UNUSUALLY HIGH RATIO OF SATURATED/UNSATURATED STEROLS IN *TAMARIX GALLICA*

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Tamarix gallica L. (Syn. *T. Troupii* Hole), commonly known as "Jhau," is a shrub of the family Tamaricaceae. The fruits and leaves have been used as an astringent for dysentery and chronic diarrhea, lucoderma, spleen troubles and eye diseases. The steam from cooked leaves is reported to be useful in the treatment of piles, ulcers and wounds (1). Previously, only the 3,3'-dimethyl ether of ellagic acid (2) and polyphenols (3) had been isolated from *T. gallica*. In view of the important medicinal values and the fact that almost no systematic work has been done so far, a chemical study of the stem and leaves of this shrub was undertaken. This paper deals with the composition of sterols in the light petroleum extract.

The aerial parts of *T. gallica*, consisting of a slender stem and minute scaly leaves, were collected from the sandy soil near Ganga (Dist. Bulandshahr, India). The plant material was air-dried, ground and extracted 3 times with light petroleum (bp 60–80°) under reflux for 36 hrs. The extract was chromatographed on a column of activated alumina.

The column, on elution, with benzene-chloroform (3:1), yielded a sterol fraction which, upon crystallization from methanol, gave shiny flakes (1.8% of the extract). This sterol fraction exhibited a positive Liebermann-Burchard test and gave a yellow color with tetranitromethane. The sterol fraction was then acetylated and identification of each component was achieved by comparison with authentic steryl acetate of the Rf on argentation thin layer chromatography (tlc), the relative retention time (RRt) on gas-liquid chromatography (glc), and the mass spectrum on combined gas chromatography-mass spectrometry (gc-ms).

The acetylated sterol fraction was separated by argentation tlc into three zones having the Rf-values 0.87 for the least polar zone (zone-1) and 0.80 for the medium polar zone (zone-2). The Rf of the most polar zone (zone-3) was not measured exactly. The fraction from zone-1 was a mixture of the acetates of 24-methylcholestanol (24ξ-methyl-5α-cholestan-3β-ol) (RRt: OV-1, 1.32; OV-17, 1.33) and 24-ethylcholestanol (24ξ-ethyl-5α-cholestan-3β-ol) (1.63, 1.65). The next fraction from zone-2 consisted of the acetates of cholesterol (cholest-5-en-3β-ol) (1.00, 1.00), 24-methylcholesterol (24ξ-methycholest-5-en-3β-ol) (1.29, 1.31), 24-ethyl-22-dehydrocholestanol (24ξ-ethylcholest-*E*-22-en-3β-ol) (1.44, 1.45), 24-ethylcholesterol (24ξ-ethylcholest-5-en-3β-ol, most probably sitosterol) (1.60, 1.63) and 24-ethyllathosterol (24ξ-ethyl-5α-cholest-7-en-3β-ol) (1.81, 1.92). The fraction from zone-3 was shown to contain the acetates of 24-ethyl-22-dehydrocholesterol (24ξ-ethylcholesta-5,*E*-22-dien-3β-ol, most probably stigmasterol) (1.40, 1.43) and avenasterol (24-ethyl-5α-cholesta-7,*Z*-24[28]-dien-3β-ol) (1.89, 2.15) along with several saturated and monoenic steryl acetates, identified in zones-1 and 2, which presumably resulted from "tailing" of the components on the argentation tlc.

The approximate content of each of these nine sterols in the acetylated total sterol fraction as determined by

glc on OV-1 column was: 24-methylcholestanol (2.7%), 24-ethylcholestanol (24.6%), cholesterol (0.3%), 24-methylcholesterol (3.1%), 24-ethyl-22-dehydrocholestanol (1.0%), 24-ethylcholesterol (60.8%), 24-ethyllathosterol (2.8%), 24-ethyl-22-dehydrocholesterol (4.4%), and avenasterol (0.3%). It is worth mentioning here that the high content of saturated sterols, 27.3% of the total sterols, is a unique finding in plants. Occurrence of such a high proportion of saturated sterols has previously been reported only in the endosperm of *Zea mays*, in the year 1924, in which ca. 15% of 24-ethylcholestanol was found in the sterol mixture (4). The other noteworthy finding in this study is the identification of 24-ethyl-22-dehydrocholestanol. This sterol has previously been identified only in the roots of *Bupleurum falcatum* (5), in the slime mould, *Dictyostelium discoideum* (6,7) and in the seeds of *Lycopersicon esculentum* (8).

EXPERIMENTAL

The following nine authentic sterols (8,9) were used as the acetate derivatives for comparisons by argentation tlc, glc and gc-ms data: 24-methylcholestanol, 24-ethylcholestanol, cholesterol, 24-methylcholesterol, 24-ethyl-22-dehydrocholestanol, sitosterol, 24-ethyllathosterol, stigmasterol and avenasterol. Glc was performed either on OV-1 or OV-17 SCOT glass capillary column (30 m x 0.3 mm i.d.) at 255°, injection at 275°, and nitrogen carrier gas at 0.60 ml/min. (split ratio ca. 100:1). RRt in glc was given relative to cholesteryl acetate. Combined gc-ms (70 eV, $>m/z$ 200; 2% OV-17, 2 m x 3 mm i.d. glass column) was performed by methods described previously (9). The other techniques used in this work have been described earlier (7). The following gc-ms data for the acetates of *T. gallica* sterols, as indicated only by prominent ions, were consistent with those recorded from authentic specimens: 24-methylcholestanyl acetate, m/z 444 (M^+, relative intensity 48), 429 (12), 384 (47), 369 (34), 276 (35), 275 (43), 230 (27), 217 (26), 216 (51), 215 (100) and 201 (20); 24-ethylcholestanyl acetate, m/z 458 (M^+, rel. int. 50), 443 (13), 398 (45), 290 (10), 276 (35), 275 (41), 230 (25), 217 (27), 216 (50), 215 (100) and 201 (20); cholesteryl acetate, m/z 368 (M^+–HOAc, rel. int. 100), 353 (29), 260 (23), 255 (28), 247 (33) and 213 (27); 24-methylcholesteryl acetate, m/z 382 (M^+–HOAc, rel. int. 100), 367 (25), 275 (7), 274 (19), 261 (17), 255 (20) and 213 (18); 24-ethyl-22-dehydrocholestanyl acetate, m/z 456 (M^+, rel. int. 15), 413 (3), 395 (7), 381 (23), 353 (32), 344 (14), 315 (7), 285 (19), 284 (25), 259 (20), 258 (23), 257 (100), 255 (17), 229 (19) and 215 (15); 24-ethylcholesteryl acetate, m/z 396 (M^+–HOAc, rel. int. 100), 381 (23), 288 (16), 275 (17), 255 (21) and 213 (17); 24-ethyllathosteryl acetate, m/z 456 (M^+, rel. int. 100), 441 (22), 396 (18), 381 (16), 315 (10), 288 (10), 273 (14), 255 (79), 229 (30) and 213 (38); 24-ethyl-22-dehydrocholesteryl acetate, m/z 394 (M^+–HOAc, rel. int. 100), 379 (6), 351 (18), 282 (9), 255 (58), 253 (7), 228 (14) and 213 (14); and avenasteryl acetate, m/z 454 (M^+, rel. int. 2), 439 (2), 379 (3), 357 (38), 341 (5), 314 (26), 313 (100), 296 (8), 288 (10), 255 (15), 253 (15), 227 (11) and 213 (20).

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