Afr J Med. med Sci (1998) **28**, 181-183

Phytochemical and microscopic evaluation of cultivated *Datura innoxia* miller

K.A. Abo and O.A. Oni

Department of Pharmacognosy, Faculty of Pharmacy, College of Medicine, University of Ibadan, Nigeria.

Summary

Using a sensitive analytical method, the total alkaloid content of *Datura innoxia* cultivated in Ibadan is reported at different stages of growth. Furthermore, the total hyoscine content of *D. inoxia* has been shown to peak during the harmattan season and to be at its lowest in the rainy season in Ibadan. The diagnostic microscopic features of this little studied species is also described.

Keywords: *Datura Innoxia, hyoscine content, seasonal variations*

Résumé

En utillsant une methode analytique sensitive, le contenue total en alkaloide de *Datura innocia* cultivè à Ibadan e ètè reportè à differents stages de development. Deplus, le contenu total de l'hyoscine de *D. innoxia* a atteint sa valeur optimale pendant la saison de l'harmattan, et sa plus faible valeur pendant la saison de pluie à Ibadan. Les characteristiques du diagnostique micorscopicque de cette peititè ètude est ausiio discutè.

Introduction

Datura innoxia Miller (Solanaceae) is one of the 10 Datura species growing worldwide. It is used in traditional medicine for the treatment of snake bite, cough, asthma, as an intoxicant and for rituals.[1-3].

D. Innoxia and *D metel* are the two commonly found *Datura* species in Nigeria. Both species are easily confused by the indigenous population because of their very close morphologiccal and histological features [4]. Both species are called "Apikan" in Yoruba and "Hau-kata yaro" in Hausa. There is very limited research on *D. Innoxia* growing in Nigeria.

Datura stramonium and *Atropa belladonna* are the two official plant species described in many pharpacopoeias as the natural sources of hyoscine (scopolamine) and atropine (DL-hyoscyamine). These two European species are the principal raw materials for the preparation of galencials such as Stratmonium and Belladonna tinctures BP which are used in pharmacy for the formulation of many cough, diarrhoeal and peptic ulcer preparation.

In our continued search for possible substitutes for *D stramonium.*, the total alkaloid content of various morphological parts of *D. Innoxia* at different stages of growth in Ibadan was investigated. This communication also describes seasonal variations of total leaf alkaloids calculated as hyoscine, the major alkaloid of many tropical *Datura* Species [5,6]. A microscopic description of the diagnostic features of *Datura* species in power form is also presented.

Correspondence: Dr. K.A. Abo, Dept. of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria.

Materials and methods

Plant Material

D. Innoxia was raised by authors from seeds (collected from Kano) grown in Ibadan (1 m apart) on loose soil of about 0.64% total nitrogen and maintained with abundant farmyard manure. The species was authenticated at the Forestry Research Institute of Nigeria, Ibadan, where herbarium specimen had been deposited. Fresh organs of *D. Innoxia* were collected in the mornings, dried at about 60 ^{0}C, powdered and stored in amber-coloured containers ready for analysis. For seasonal studies, leaves were harvested weekly over a period of one year. Five replicate determinations were carried out on each batch.

Microscopy

The powdered sample of the leaves was sieved and the collected fine powdered was cleared with chloral hydrate solution over a bunsen flame, stained with phloroglucinol /conc. HCl and immediately mounted in glycerol and examined under the Olympus binocular research microscope. The result is shown in Figure 1.

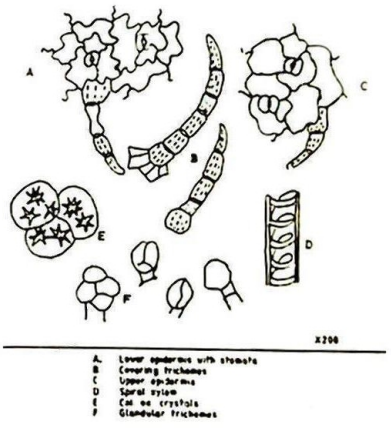

Fig. 1: Microscopic of powdered *D innoxia*

Microchemical tests

About 0.1 ml of the extracts containing the alkaloidal salt was placed in a series of test tubes and about three drops of alkaloidal reagents (Dragendorffs', Mayers' and Wagners') were separately added to each of the test tubes and the colour of the precipitates noted.

The Vitali-Morin test was performed on aliquots of each extract. Two drops of glacial acetic acid were added to each dried extract in porcelain dishes. This was evaporated and cooled. About three drops of fuming HNO_3

were then added to each residue and evaporated to dryness. A few drops of 3% KOH/MeOH were added to each dish. And the colour noted.

Assay for total alkaloid content

The determination of total alkaloid content of various organs and batches of *D. Innoxia* was performed on about 1g (accurately weighed) powdered sample using a sensitive micro-method for the quantitation of total alkaloid of *Datura* species as previously described by Abo and Ikhile [7]. The purified alkaloidal bases were heated on a water bath for 15 minutes. This was reconstituted in about 1ml $CHCl_3$ and transferred to a beaker containing 10 0.01 M H_2SO_4. Excess $CHCl_3$ was evaporated on a steam bath and cooled. A few drops of methl red indicator were added and the mixture titrated potentiometrically with 0.02M NaOH solution. The results of the assay are shown in Table 1 and 2.

Table 1: Total alkaloid content of organs of cultivated *Datura innoxia*

Plant organ	Period of harvest	Total hyoscine content (% w/w)
Leaf	At peak flowering	0.27 ± 0.007
Fruit	At fruit dehiscence	0.33 ± 0.003
Flower	At peak flowering	0.44 ± 0.023
Root	Towards end of growing season	0.39 ± 0.012

Table 2: Seasonal distribution of total leaf alkaloids in *D. innoxia*

Month of harvest	Average rainfall (mm/day)	Water-soluble extractive	Hyoscine content (% w/w)
January	1.07	13.50	0.37 ± 0.01
February	2.24	15.25	0.28 ± 0.00
March	3.29	14.32	0.24 ± 0.01
April	1.88	11.50	0.22 ± 0.02
May	4.01	8.00	0.14 ± 0.00
June	9.09	11.50	0.16 ± 0.03
July	5.00	13.50	0.26 ± 0.01
August	0.64	15.00	0.28 ± 0.01
September	10.10	16.00	0.25 ± 0.01
October	7.52	12.00	0.33 ± 0.01
November	1.60	9.75	0.53 ± 0.22
December	0.00	10.50	0.56 ± 0.11

Results and discussion

Alkaloids of *Datura* act on the autonomic nervous system. Various tropane alkaloids such as atropine, hyoscine and meteloidine have been reported in *D. Innoxia* growing wild in India [8,9]. These alkaloids and related drugs are sold in prescription and in a number of proprietory mixtures for the treatment of gastrointestinal diseases, cold, hay fecer, parkinsonism and asthma. Atropine is used to a large extent in ophtalmic practice to dilate the pupil of the eye. *Datura* species and their alkaloids are still in demand for the preparation of tinctures and many anti-diarrhoeal and peptic ulcer formulations.

The identification tests and the purple colour observed in the Vitali Morin test showed that all extracts of *D. innoxia* contained tropane alkaloids. Table 1 shows the total hyoscine content of organs of the cultivated *D. innoxia* at different stages of growth. It shows that hyoscine is concentrated in the flowers [0.44%] and minimal in the leaf (0.27%) at peak flowering . The high level of alkaloids in the flower and roots is consistent with earlier findings on an Indian variety of *D. Innoxia* [10]. Table 2 shows seasonal distribution of total leaf of the species. The figure for each month are the means of the appropriate weekly determinations presented as ± S.D. The total alkaloid content reached peak levels during the hot dry season (October, - January), the maximum content being recorded in the harmattan period. Hyoscine is formed in the root [9] from hyoscyamine via 6-hydroxy hyoschamine. The extent to which this conversion occurs often depends on the age of the plant, particular variety or species and geographical source; and is an important factor in determining the ultimate uses and pharmacological properties of the plant. [11].

Total alkaloid content was lowest during the rainy season in Ibadan, falling below the acceptable level of 0.25% (for leaf) in May and June. The low alkaloid level recorded during the rainy season is consistent with earlier findings by Gupta *et al* [6] and *Abo et al* 12 in various tropical *Datura* species. Chromatographic examination of the extracts show that hyoscine occurred in greater proportions than hyopsyamine where both alkaloids coexisted. Little or no hyoscyamine was detected in the leaf extracts during the rainy season.

The cultivated species was a bushy annual herb attaining a height of about 1.5 m. The powdered leaf sample was characterised by epidermal cells with wavy (lower epidermis) anticlinal walls; numerous anisocytic stomata; glandular trichomes (unicellular, bicellular heads and unicellular and multicellular stalks); numerous lignified spiral xylem and mesophyll cells containing rosettes of calcium oxalaet crystals [Figure 1].

This study has provided basic information on alkaloidal distribution in *D. innoxia* and the best period of harvest for use as a source of drugs. This species could be standardised as a possible substitute for *D. Stramonium* in Nigeria.

Acknowledgements

The authors are grateful to the staff of the Climatological Centre of the Geography Department, University of Ibadan for the rainfall data and staff of I.I.T.A., Ibadan, for soil analysis.

References

1. Kokwaro JO. Medicinal plants of East Africa. Kampala: East African Lt. Bureau 1976; 204
2. Lewis WH, Edwin-Lewis PFM. Medical Botany. New York: John Wiley and Son, 1977; 419-425.
3. Dalziel JM. The useful plants of West Tropical Africa. London: Crown Agents, 1937; 428-430.

4. Kosova V, Chladek M. Pharmacognosy of *Datura metel* and of *D. Innoxia*. Pharmazie 1957; 12: 620-627

5. Karnick CR, Saxena MD. Variability of alkaloidal production in *Datura* species. Planta Medica 1970; 18 (3): 266-269.

6. Gupta S, Prabhakar VS, Madam CL. The distribution of total alkaloids and major components in the different organs of *Datura metel* var. *fastuosa* at various stages of growth. Planta Medica 1973; 23 (4): 370-376.

7. Abo KA, Ikhile BU. A Sensitive micro-method for the quantitation of total alkaloids. Phyt. Res. 1987; 1 (4): 191-192.

8. Verza-Petri G, Sarkany S. Comparative study of Asiatic and American species for drug production. Acta Pharm Hung. 1961: 31: 22-24.

9. Evans WC, Patridge MW. Alkakoids of *Datura innoxia* Nature. 1953; 171: 656

10. Shah CS, Khanna PN. Alkaloid content of *Datura metel* and *D. innoxia* var. *Fastuosa*. Ind. J, Pharm. 1964; 26-140.

11. Evans WC. In: Hawkes JG, Lester RN, Skelding AD (eds.) The biology and taxonomy of the Solanaceae. London: Academic Press, 1979: 241-254.

12. Abo KA, Salami OO, Adelegan IO. Variation of total hyoscine content of Cultivated . *Datura metel* Linn. Afri J. Med. Sci. 1993; 22: 45-47.