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FATTY ACIDS AND ESTERS FROM *PANAX PSEUDO-GINSENG* RHIZOMES

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Key Word Index—*Panax pseudo-ginseng*, Araliaceae, rhizomes, 14-hydroxyheneicosanoic acid, dotriacontanyl palmitate, fatty acids, pentatriacontane, sitosterol-β-D-glucoside

Abstract—In addition to methyl palmitate, palmitic acid, stearic acid, eicosanoic acid, triacontanoic acid, hexatriacontanoic acid, pentatriacontane, sitosterol and sitosterol-β-D-glucoside, two new compounds isolated from the rhizomes of *Panax pseudo-ginseng* have been characterized as 14-hydroxyheneicosanoic acid and dotriacontanyl palmitate, respectively, by physico-chemical studies

INTRODUCTION

The *Panax* spp (Araliaceae), *P ginseng* and *P quinquefolium* have carrot-like roots, while *P japonicus* and *P pseudo-ginseng* have long horizontally creeping rhizomes [1] They are reported to be used as tonics and remedies for various ailments Recent years have seen considerable progress in the study of ginseng as the most important non-specific resistance drug Since no work has been done on *P pseudo-ginseng*, found in India, a detailed chemical investigation was undertaken

RESULTS AND DISCUSSION

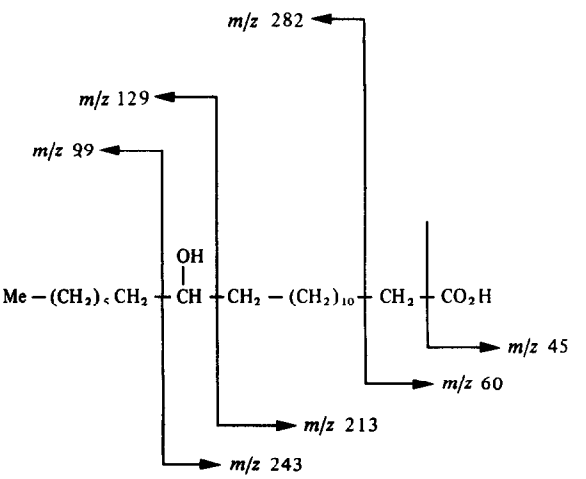
Silica gel CC of the *n*-hexane fraction of the 70% ethanol extracts furnished 11 compounds **1–11** Compound **10**, mp 84° showed IR bands for a hydroxyl group at 3400 cm^{-1} and a carboxyl group at 1705 and 2500–3000 cm^{-1} Its long-chain nature was revealed by the presence of bands at 2920, 2840, 1455 and 715 cm^{-1} The mass spectrum of the compound had a $[M]^+$ at *m/z* 342 suggesting the molecular formula as $C_{21}H_{42}O_3$ The significant ions at *m/z* 45, 60 and the loss of 60 mass units from the $[M]^+$ to give an ion at *m/z* 282 were characteristic of a terminal carboxyl group The ion at *m/z* 60 was due to McLafferty rearrangement [2] involving β-fission of the carboxyl function The rest of the mass spectrum consisted of two series of ions resulting from cleavage of a C–C bond with retention of charge either on an oxygen-containing fragment or on the alkyl fragment (see Experimental) The location of the hydroxyl group at C-14 was determined from the prominent α-fission ions obtained at *m/z* 243, 213, 129 and 99 The ^1H NMR spectrum of **10** showed a triplet, $J = 6$ Hz at δ2 24 for a methylene group adjacent to the carboxyl group and a multiplet at δ3 26 for the –CH(OH)– proton The above data were best accommodated by the structure of 14-hydroxyheneicosanoic acid (**10**)

Compound **2**, mp 85° had IR absorption bands at 2910, 2840, 1455, 720, 710 (long chain), 1380 (methyl), 1725 cm^{-1} (ester group) The mass spectrum of this compound displayed an $[M]^+$ at *m/z* 704 suggesting the molecular formula as $C_{48}H_{96}O_2$ Alkaline hydrolysis of this ester yielded an acid, mp 65°, $[M]^+$ *m/z* 256, $C_{16}H_{32}O_2$, identified as palmitic acid by comparison with an authentic specimen, and an alcohol, mp 86° identified as dotriacontanol by comparison with an authentic sample [3] Compound **2**, therefore, was characterized as dotriacontanyl palmitate

Compound **1**, mp 71°, was identified as pentatriacontane by IR, mass spectrometry and comparison with authentic material [4] Compound **3** was hydrolysed to give palmitic acid and identified as methyl palmitate by comparison with authentic material (IR, mass spectrum and co-TLC) Compounds **4**, mp 65°, **6**, mp 70°, **7**, mp 75°, **8**, mp 90° and **9**, mp 88° were identified as palmitic acid, stearic acid, eicosanoic acid, triacontanoic acid and hexatriacontanoic acid, respectively, by comparison with authentic samples, whereas compounds **5**, mp 137° and **11**, mp 280–285° were identified, respectively, as sitosterol and its β-D-glucoside

EXPERIMENTAL

Mps are uncorr IR spectra were recorded in KBr and the 60 MHz ^1H NMR spectrum was measured in $CDCl_3$ with



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TMS as int standard TLC was carried out on silica gel G in at least three different solvent systems and the spots were visualized by exposure to I_2 vapour Plant material was collected from the Tung area of Darjeeling, West Bengal and identified in the Botany Department of this Institute where a voucher specimen has been deposited

Extraction and isolation of compounds Dried and powdered rhizomes (268 g) of *P pseudo-ginseng* Wall were extracted with EtOH (70%, 9 × 600 ml) The solvent was removed *in vacuo* and the residue dissolved in EtOH (70%, 300 ml) It was then extracted successively with *n*-hexane (6 × 200 ml) and *n*-BuOH (7 × 200 ml) Removal of solvent from the hexane extract gave a residue (3 g) which was chromatographed over silica gel (65 g, 60–120 mesh, BDH) Elution was carried out in hexane, hexane–C_6H_6 (3 1, 1 1, 1 3), C_6H_6, C_6H_6–$CHCl_3$ (3 1, 1 1, 1 3), $CHCl_3$, $CHCl_3$–MeOH (19 1) and MeOH Fractions (25 ml each) were collected and each was monitored by TLC

Pentatriacontane (**1**) Removal of solvent from hexane fractions (1–4) afforded a residue, 5 mg, mp 71° (Me_2CO), identified by mmp, co-TLC, IR, MS

Dotriacontanyl palmitate (**2**) Removal of solvent from hexane fractions (6–14) furnished a residue, 17 mg, mp 85° (Me_2CO) IR ν_{max} cm^{-1} 2910, 2840, 1725, 1455, 1380, 1250, 720, 710 MS *m/z* (rel int) 704 $[M]^+$ ($C_{48}H_{96}O_2$) (0 2), 448 (2), 420 (4), 257 (1), 256 (2), 127 (27), 113 (32), 99 (37), 85 (67), 71 (83), 57 (100), 43 (58) The compound (10 mg) was refluxed with 5% EtOH–KOH (10 ml) for 5 hr At the end of the reaction the mixture was diluted with H_2O (50 ml) and extracted with Et_2O (4 × 25 ml) The extract was washed with H_2O (2 × 25 ml) and dried (Na_2SO_4) Removal of solvent gave a residue, mp 86° (Me_2CO–hexane), identified as dotriacontanol (mmp, IR, MS, co-TLC) MS *m/z* (rel int) 448 $[M-H_2O]^+$ (1), 420 $[M-46]^+$ (2), 392 (1), 364 (1), 336 (1), 308 (1), 280 (1), 252 (1), 224 (2), 196 (2), 168 (3), 140 (5), 127 (8), 113 (8), 99 (10), 85 (35), 71 (55), 57 (100), 43 (80), 41 (30) The aq layer remaining after the above extraction was acidified with dil HCl and extracted with Et_2O (4 × 25 ml) The extract was washed with H_2O (2 × 25 ml) and dried (Na_2SO_4) Removal of solvent provided a residue, mp 65° ($CHCl_3$–MeOH), identified as palmitic acid (mmp, IR, co-TLC, MS) IR ν_{max} cm^{-1} 3000–3500, 2910, 2840, 1700, 1460, 1285, 930, 720, 710 MS *m/z* (rel int) 256 $[M]^+$ ($C_{16}H_{32}O_2$) (3), 196 $[M-60]^+$ (5), 73 (100), 60 (99), 45 (12), 44 (5)

Methyl palmitate (**3**) Eluates from hexane–C_6H_6 (3 1) fractions (16–24) when freed of solvent gave a viscous residue, 20 mg MS *m/z* (rel int) 270 $[M]^+$ ($C_{17}H_{34}O_2$) (10), 256 (1), 241 (5), 239 (4), 227 (10), 213 (4), 211 (1), 199 (9), 185 (9), 171 (8), 157 (5), 143 (10), 129 (10), 115 (8), 101 (10), 87 (99) 74 $[CH_2{=}C(OH)OMe]^+$ (99), 59 (25), 55 (90), 43 (100) It was identified by IR, co-TLC and hydrolysis product, mp 65°

Palmitic acid (**4**) Hexane–C_6H_6 (1 1) fractions (57–72) provided a residue, 15 mg, mp 65° identified by co-TLC, mmp, IR, MS

Sitosterol (**5**) Eluates (75–96) from hexane–C_6H_6 fractions (1 1, 1 3) afforded a residue, 50 mg, mp 137° identified by mmp, co-TLC, IR, MS

Stearic acid (**6**) Removal of solvent from C_6H_6 fractions (108–118) gave a residue, 10 mg, mp 70° (MeOH). MS *m/z* (rel int) 284 $[M]^+$ ($C_{18}H_{36}O_2$) (1), 240 (1), 239 (1), 224 $[M-60]^+$ (1), 60 (12), 57 (100), 55 (65), 46 (80), 45 (70), 44 (8), 43 (88) It was identified by co-TLC, mmp, IR

Eicosanoic acid (**7**) Fractions (121–132) from C_6H_6 eluates furnished a residue, 15 mg, mp 75° ($CHCl_3$–MeOH) MS *m/z* (rel int) 312 $[M]^+$ ($C_{20}H_{40}O_2$) (5), 268 $[M-44]^+$ (3), 267 $[M-45]^+$ (2), 252 $[M-60]^+$ (3), 60 (12), 57 (46), 55 (100), 45 (28), 44 (20), 43 (80) It was identified by mmp, co-TLC, IR

Triacontanoic acid (**8**) Fractions (133–142) eluted from C_6H_6 provided a residue, 10 mg, mp 90° ($CHCl_3$–MeOH) identified by mmp, IR, co-TLC MS *m/z* (rel int) 452 $[M]^+$ ($C_{30}H_{60}O_2$) (1)

Hexatriacontanoic acid (**9**) Eluates 144–164 from C_6H_6–$CHCl_3$ (3 1) gave a residue, 5 mg, mp 88° ($CHCl_3$–MeOH) identified by mmp, co-TLC, IR MS *m/z* (rel int) 536 $[M]^+$ ($C_{36}H_{72}O_2$) (3)

14-*Hydroxyheneicosanoic acid* (**10**) Removal of solvent from C_6H_6–$CHCl_3$ (1 1) fractions (165–184) afforded a residue, 21 mg, mp 84° ($CHCl_3$–MeOH) IR ν_{max} cm^{-1} 3400, 2500–3000, 2930, 2840, 1705, 1455, 1370, 1240, 1160, 715 ^1H NMR δ0 80 (3H, *t*, *J* = 6 Hz, H_3-21), 1 20 (30H, *br s*, $(CH_2)_{15}$), 2 24 (2H, *t*, *J* = 6 Hz, H_2-2), 2 08 (4H, *m*, H_2-13, H_2-15), 3 26 (1H, *m*, H-14) MS *m/z* (rel int) 342 $[M]^+$ ($C_{21}H_{42}O_2$) (1), 282 (1), 243 (6), 213 (10), 129 (15), 101 (8), 99 (8), 87 (8), 85 (12), 73 (35), 71 (30), 60 (30), 59 (11), 57 (65), 45 (18), 44 (21), 43 (88), 41 (100)

Sitosterol-β-D-glucoside (**11**) Fractions obtained from MeOH eluates were freed of the solvent to provide a residue, 10 mg, mp 280–285°, identified by IR, MS, co-TLC, mmp and from hydrolysis products

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