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SHORT REPORTS

THE OCCURRENCE OF 4-HYDROXYISOLEUCINE IN STEROIDAL SAPOGENIN-YIELDING PLANTS

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Key Word Index—*Dioscorea deltoidea*; Dioscoreaceae; *Balanites aegyptiaca*; Balanitaceae; *Trigonella foenum-graecum*; Leguminosae; *Solanum laciniatum*: Solanaceae; 4-hydroxyisoleucine; steroidal sapogenin.

Plants. Dioscorea deltoidea tuber purchased in 1967 from Seth Panchhi Ram & Co., Kuth Grower, Manali, Kulu Hills, India. *Balanites aegyptiaca* seed purchased as fruits from Nigeria in 1966 from United Africa Co., Liverpool. *Trigonella foenum-graecum* commercial seed from Morocco, purchased in 1970 from Evans Gray & Hood Ltd., 110 Cannon Street, London, E.C.4; freed from weed seed. *Solanum laciniatum* fruit grown at Bath in 1973 from seed supplied by Searle de Mexico, S.A. de C.V.

Previous work. 4-hydroxyisoleucine from seeds of *Trigonella foenumgraecum* [1]. Fenugreekine, a new steroidal sapogenin-peptide ester of *Trigonella foenum-graecum* [2].

Present work [3]. Dry powdered plant material was extracted with 20% EtOH, solvent removed, residue dissolved in H_2O and purified on cation exchange resin. The amino acid fraction was displaced with *N* NH_3, conc., residue dissolved in H_2O and (2S,3R,4R)-4-hydroxyisoleucine detected by Co-TLC using silica gel and (a) phenol–H_2O (3:1), R_f 0·37 and (b) BuOH–HOAc–H_2O (4:1:1), R_f 0·43 (15 cm). The amount of free 4-hydroxyisoleucine was determined by densitometric TLC using a Joyce Loebl Chromoscan on the transmittance mode 0·06% (*Dioscorea*), 0·03% (*Balanites*), 0·11% (*Solanum*) and 0·74% (*Trigonella*), m.f.b. (2S,3R,4R)-4-Hydroxyisoleucine 200 mg/kg in 4 ml H_2O *per os* in male dutch rabbit: negative hypoglycaemic activity.

Comment. This appears to be the first record of the detection and quantitative determination of free (2S,3R,4R)-4-hydroxyisoleucine in higher plants, other than fenugreek (age of plant material may have reduced yield). All are steroidal sapogenin (or their *N*-analogue)-yielding plants of four distinct families (Dioscoreaceae, Balanitaceae, Leguminosae and Solanaceae) suggesting the co-occurrence of such saponins with 4-hydroxyisoleucine. This also suggests that the sapogenin-peptide ester, fenugreekine [2], which on hydrolysis affords a mixture of three isomeric 4-hydroxyisoleucine lactones [2], is the first example of a class of compounds that will be of common occurrence in steroidal saponin containing plants, including those investigated above and others, such as species of *Digitalis*.

Fenugreekine is reported to have pharmacological activities which include hypoglycaemic and cardiatonic properties [2]. Some amino acids are known to stimulate insulin secretion [4]. In a preliminary test the free (2S,3R,4R)-4-hydroxyisoleucine showed no hypoglycaemic activity.

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THE DEMONSTRATION OF GALACTOSAMINE IN A HIGHER PLANT: *CANNABIS SATIVA**

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Key Word Index—*Cannabis sativa*; Cannabinaceae; marijuana; glycoprotein; amino sugar; galactosamine.

* This investigation was carried out in collaboration with the United Nations Narcotics Laboratory in Geneva, as part of the U.N. Cannabis research programme established by resolution 8 (XIV) of the Commission on Narcotic Drugs.

Plant and source. Cannabis sativa was grown from South African seeds in the Botanical Garden of University of Oslo. Identification of the plants was carried out by Prof A. Nordal, Department of Pharmacognosy, Insti-