COMPONENTS OF BAUHINIA CANDICANS

ADOLFO M. IRIBARREN and ALICIA B. POMILIO^{1*}

Departamento de Química Orgánica, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Pabellón 2, Ciudad Universitaria, 1428 Buenos Aires, Argentina

Bauhinia candicans Benth. (Leguminosae) is a medicinal plant indigenous to subtropical regions of Argentina and southern Brazil. The infusion of its leaves is widely used because of its potential hypoglycemic action. No chemical reports on this plant are known.

This work deals with the identification of steroids, flavonoids, and protoalkaloids from the leaves and flowers of *B. candicans*. According to our results, the principle responsible for the hypoglycemic and hypocholesteremic activity of *B. candicans* had to be the trigonelline that occurs in its aqueous extract. This suggestion is supported by earlier pharmacological studies on this compound (1,2).

Further studies are being carried out in our laboratories.

EXPERIMENTAL²

GENERAL EXPERIMENTAL PROCEDURES. —Melting points were determined on a Fischer-Johns apparatus and are uncorrected. The uv spectra were measured on a Beckman DK-2A spectrophotometer; ir spectra were recorded in KBr pellets on a Perkin Elmer 137-B spectrophotometer. Optical rotations were measured on a Perkin Elmer polarimeter, model 141. ¹H-nmr (100 MHz) and ¹³C-nmr (25.2 MHz) were recorded in the stated solvents on a Varian XL-100 with a Fourier-transform accessory. Chemical shifts (δ) are expressed in ppm downfield from TMS as internal reference. The ms spectra were taken with a Varian MAT CH-7A spectrometer at 70 eV; glc were performed with a Hewlet Packard 5830-A (FID detector); glc/ms with a Varian 1400 chromatograph coupled to a Varian MAT CH-7A spectrometer. The system is interfaced to a Varian MAT spectra system 166 computer.

PLANT MATERIAL—. Leaves and flowers of *B. candicans* were collected in November in Buenos Aires (Argentina). Voucher specimens are deposited in Instituto de Botánica Darwinion (Argentina) under the number SI27581. The plant material was identified by Prof. Emilio Ulibarri.

ISOLATION AND IDENTIFICATION.—Dried, ground leaves were successively extracted with petroleum ether and ethanol. Upon concentration of the former extract, a solid separated out, which was mainly composed of triacontanol identified by spectral data (3).

The filtrate was chromatographed on a silica gel column using gradients of petroleum etherchloroform-acetone. One of the fractions yielded sitosterol, campesterol, stigmasterol, cholesterol, and stigmast-3,5-dien-7-one identified by glc and glc/ms.

The ethanolic extract was suspended in 10% acetic acid and filtered. The acidic filtrate was exhaustively extracted with ethyl acetate, and the acidic aqueous layer was basified to pH 12 (K_2CO_3), then further extracted with chloroform. The remaining aqueous layer was treated with Reinecke salt. The reineckate obtained was dissolved in acetone and passed over a resin column (Amberlite IRA-400, HO⁻ form; ethanol), yielding choline that was identified by spectral data and comparison with a standard.

Upon acidification to pH 1 and treatment with Reinecke salt as described above, trigonelline was obtained. Its spectral data were coincident with those reported $[^{1}H-nmr(4), ms(5)]$.

Trigonelline acetate: ¹³C-nmr (25.2 MHz, D_2O) ppm: 23.4(*C*H₃COO⁻,q); 49.1(NCH₃,q); 128.4(C-5,d); 137.6(C-3,s); 145.5(C-2,d); 146.7(C-6,d); 147.8(C-4,d); 179.7(COOH,s); 180.4 (CH₃COO⁻,s).

The ethyl acetate layer was chromatographed on a silica gel column using gradients of chloroformethanol as eluents. Three main fractions were obtained. The first one was purified by silica gel H column chromatography yielding sitosterol 3-0- β -glucoside identified by hydrolytic and spectroscopic methods (6,7).

Sitosterol 3-O-β-glucoside: ¹³C-nmr (25.2 MHz, Py-d₅) ppm: 12.1(C-18)^a; 12.3(C-29)^a; 19.1(C-

¹Research member of the National Research Council of Argentina (CONICET).

²Full details of the isolation and identification of the compounds are available on request to the senior authors.

21)^b; 19.3(C-26)^b; 19.5(C-19); 20.1(C-27); 21.4(C-11); 23.5(C-28); 24.6(C-15); 26.5(C-23); 28.6(C-16); 29.6(C-25); 30.4(C-2); 32.2(C-7 and C-8); 34.3(C-22); 36.5(C-20); 37.0(C-10); 37.6(C-1); 39.4(C-12); 40.0(C-4); 42.6(C-13); 46.1(C-24); 50.4(C-9); 56.3(C-17); 56.9(C-14); 62.9(C-6^G); 71.7(C-4^G); 75.3(C-2^G); 78.1(C-3); 78.5(C-3^G); 78.6(C-5^G); 102.6(C-1^G); 121.9(C-6); 140.8(C-5) (^{a,b} these assignments may be interchanged).

Fraction 2 provided kaempferol 3-0- β -rutinoside that was repeatedly purified by column chromatography on silica gel H (ethyl acetate-ethanol) and Sephadex LH-20 (methanol). It was identified by hydrolytic and spectral data (8).

Fraction 3 yielded kaempferol 3-0- β -rutinoside 7-0- α -rhamnopyranoside (8,9,10), that was purified and characterized as mentioned above.

Treatment of both flavonoids with dichloromethyl methyl ether (11) confirmed the presence of rutinose.

These two flavonoids were also obtained from the ethanolic extract of fresh white flowers of *B. candicans.* The main component of this extract was 3-0-methyl-*D*-inositol (*D*-pinitol). Its spectra and physical data were concordant with those reported (12).

ACKNOWLEDGMENTS

We want to thank Prof. Emilio Ulibarri (Instituto de Botánica Darwinion, Argentina) for the identification of the plant material and Lic. Gustavo Aldomá for ¹H-nmr spectra.

We are also indebted to CONICET (Argentina) and CEFAPRIN (Argentina) for financial support.

LITERATURE CITED

- 1. J. Shani, A. Goldschmied, B. Joseph, Z. Ahronson, and F.G. Sulman, Arch. Int. Pharmacodyn. Ther., 210, 27 (1974).
- 2. G. Weiczel, P. Wahl, and E. Buddecke, Z. Physiol. Chem., 327, 109 (1962).
- 3. D. J. Luzbetak, S.J. Torrance, J.J. Hoffmann, and J.R. Cole, Lloydia, 42, 315 (1979).
- 4. The Sadtler Standard Spectra, Sadtler Res. Lab. ¹H-nmr spectra No. 8809 (1968).
- 5. E. Larsen, H. Egsgaard, and H. Holmen, Org. Mass Spectrom., 13, 417 (1978).
- 6. A.N. Misra and H.P. Tiwari, Phytochemistry, 12, 393 (1973).
- 7. A.K. Dzizenko, V.V. Isakov, N.I. Uvarova, G.I. Oshitok, and G.B. Elyakov, *Carbobyd. Res.*, 27, 249 (1973).
- 8. K.R. Markham, B. Ternai, R. Stanley, H. Geiger, and T.M. Mabry, *Phytochemistry*, **34**, 1389 (1978).
- 9. H. Schels, H.D. Zinsmeister, and K. Pfleger, Phytochemistry, 16, 1019 (1977).
- 10. H.-F. Aly and H. Geiger, Phytochemistry, 14, 1613 (1975).
- 11. H. Gross and I. Farkas, Chem. Ber., 93, 95 (1960).
- 12. D.E. Dorman, S.J. Angyal, and J.D. Roberts, J. Am. Chem. Soc., 92, 1351 (1970).

Received 3 November 1982

THE ALKALOIDS OF HUNNEMANIA FUMARIAEFOLIA

M.A. EL-SHANAWANY,

Department of Pharmacognosy, Faculty of Pharmacy. University of Assiut, Assiut, Egypt

A.M. EL-FISHAWAY, D.J. SLATKIN, and P.L. SCHIFF, JR.

Department of Pharmacognosy, School of Pharmacy, University of Pittsburgh, Pittsburgh, PA 15261

Hunnemania fumariaefolia Sweet (Papaveraceae), also known as the tulip poppy or Golden Cup, is an ornamental herb usually cultivated as a garden plant for its bright yellow flowers (1). Extracts of the tops of this species have been found to inhibit the growth of a variety of microorganisms (2), with this antimicrobial activity subsequently assigned, via bioassay-guided isolation, to the alkaloidal artifacts sanguinarine pseudomethanolate and pseudoethanolate, chelerythrine pseudomethanolate and pseudoethanolate (3). Earlier investigations resulted in the isolation of numerous