

NIGERIAN GARDENIA ERUBESCENS STAFF & HUTCH., A POTENTIAL COMMERCIAL SOURCE OF MANNITOL

M.M. HUSSAIN,* M. SHOK, A. GHANI AND E.N. SOKOMBA

*Department of Pharmacognosy & Drug Development,
Faculty of Pharmaceutical Sciences,
Ahmadu Bello University, Zaria,
Nigeria.*

ABSTRACT

A phytochemical investigation on the various parts of *Gardenia erubescens* Stapf & Hutch. (Rubiaceae), used in traditional medicine in Northern Nigeria resulted in the isolation of D-Mannitol as the principal chemical constituent of the plant. The leaves, stem bark, roots, stem wood and fruits of the plant were found to contain 5.0%, 3.4%, 1.8%, 0.4%, and 0.25% of D-Mannitol respectively. The possibility of the use of *G. erubescens* as a potential source for commercial production of mannitol has been discussed.

INTRODUCTION

The various parts of *Gardenia erubescens* Stapf & Hutch. (Family Rubiaceae), a tropical shrub grown in Nigeria, are used in traditional medicine in northern Nigeria. Although a number of species of *Gardenia* have been investigated phytochemically (Kuhn & Weigand, 1928; Rama Rao et al, 1970; Krishnamurti et al, 1971; Endo & Taguchi, 1973; Inouye et al, 1974; Gupta et al, 1975; Billore et al, 1976; Takeda et al, 1976; Chhabra et al, 1977a, 1977b; Joshi et al), 1979, there appears to be no phytochemical report on *G. Erubescens*. Mannitol has been reported as one of the chemical constituents of many of the *Gardenia* species investigated, which include the South-East Asian *G. Lucida*, *G. turgida*, *G. latifolia*, *G. gummifera* and *G. grandiflora* (Reddy et al, 1973, 1975 and 1977; Dutta et al, 1966; Hayashi, 1944) and the African *G. promodora*, *G. Vogelli* and *G. florida* (Delaude & Kapundu, 1975). *G. lucida* has been

reported to contain about 3 per cent mannitol (Dutta et al, 1966) and a dried resinous exudate from *G. turgida* yielded about 40 per cent mannitol (Forster & Rao 1925). In this work phytochemical investigation has been carried out to detect and isolate mannitol present in various parts of *G. erubescens* and to estimate the mannitol content quantitatively.

The isolation, characterisation and quantitative estimation of mannitol from the local plant have been described in this paper.

EXPERIMENTAL

Plant material: Various parts (leaves, roots, stem bark, stem wood and fruits) of the plant were collected from plants growing wild along Zaria-Kaduna Highway of Kaduna State, Nigeria. The plant was identified in the herbarium of Biological Sciences Department of Ahmadu Bello University, Zaria, Nigeria, where a voucher specimen (No. 119; T.C.N. Baker) has been deposited. The dried plant parts were reduced to moderately coarse powders by using mechanical grinders.

Extraction of plant materials. The powdered plant materials (250g each) were first defatted by extracting twice with hot petroleum ether (bp 60-80 °C) using a Soxhlet extractor. The defatted materials were then extracted exhaustively with hot methanol. The filtered extracts were then concentrated by using a rotary evaporator to about 200 ml each and allowed to stand in a refrigerator.

Isolation of D-Mannitol. An amorphous, dirty, yellowish white resinous residue separated out after 24 hrs of refrigeration. The supernatant mother liquor was sepa-

rated by careful decantation, which was further concentrated and cooled to obtain a second crop of the resinous residue. The combined residue was washed with hot benzene to remove the oily impurities. The oil-free residue was then dissolved in water and treated with activated charcoal to remove the colouring matter. The decolourised aqueous solution was filtered twice to remove the charcoal particles. The clarified solution was evaporated to dryness on a water bath. The resultant mass was crystallised twice from hot aqueous methanol to give white soft needles.

Quantitative estimation of D-Mannitol. The quantity of D-mannitol present in each part of the plant was separately estimated by the titrimetric method described in the British Pharmacopoeia (1980). Methanolic extract of each plant part containing the equivalent of approximately 400 mg of mannitol was used for this estimation.

RESULTS AND DISCUSSION

Characterisation of D-Mannitol. The above crystallised compound was characterised and identified as D-mannitol by the following physical and chemical properties of the compound: sweet taste, high solubility in water, prismatic needle shape of the crystals, mp (165-66 °C), mixed mp (165 °C), optical rotation (+ 23 to 24 ° in water), co-chromatography and chromogenic reactions. The identity of D-mannitol was further confirmed by preparing its acetyl (hexa-acetate, mp 119-21 °C, $[\alpha]_D^{25} = +25.1$ in CHC 3) and benzoyl (hexabenzoylate, mp 148-49 °C, $[\alpha]_D^{25} = +53.90$ in CHC 3) derivatives. (Melting points were

determined by a Callenkamp Melting point apparatus and are uncorrected. Optical rotations were measured by a Ballingham + Stanely Polarimeter, Model B+S).

D-Mannitol was found to be present in all the parts examined, with the highest quantities in the leaves and stem bark. The percentage quantity of the compound found in each part is tabulated in Table 1 below.

Table 1: D-Mannitol content of the various parts of *Gardenia erubescens* Stapf & Hutch. (% dry weight)

Plant part	Quantity of mannitol
Leaves	5.00 + 0.25
Stem bark	3.40 + 0.17
Roots	1.80 + 0.09
Stem wood	0.40 + 0.02
Fruits	0.25 + 0.01

Although mannitol is a common constituent of the plants of Rubiaceae as it is in those of Umbelliferae, Oleaceae and Scrophulariaceae (Swain, 1963; Subramanian & Nair, 1971), so far we are not aware of any Nigerian plant of these families that has such a high quantity of mannitol. In view of the fact that mannitol can easily be isolated from all parts of the plant, especially in substantial quantities from the leaves (5.0%) and stem bark (3.4%) (cf. Table 1), *G. erubescens* could be profitably utilized as a good source for commercial production of mannitol. The method of isolation is also simple, straightforward and cheap. Presently, *Fraxinus ornus* Linn. (Manna) is regarded as the principal natural source of mannitol for its commercial production, and it is also obtained by hydrolysis of waste turnings of Ivory nuts (Claus & Tyler, 1970). Apart from these two sources, apparently there appears to be no other natural source where mannitol occurs in substantial quantity. Thus Nigerian *G. erubescens* promises to be a potential commercial source of D-mannitol.

REFERENCES

1. BILLORE, S.K. Mehta, S.C. & Mall, L.P., Changes in chlorophylls a,b and carotenoids in summer leaves of tree species in a dry deciduous forest, *J. Indian Bot. Soc.*, 1976, 55 (1), 56-59.
2. British Pharmacopoeia. Her Majesty's Stationery Office, University Press, Cambridge, 1980, pp. 270-71.
3. Chhabra, S.C., Gupta, S.R. & Sharma, N.D., A new flavone from *Gardenia gum*, *Phytochemistry*, 1977a, 16 (3), 399.
4. Chhabra, S.C., Gupta, S.R.; Sharma, C.S. & Sharma, N.D., A new Wogonin derivative from *Gardenia gum*, *Phytochemistry*, 1977b, 16 (7), 1109.
5. Claus, E.P. & Tyler, V.E., *Pharmacognosy*, 6th edn., Lea & Febiger, Philadelphia, USA, 1970, pp. 57-58.
6. Delaude, C. & Kapundu, M., Presence of D-mannitol in African Rubiaceae....., *Bull. Soc. R. Sci. Leige*, 1975, 44 (5-6), 493-94.
7. Dutta, H.A., Ganguly, S.N. & Bhattacharya, A.K., Isolation of D-mannitol from *G. lucida* and *P. perviflora*, *J. Indian Chem. Soc.*, 1966, 43(5), 390.
8. Endo, T. & Taguchi, H., The constituents of *Gardenia jasminoides*, *Chem. Pharm. Bull.*, 1973, 21, 2684-86.
9. Forster, M.O. & RAO, K.A.N., D-mannitol from *Gardenia turgida*, *J. Chem. Soc.*, 1925, 127, 2176.
10. Gupta, S.R., Seshadri, T.R., Sharma, C.S. & Sharma, N.D., Chemical investigation of Dikamali gum: Isolation of a new flavone, 4-hydroxywogonin, *Indian J. Chem.*, 1975, 13, 785.
11. Hayashi, K., glycosidic dye and several colourless constituents of the Japanese Wronshy (*Gardenia grandiflora*), *Proc. Imp. Acad. (Tokyo)*, 1944, 20, 311-17.
12. Inouye, H., Takeda, Y. & Nishimura, H., On the constituents of *Gardenia* species, *Phytochemistry*, 1974, 13, 2219-24.
13. Joshi, K.C., Singh, P. & Pardasani, R.T., Chemical examination of the roots of *Gardenia turgida* Roxb., *J. Indian Chem. Soc.*, 1979, 56, 327-28.
14. Krishnamurti, M., Seshadri, T.R. & Sharma, N.D., Minor components of Dikamali gum, *Indian J. Chem.*, 1971, 9 (2), 189-90.
15. Kuhn, R.I. & Weigand, W. The colouring matter of Chinese fruits of *Gardenia grandiflora*, *Helv. Chem. Acta*, 1928, 11, 716-24.
16. Rama Rao, A.V.; Venkataraman, K., Chakrabarti, P., Sanyal, Ak.K. & Lose, P.K., Five Flavones from *Gardenia lucida*, *Indian J. Chem.*, 1970, & (5), 398-400.
17. Reddy, G.O.S., Ayengar, K.N.N. & Rangaswami, S., Triterpenoids of *Gardenia turgida*, *Phytochemistry*, 1973, 12 (7), 1831.
18. Reddy, G.C.S., Ayengar, K.N.N. & Rangaswami, S., Triterpenoids of *Gardenia Intifolia*, *Phytochemistry*, 1975, 14 (1), 307.
19. Reddy, G.C.S., Rangaswamy, S. & Sunder, R. Triterpenoids of the stem bark of *Gardenia guwanifera*, *Planta Medica*, 1977, 14 (32), 206-11.
20. Sgewamanian, S.S. & Nair, A.G.R., Distribution of mannitol and flavonols in some Rubiaceae plants, *Phytochemistry*, 1971, 10, 2125-27.
21. Swain, T. *Chemical Plant Taxonomy*, Academic Press, London, 1963, p. 319.
22. Takeda, Y., Nishimura, H. & Inouye, H., Studies on monoterpens glycosides ... two further new glycosides from the fruits of *Gardenia jasminoides* *ollia* forma *grandiflora* (Lour) Makino, *Chem. Pharm. Bull.*, 1976, 24 (11), 2644-46.

OLUMO '89

Contd from page 28

tested for their genuiness. This he said will ensure the speedy trial of all concerned because justice delayed is certainly justice denied.

The Chief Executive agreeing that the PSN and pharmacists need a change of image, called on government to put together a seminar involving the task forces, PSN members and government officials to study and decide on strategies for effective implementation of Decree 17, of 1989.