloids have diverse but important physiological effects on both man and animal. They are good medicinal value either as analgesics or anesthetics *Datural mete* does not possess Anthroquinone or combined Anthroquinones *Datural metel* possess Tannin as well. (Daunay, M.C, 1999) Tannin have been suggested to be involved with antibacterial and anti-viral activities, while tannins and flavonoids are thought to be responsible for antidiarrheal activity (Daunay, *et al,*2001) This can be used on tanning animal hides into leather.

The phenolic content of *Datura metel* is 0.04% it looks negligible but there work cannot be over emphasized. *Datura* mete is reach in vitamin C with a value of 7.5mg/100g.

Conclusion

The results obtained from this research were shown that *Datura* species are good sources of nutrients e.g. calcium, potassium, magnesium, zinc etc which can be good for the development and maintenance of good health and body growth if well processed and if refined can be a good scavenger of free radicals. (Onayemi O. *et al,* and lyon, F. *et al*) *Datura metel*, a non edible egg plant should be refused for a locally available raw material for pharmaceutical, industry and agro-allied properties, hence, further research is needed. (Dalziel, J.M (1999) and Chweya (1995).

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