

Qualitative and quantitative evaluation of non-biologically active by-product of
Dracaena Manni fruit pulp Baker (Fam. Agavaceae)

ODOH, U. E.* EZUGWU C.O. AND EBEBE I. M.

Department of Pharmacognosy, University of Nigeria, Nsukka

*Correspondence

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The non-biologically active by-products of *Dracaena manni* fruit pulp were evaluated for its unutilized bioactive principles. A 1.0 kg of powdered fruit pulp of *D. manni* was extracted successively with petroleum ether and the non-biologically active products studied both qualitatively and quantitatively. Preliminary phytochemical analysis and quantitative examination revealed the presence of carbohydrates ($21.50 \pm 1.22\%$) proteins ($19.25 \pm 1.64\%$), fats and oils ($17.55 \pm 0.92\%$) and saponins ($41.70 \pm 2.40\%$). The physical and chemical properties of the oil were: specific gravity at 15 °C (0.0849), refractive index at 20 °C (1.506), viscosity (290 centipoises), saponification value (151.47), acid value (3.843), ester value (147.63) and unsaponifiable matter (1.1%). The detergent capacity of the saponin fraction gave the values of the foam number [(0.18, 0.20 and 0.30 ml/gm), foam index (8.33, 12.50 and 20.00 ml/gm) and foam capacity (0.09, 0.10 and 0.15 ml/gm)], for *D. manni* extract, quillaia bark extract and commercial detergent (0.10) (OMO®) respectively. The commercialization of *Dracaena manni* fruit-pulp could be viewed from the nutritional content of the plant i.e. protein, which suggest possible presence of essential amino acids in the fruit. The carbohydrate content can be explored as animal feeds or as excipients in various pharmaceutical dosage forms. Oils could serve as cheaper alternative in industrial production of soap and the detergency of the saponin fraction can as well be utilized.

Keywords: Qualitative and quantitative evaluation, non-biologically active, *Dracaena manni*, nutritional content and commercialization

INTRODUCTION

Scientific investigations on the plant have been centered on secondary metabolites with pronounced biological activities, but the commercial development of *Dracena* on this basis looks expensive. Hence the need to explore other by-products of the plant, which could subsidize the commercialization of the plant for its bioactive principles. This venture comprises a qualitative and quantitative evaluation of by-products like carbohydrates, proteins, saponins and fats and oils, which conventionally seen to enjoy a wide scope of industrial application.

Carbohydrate for example is a complex whole of excipients, which are extensively used, in pharmaceutical industries. Though widely distributed in plants, large-scale production is limited to only few plants. In pharmaceutical industries, carbohydrate in the form of cellulose is used in the form of its derivatives resulting from different chemical treatments These ranges from esterification,

etherification to acid hydrolysis. The products so formed, ethylcellulose, microcrystalline cellulose, cellulose acetate phthalate, etc are used as pharmaceutical excipients-binders, thickeners, disintegrants, diluents, tablet coating material and packaging materials. Similarly, starch, a carbohydrate, remains an important ingredient in tablet manufacturing. It serves as binders, disintegrants and glidants in tableting. The absorbent property has enhanced its use in the production of dusting and cosmetic powders. Like cellulose, restricted number of plants are involved in the large-scale production of starch. This may be due to cost and the suitability of the starch to well-defined pharmaceutical standards.

The occurrences of proteins in plants are estimated by the determination of the amount of nitrogen present in the plant part. Apart from its use as a food supplement, the constituent amino acid units could

be evaluated for presence of rare amino acids or used in the semi-synthesis of important amino acids.

Saponins are reputed locally due to their ability to reduce surface tension of water, hence are used in soap making. This could be exploited industrially in the manufacture of natural laundry detergents.

Fixed oils, e.g. castor oil, coconut oil, corn oil, *Dracaena* oil, theobroma oil, etc have various uses in industrial operations based on their physical and chemical properties like viscosity, saponification value, and specific gravity. They are useful excipients in pharmacy. For example, corn oil has been used as a solvent for injections (1). Theobroma oils are used in the formulation of suppository in which in they serve as the suppository and ointment bases (2). They have also found use in cosmetic and paint industries, especially the "drying oils". *Dracaena* oil in effect needs to be assessed for possible uses in some of these areas.

Dracaena mannii is reputed locally for therapeutic activities. The leaves have been implicated in the treatment of backache and rheumatism (3). The swollen roots of some species serve as aphrodisiac (4). In Liberia and Guinea, the ashes of the leaves are used locally for soap making (4)

Phytochemical analysis of the fruit-pulp revealed the presence of proteins, carbohydrates, saponins, fats and oils and steroids/triterpenoids. Previous chemical investigations on the fruit pulp revealed a triglycoside of pennogenin, which was found to be fungicidal (5). Malcolm and Sofowora (6) reported an antibacterial activity of the extract from the bark against gram-positive and gram-negative organisms especially *Staph. aureus*.

This study therefore aim at evaluating of the by-products of *Dracaena* fruit pulp coupled with comparative analysis with commercially used products such that it could contribute to the commercial development of the plant.

EXPERIMENTAL

Plant material

Fruits of *D. mannii* plant growing wild at Nsukka, Nigeria were collected in November, 2001. They were identified by Mr. A. Ozioko, a taxonomist in the Department of Botany, University of Nigeria, Nsukka Voucher specimen (UNN / PCOG / 012) has been deposited at the herbarium of the Department of pharmacognosy, University of Nigeria, Nsukka.

QUALITATIVE EVALUATION OF PLANT METABOLITES

Phytochemical test

Phytochemical test to detect the presence of required by-products were carried out following standard procedures (7).

Determination of the physical and chemical properties of expressed oil-

Physical and chemical properties such as specific gravity, refractive index, and viscosity, acid value, saponification value, ester value and unsaponifiable matter of the oil were all determined following standard method (8-10),

Specific Gravity

The specific gravity of a liquid is the weight of a given volume of the liquid at the specified temperature compared with the weight of an equal volume of water at the same temperature, all weighing being taken in air. Measurements are done by weighing a dry pycnometer (a gm) then filling it with distilled water and noting the weight (b gm). The pycnometer is then washed and dried then filled with oil and the weight taken (c gm).

Acid Value

The acid value is the number of mg of potassium hydroxide required to neutralize the free acid in 1 g of the substance under analysis. 5 gm of the oil was weighed into a 250 ml flask and 50 ml of a mixture of equal volumes of Ethanol (96%) and ether which has been neutralized after the addition of 1 ml of dilute phenolphthalein solution added. The solution obtained was titrated with 0.1 M. potassium solution with constant shaking until a pink colour which persisted for fifteen seconds was obtained. The volume of titrant used was noted (V) and the acid value calculated from the following expression.

$$\text{Acid Value} = \frac{V \times 0.00561 \times 1000}{\text{weight(gm) of the oil}}$$

Ester Value

This is the number of mg of potassium hydroxide required to saponify the esters present in 1 gm of the oil.

$$\text{Ester Value} = \text{Saponification value} - \text{Acid value.}$$

Saponification Value

Two gram of the oil sample was weighed out into a 25 ml conical flask, 25 ml of N/2 alcoholic-caustic potash solution was added. The flask was connected to a reflux condenser and heated on an electric hot plate for 1 hour during which the sample was completely saponified as indicated by the absence of any oil matter and appearance of clear solution.

After the cooling of the flask and the condenser, the inside of the condenser was washed down with 10 ml of hot ethyl alcohol, 1 ml of indicator solution of phenolphthalein and the solution titrated with N/2 standard hydrochloric acid.

Also the blank determination was conducted following the illustrated procedure above without the oil and the value calculated using the saponification equation.

Unsaponifiable matter

These include all components of the oil not saponified by potassium hydroxide and includes triterpenes and sterols. The oil was weighed and the percentage of the total weight of the oil calculated.

Viscosity test

In the oil sample contained in a cup, the viscometer (ferranti portable viscometer) was immersed and the motor switched on and the reading of the viscosity on the calibrated dial was taken.

Refractive index test.

Using abbe's refractometer, a smear of the oil sample was placed on the lower angled mirror. With fine adjustments of telescope tubes until a black shadow appeared in the center of the cross wire indicator, then the reading was taken at the temperature of 20 °C.

Estimation of the detergency of the extract

The detergent properties of the extract was estimated as foaming capacity, foam number and foam index and compared with quillaia extract and commercial detergent OMO®. The foaming properties (foaming capacity, foam number and foam index) were determined according to the Lawhon *et al* (1972) procedure (11). 2.5 g of sample was suspended in distilled water and the pH adjusted to 7.0. The suspension was whipped in a Kenwood chef food mixer for 10 min. The suspension was immediately poured into a 100 ml measuring cylinder and the foam height and volume of liquid collected at the bottom of the cylinder were measured at intervals. The foam properties were then calculated as described by Lawhon *et al.* (1972).

QUANTITATIVE EVALUATION OF THE BY-PRODUCTS

Determination of fat or oil content

1.5 kg of the powdered crude drug were packed into the soxhlet extractor pot and then mounted on a Heating mantle. 500 ml of pet. ether solvent was poured through the material into the pot at the

bottom. A condenser was fitted in position and then the heater switched on. The temperature of the heater was adjusted such that it is not more than the boiling point of the solvent. The extraction was allowed to run until the small arm of the thimble showed a clear solvent. Residual solvents were recovered by drying in an oven at 60 °C for 24 hour. The oil was then dried at 100°C in an oven and weighed. The oil content in the sample was calculated thus:-

$$\text{Percentage weight of oil} = \frac{W_o}{W_m} \times \frac{100}{1}$$

Where W_o = weight of oil; and W_m = weight of plant material.

Determination of percentage saponin content

1.0g of the extract was dissolved in 100 ml of petroleum ether. The saponin in the solution was precipitated with diethyl ether and then weighed. Percentage saponins content was calculated thus:-

$$\text{Percentage saponins} = \frac{W_s}{W_m} \times \frac{100}{1}$$

where W_s = weight of saponin; and W_m = weight of extract.

Determination of carbohydrate content

25.0 g of the dried *D. manni* fruit pulp was de-resinified using Benzene-Ethanol (2:1) and then carbohydrate extracted following procedures outlined by Harbone (7). The percentage carbohydrate content was calculated thus: -

$$\text{Percentage carbohydrate} = \frac{W_c}{W_m} \times \frac{100}{1}$$

Where W_c = weight of carbohydrate; and W_m = weight of plant material

Determination of protein content

Protein content of the fruit pulp was determined following Micro - Kjeldahl method as described by Pederson (12). 2 gm of the sample was weighed and digested with 5 ml concentrated H_2SO_4 using mercury tablet as catalyst. After digestion, the digest was made up to 100 ml with distilled water. 5 ml of this digest was mixed with 5 ml Boric acid using methyl red indicator. The titration was done using 0.01N HCl as the titrant and the estimation of nitrogen content of the sample was done.

$$\% \text{ protein} = \frac{\text{Titre value} \times 0.0001401 \times 20 \times 6.25}{\text{sample weight}} \times \frac{100}{1}$$

RESULTS AND DISCUSSIONS

Phytochemical analysis of the fruit pulp of the plant revealed the presence of the following by-products – proteins, carbohydrates, oils and saponins (Table 1). The protein content was found to be 19.25% and compared well with other proteinous edibles like groundnut, 23.2 and melon, 19.4-25.8% (13). This

shows that if detoxified, it could be a potential substitute for any of them as vegetable protein source. Furthermore, the hydrolysis of the protein to its amino acid building units could be exploited since it could serve as a possible cheap source of rare amino acids or could be used as starting materials in the semi-synthesis of some important amino acids.

Table 1: Results of the preliminary phytochemical analysis to assess the presence of the by-products in the fruit pulp.

Test	Result
Test for proteins	
Millions test	++
Xanthoprotein test	+
Biuret's test	+
Test for Carbohydrates	
Molisch's test	+
Fehling's test	++
Bardfoed's test	+
Iodine test	++
Test for Fats and Oils	
Paper test	+
Test for Saponins	
Frothing test	+++
Emulsification test	+

A general view of the data obtained for the physical and chemical properties of oil fraction (Table 2) shows that the oil would be suitable for industrial application. The saponification value of the oil is a little bit close to that of palm oil, 195-205 and olive

oil, 188-196 (14) and so can serve as cheap alternative in soap making. The fact that *Dracaena* oil is a drying oil, suggests that it can find use in cosmetic and paint industries.

Table 2: physical and chemical properties of the oil.

Specific gravity (15°C)	0.0849
Refractive Index (20°C)	1.506
Viscosity	290 centipoises
Acid value	3.843
Saponification value	151.47
Ester value	147.63
Unsaponifiable matter	1.1%

Table 3: Result of Quantitative estimation of the by-products in fruit pulp.

By – Products	Percentage Composition
Proteins	19.25 ± 1.64
Fats and Oils	17.55 ± 0.92
Carbohydrate	21.50 ± 1.22
Saponin	41.70 ± 2.40

The high content of cellulose contained in the fruit pulp could warrant its use as animal feed, since animals are able to digest cellulose through the activity of luminal microorganism (15). This use is also augmented by the presence of large quantity of proteins. In addition to this, carbohydrates in the form of cellulose and/or starch has find wide application in the area of pharmaceuticals. This ranges from its use as excipients in various dosage forms to the use as packaging materials. In effect, carbohydrate in the *Dracaena* fruit pulp being of suitable quality and quantity could provide a cheap source of cellulose and / or starch for the above purpose.

The detergency property of the saponin fraction was quite remarkable (Table 4). The foam capacity, 0.09 ml/gm compared favourably with that of quillaia extract (0.1 ml/gm) and that of synthetic detergent Omo®, (0.15 mg/gm) (2) and being of natural original, it lacks the tendency to attack materials and fibre as is the case with synthetic detergents. Moreso, the saponins resembles sex hormones in structure, thus could be exploited for as a possible starting materials in the semi-synthesis of these hormones. The articulation of all these findings may form basis for the preliminary development of the by-products of *Dracaena* plant for commercial purpose.

Table 4: Result of the Detergent capacity of the substances.

Substance	Foam capacity (ml/gm)	Foam Index	Foam Number
Dracaena extract	0.09	8.33	0.18
Quillaia extract	0.10	12.50	0.20
Omo®	0.15	20.00	0.30

REFERENCES:

1. Tyler, V. E., Brady, L. R. and Robberts. J.E. Pharmacognosy, 11th edn, Balliere Tindal, London, (1976) pp 31 & 4.33.
2. Ajibola, A. O., Okunji, C., O. (1984) Physiochemical properties of Theobroma oil and its pharmaceutical application. *Nig J. Pharm. Scs.*, 15: 25 - 27
3. Keay, R. N. J., Onochie, C.F.A. and Stanfield, D.R., "Nigeria Trees 11, Feb. Dept. of Forestry Research, Ibadan, 1964, Vol. 11, 439 - 440.
4. Irvine, F.R., "Woody plants of Ghana with special Reference to their uses", Oxford University Press, London, 1961, 769 - 771.
5. Okunji, C.O. and Iwu, M.M., (1988), Antimicrobial Activity of a pennogenin triglycoside from *Dracaena mannii* fruit pulp. *Int. J. Crude Drug Res.*, 26: 4 - 11
6. Malcom, S.A. and Sofowara. E.A.. (1969). Antimicrobial Activity of selected Nigeria Folk Remedies and their Constituents. *J. Nat. Products.*, 32: 512
7. Harborne, J.B. "Phytochemical Methods: A Guide to Modern Techniques of plant Analysis", Chapman and Hall, London, 1973. 14 - 255.
8. United States pharmacopoeia XX1 - National formulary XIV (1980) 20th edn, United States pharmacopoeial Conv. Inc., Rockville.
9. African Pharmacopoeia (1986), OAU/STRC scientific Publications, Lagos, Vol. 2, pp 1323-1325.
10. British Pharmacopoeia (1973), Her Majesty Stationary Office, London, Vol. 1, pp 1623-1628.
11. Lawhon, J.T., Rooney, L.W., Carter C.M., Multil, K.F. (1972). Evaluation of protein concentrate produced from glandless cotton flour by a wet extraction process. *J. Food Sci.* 37, 778 - 782.
12. Pederson, C.S., "Microbiology of food fermentation", the Avi Pub. Co. Ltd, London, 1971, 41 - 271
13. Ihekoronye, A.I. and Ngody, P.O., "Integrated Food Science and Technology for the Tropics. 1st ed.. Macmillan Publishers. London, 270 - 310.
14. Aurand, L.W., Woods, A.E. and Wells, W.R.. "Food composition and Analysis", Van Nostrand-Reinhold company, New York, 1987, 31 - 33.
15. Ramstad. E., "Modern pharmacognosy. 1st ed., McGraw-Hill, London, 1959. 54.