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Chemical Studies on the Roots of *Ichnocarpus Frutescens*

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Abstract

Systematic fractionation of ethyl acetate portion of the methanolic extract of defatted roots of Ichnocarpus frutescens led to the isolation of triterpene acid, ursolic acid. This is the first report of the occurrence of ursolic acid from its roots.

.**Keywords**: *Ichnocarpus frutescens*, triterpene acid, ursolic acid, isolation, root

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INTRODUCTION

Sariva has been used to treat a vast range of ailments through ages especially for its blood purifying and cooling attributes. Because of its good fragrance, it is also known as sugandha. In charka samhita , it is classified under the sugandha dravyas (aromatic drugs), varnya dravyas (complexion improving herbs), dahaprasamana (herb relieving burning sensation on the skin) and a potent panacea for Rakta pitta (bleeding disorder and diarrhoea).Sariva traditionally is being given to pregnant women who have a tendency of abortion as this helps to secure their foetal growth with advantage.

Four different plant materials viz *Decalepis hamiltonii, Cryptolepis buchananii Ichnocarpus frutescens* and *Hemidesmus indicus* are indiscriminately sold in the local crude drug market, under the name sariva. *Ichnocarpus frutescens* (Apocynaceae) also known as sarasaparilla is used in the indigenous system of medicine in the treatment of fevers, gout, rheumatism, arthritis, epilepsy, venereal diseases, herpes and skin diseases[1,3,5]. Although chemical investigation has been carried out on the aerial parts of the plant [2, 4], no work is reported from its roots in literature. Chemical investigation conducted on its roots led to isolation of a triterpene acid, ursolic acid hitherto not reported in literature its root.

Plant materials

The roots of *Ichnocarpus frutescens* were collected during the month of June from Tirukovilur, Tamilnadu, India. The plant material was taxonomically identified and authenticated by Prof. P. Jayaraman, Taxonomist, Plant Anatomy Research Centre, Chennai, India. A voucher specimen (PARC/24/06) has been deposited in the Herbarium for future reference.

Extraction and isolation

The plant material (1Kg) was extracted using 95% methanol and the solvent was completely removed from the extract by vacuum distillation to yield a reddish brown residue (54 g). This crude methanolic extract was subjected by partition among petroleum ether (60-80°C), ethyl acetate and chloroform. The ethyl acetate extract was separated by consecutive partitions between diethyl ether- 1% sodium hydroxide (1:2.5 ml, 12 times). The combined organic layer, dried in vacuum, gave fraction 1 (18.4g). The combined alkaline aqueous layers were acidified with acetic acid until pH 5 and then re-extracted with diethyl ether 12 times; the combined diethyl ether layers were dried in vacuum to give fraction II (35.6 g). Fraction II was chromatographed on silica gel using hexane, chloroform and methanol (9:1) eluates, dried, residue crystallized from methanol, ursolic acid (14 g).

RESULT AND DISCUSSION

The compound has MP 280-81°C; soluble in ethanol and chloroform, responded positively with Liebermann-Salkowski reagent (blue colour), EIMS, M+1 at m/z 457.1 which gives M^+ at 456; It analyzed for the molecular formula $C_{30}H_{48}O_3$. λ_{MeOH}^{Max} 205,255 and 293nm; υ_{KBr}^{Max} (cm^{-1}), 3523 (hydroxyl group), 2925 (C=C) and 1714, 1689 (C=O); Double bond equivalent calculation indicate that the molecule has 7 double bond equivalent. Only two of them are accounted for by a carbonyl and an olefinic bond which suggests the molecule to be pentacyclic in nature. with acetic anhydride and triethylamine in cold, is furnished an acetate derivative m.p ; 244-245°C; with benzoyl chloride and sodium hydroxide it afforded a benzoate m.p 215-216°C; with methanol and concentrated sulfuric acid it yielded a methyl ester m.p 173°C. From the above data the compound appeared to be ursolic acid which was further confirmed by its spectral analysis.

υ_{KBr}^{Max} (cm^{-1}), 3523, 2925, 1714, 1689,1456,1377,1029 and 999 cm^{-1}, 1H-NMR ($CDCl_3$, 400 MHz); ppm δ 5.10 (1H, t, *J* = 5 Hz, H-12), 2.98 (1H, dd, *J* = 11.0, 5.1 Hz, H-3α), 2.09 (1H, d, *J* = 11.3 Hz, H- 18), 0.87, 1.02, 0.73 and 0.84 (each 3H, s, H-23, H-27, H-26 and H-24), 0.89 (3H, d, *J* = 6.5 Hz, H-30), 0.79 (3H, d, *J* = 6.4 Hz, H-29) and 0.66 (3H, s, H-25); 13C-NMR (DMSO-d6,100 MHz): ppm: δ 38.3 (C-1), 26.9 (C-2), 76.8 (C-3), 38.4 (C-4), 54.8 (C-5), 18.0 (C-6), 32.7 (C- 7), 38.4 (C-8), 47.0 (C-9), 36.5 (C-10), 22.8(C-11), 124.6 (C-12), 138.2 (C-13), 41.6 (C-14), 27.6 (C-15), 23.8 (16), 46.8 (C-17), 52.4 (C-18), 38.5 (C-19), 38.4 (C-20), 30.2 (C-21), 36.3 (C-22), 28.3 (C-23), 15.2 (C- 24),16.1 (C- 25), 16.9 (C-26), 23.3 (C-27), 178.3 (C- 28), 17.0 (C-29) and 21.1 (C-30). Mass spectrum recorded M+1 at m/z 457 which gives molecular ion peak (M+) at m/z 456 with prominent fragment ions at m/z 248 and m/z 208. Finally the identity of the compound was established to be ursolic acid by direct comparison of m m.p, co-TLC and superimposible IR).

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