D-(+)-Pinitol, a Component of the Heartwood of *Enterolobium cyclocarpum* (Jacq.) Griseb.

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D-(+)-Pinitol, a natural product of the group of cyclitols, was purified for the first time from an aqueous extract of the heartwood of *Enterolobium cyclocarpum*, and its chemical structure was determined.

Key words: D-(+)-Pinitol, *Enterolobium cyclocarpum*, Heartwood

Introduction

Enterolobium cyclocarpum (Jacq.) Griseb. (Leguminosae) is an important agroforest species, with extensive distribution from southern Mexico to northern Brazil. Heartwood, due to its strong resistance to biodegradation, is highly appreciated by the furniture industry. Some extracts with organic solvents from the heartwood have been obtained with toxic effects (Carter *et al*., 1975; Rutiaga Quiñones *et al*., 1995; Dominguez and Franco, 1979) and others contained trypsin inhibitors (Aguilar and Zolla, 1982). However, a phytochemical study to find the bioactive compounds in aqueous extracts of the heartwood of this tropical tree is still missing.

Experimental

General experimental procedure

Optical rotation was measured on a Perkin Elmer 341 polarimeter. 1D and 2D NMR spectra were obtained with a Varian Mercury 400 MHz spectrometer. Column chromatography was performed on silica gel (70–230 mesh, Merck).

Plant material

The wood of *E. cyclocarpum* was collected on July 15, 2000 at El Copalito, Michoacan, Mexico (coordinates: 19° 10′ 53′′ N and 101° 28′ 30′′ W) at an altitude of 1640 m above sea level and identified by M. C. Xavier Madrigal, taxonomist at the UMSNH (voucher 10277).

Extraction and isolation

100 g of air-dried and powdered heartwood were extracted with deionized boiling water (2 × 550 mL) for 20 min. After removal of the water by lyophilization, the resultant extract (56 mg) was subjected to silica gel column chromatography for the separation of pinitol by using hexane as mobile phase. The polarity of the mobile phase was increased by sequentially adding 20 %, 50 %, 80 % ethyl acetate in hexane, then pure ethyl acetate, and finally 20 %, 30 %, 40 % and 50 % methanol in ethyl acetate. Out of 134 fractions collected, fractions 51–67 (5 mg) gave D-(+)-pinitol. NMR spectra were obtained in D_2O, confirmed with pyridine-(d_5), and compared with those reported by Misra and Siddiqi (2004).

Results and Discussion

The following characteristics of D-(+)-pinitol were found: ^{1}H NMR (Fig. 1, 400 MHz, D_2O): δ = 3.85 (2H m, H-1, H-6), 3.66 (1H, dd, $J_{2,3}$ = 9.90 Hz, $J_{2,1}$ = 2.6 Hz, H-2), 3.61 (1H, dd, $J_{5,4}$ = 9.98 Hz, $J_{5,6}$ = 2.6 Hz, H-5), 3.50 (1H, dd, $J_{4,3}$ = 9.53 Hz, $J_{4,5}$ = 9.98 Hz, H-4), 3.45 (3H, s, OMe), 3.19 (1H, dd, $J_{3,2}$ = 9.90 Hz, $J_{3,4}$ = 9.53 Hz, H-3); ^{13}C NMR (Fig. 2, 100 MHz, D_2O): δ = 82.96 (C-3), 72.32 (C-1), 71.89 (C-5), 71.67 (C-2), 70.73 (C-4), 70.02 (C-6), 59.88 (OMe). COSY correlations between the proton signals at H-3 (δ 3.19), H-4 (δ 3.50), H-2 (δ 3.66), between H-2 (δ 3.66) and H-1 (δ 3.85), and between H-4 (δ 3.50) and H-5 (δ 3.61) were found. Furthermore, H-5 coupled with H-6, what allowed to completely elucidate this structure. The ^{13}C NMR chemical shifts of all hydrogenated carbon atoms were assigned unambiguously using the HETCOR spectra. The optical rotation angle of pinitol was $[\alpha]_D^{25}$ = +67° (*c* 0.30, H_2O) [Calle *et al*. (1986): $[\alpha]_D^{25}$ = +65° (*c* 0.4, H_2O)].

D-(+)-Pinitol has been isolated from several plants and its effect on the glucose metabolism is

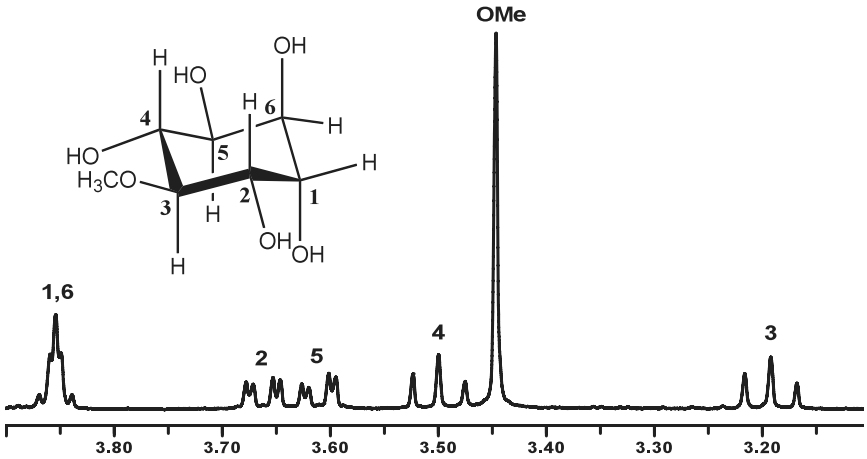

Fig. 1. ^{1}H NMR spectrum of D-(+)-pinitol at 400 MHz in D_2O.

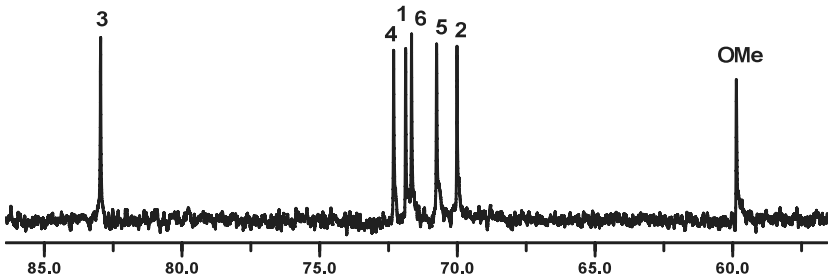

Fig. 2. ^{13}C NMR spectrum of D-(+)-pinitol at 100 MHz in D_2O.

well known (Narayanan *et al*., 1987; Numata *et al*., 1979). Furthermore, an insecticidal effect has been described caused by this cyclitol on the larval growth of *Heliotis zea, Aedes aegypti* and *Culex quinquefasciatus* (Chaubal *et al*., 2005; Dreyer *et al*., 2005). This is the first time that D-(+)-pinitol was isolated from the heartwood of *E. cyclocarpum* (local name parota).

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