

## SHORT COMMUNICATION

# STRUCTURE OF NODOLIDATE FROM THE FLOWERS OF *CASSIA NODOSA*

S. A. I. RIZVI, SURESH C. VARSHNEY, S. L. ABBAS\* and N. JAHAN

Chemical Laboratories, University of Allahabad, Allahabad, India

(Received 19 October 1971)

**Abstract**—A new compound, nodolidate, has been isolated from the flowers of *Cassia nodosa* and characterized as (—)-7-acetoxy-9,10-dimethyl-1,5-octacosanolide. A mixture of fatty acids, hydrocarbons, campesterol and sitosterol along with ceryl alcohol were also isolated.

*Cassia nodosa*, a plant of the medicinal<sup>1,2</sup> genus *Cassia* (Leguminosae, subfamily Caesalpinioideae), has insecticidal activity,<sup>3</sup> contains a fixed oil<sup>4</sup> and a galactomannan<sup>5</sup> in the seeds, a new anthraquinone glycoside<sup>6</sup> and a mixture of phytosterolins and sterols<sup>7</sup> in the flowers. During the present investigation nodolidate was isolated from the ether insoluble fraction of a benzene extract of the flowers whereas the ether soluble portion contained a mixture of acids and an unsaponifiable fraction. The acids were palmitic, stearic, arachidic, behenic, lignoceric, oleic and linoleic acids. Presence of linolenic acid in a small quantity could only be recorded by spectrophotometry after alkali isomerisation. The unsaponifiable matter yielded a 'γ-sitosterol' and ceryl alcohol after column chromatography on alumina; they were identified by m.m.p. and IR comparison with authentic samples. 'γ-Sitosterol' is known to be mixture<sup>8-10</sup> and our material was identified by m.p. as an approximately equal mixture of campesterol and sitosterol.

Nodolidate,  $C_{32}H_{60}O_4$ , m.p. 69–70°, was found to be an optically active compound containing one acetyl and three C-methyl groups; IR spectra indicated it to be either an aliphatic (saturated) ester or a lactone. Saponification followed by the acidification yielded a monohydroxy lactone,  $C_{30}H_{58}O_3$ , m.p. 86–87°,  $\nu_{\max}^{KBr}$  3450 (OH) and 1725  $cm^{-1}$  ( $\delta$  or higher lactone); formed a monoacetate,  $C_{32}H_{60}O_4$ , identical to nodolidate (m.m.p. 69–70°). The hydroxylactone was therefore named nodolidol; its IR spectra showed the presence of a secondary alcoholic group ( $\nu_{\max}^{KBr}$  1390, 1278 and 1120  $cm^{-1}$ ). Oppenauer oxidation of nodolidol produced nodolidone,  $C_{30}H_{56}O_3$ , m.p. 58–59°, which formed a

\* Present address: Department of Agronomy, Allahabad Agricultural Institute, Allahabad.

<sup>1</sup> K. R. KIRTIKAR and B. D. BASU, *Indian Medicinal Plants* 2nd Edn, Vol. II, pp. 854–79, Lalit Mohan Basu, Allahabad (1933).

<sup>2</sup> R. N. CHOPRA, S. L. NAYAR and I. C. CHOPRA, *Glossary of Indian Medicinal Plants*, pp. 53–55, CSIR, India (1956).

<sup>3</sup> H. K. PLANK, *J. Econ. Entomol.* **37**, 737 (1944).

<sup>4</sup> S. A. I. RIZVI, P. C. GUPTA, K. C. SRIVASTAVA and R. K. KAUL, *Planta Med.* **16**, 317 (1968).

<sup>5</sup> S. A. I. RIZVI, P. C. GUPTA and R. K. KAUL, *Planta Med.* **20**, 24 (1971).

<sup>6</sup> S. A. I. RIZVI, P. C. GUPTA and R. K. KAUL, *Planta Med.* **19**, 222 (1970).

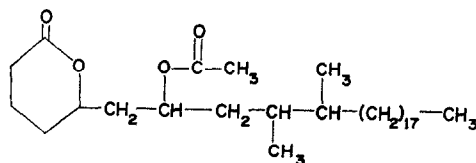
<sup>7</sup> S. A. I. RIZVI, JAWAHAR LAL and P. C. GUPTA, *Phytochem.* **10**, 670 (1971).

<sup>8</sup> W. J. A. VANDEN HEUVEL, E. O. HAAHTI and E. C. HORNING, *J. Am. Chem. Soc.* **83**, 1513 (1961).

<sup>9</sup> W. J. THOMSON, W. E. ROBBINS and G. L. BAKER, *Steroids* **2**, 505 (1963).

<sup>10</sup> I. NISHIOKA, N. IKEKAWA, A. YAGI, T. KAWASSAKI and T. TSUKAMOTO, *Chem. Pharm. Bull. Tokyo*, **13**, 379 (1965).

2,4-dinitrophenylhydrazone, m.p. 130–131° and an oxime, m.p. 79–80°. Reduction of nodolidone by Huang-Minlon<sup>11</sup> modification of the Wolff Kishner method yielded nodolide,  $C_{30}H_{58}O_2$ , m.p. 77–78°; IR spectra showed absorptions for an aliphatic  $\delta$  or higher lactone. Treatment of nodolide with Na–Hg in acid solution produced a monobasic acid,  $C_{30}H_{60}O_2$ , m.p. 54–55°, which was named as nodoic acid. From the positive optical rotation and the mass spectral studies of the methyl ester, which showed the presence of a methyl group each on  $C_9$  and  $C_{10}$ , the structure of nodoic acid was established as (+)-9,10-dimethyloctacosanoic acid. Reduction of nodolide with  $LiAlH_4$  produced an optically active diol,  $C_{30}H_{62}O_2$ , m.p. 92–93°. Oxidation of nodolide as well as the diol with alkaline  $KMnO_4$  produced the same two acids in major yields. Acid I,  $C_{25}H_{50}O_2$ , m.p. 62–63°,  $[\alpha]_D^{20} + 16^\circ$  ( $CHCl_3$ ) formed an amide and a methyl ester. Mass spectral studies of the methyl ester showed the presence of one methyl group each on  $C_4$  and  $C_5$  and therefore the structure of acid I was established as (+)-4,5-dimethyltricosanoic acid. Acid II,  $C_5H_8O_4$  (dibasic), m.p. 96–97°, was identified as glutaric acid by co-chromatography and m.m.p. determination with an authentic sample and also through the preparation of diamide. Thus the structures for nodolide and the diol were assigned as 9,10-dimethyl-1,5-octacosanolide and 9,10-dimethyloctacosane-1,5-diol respectively. Reduction of nodolidol with  $LiAlH_4$  gave a triol,  $C_{30}H_{62}O_3$ , m.p. 100–101°. Alkaline  $KMnO_4$  oxidation of nodolidol as well as the triol gave the same two acids, namely glutaric and (+)-2,3-dimethylheneicosanoic<sup>12,13</sup> (m.p. 65–66°) acids. Thus nodolidol is either 9,10-dimethyl-6-hydroxy-1,5-octacosanolide or 9,10-dimethyl-7-hydroxy-1,5-octacosanolide. Since the triol is highly resistant to periodate oxidation, it lacks vic hydroxyl groups and therefore the first possibility can be ruled out. The structures of the triol and nodolidone are established as 9,10-dimethyloctacosane-1,5,7-triol and 9,10-dimethyl-7-keto-1,5-octacosanolide respectively. The NMR spectrum of nodolidate showed a (9H) signal at 9.12  $\tau$  for three  $-CH_3$  groups, a large (36H) signal at 8.74  $\tau$  for 18 methylene groups, a (2H) signal at 8.56  $\tau$  for two  $\equiv CH$  (aliphatic) groups, a (4H) signal at 8.40  $\tau$  for two  $-CH_2-\dot{C}H-O-$  groups, a (2H) signal at 8.22  $\tau$  for  $-O=\dot{C}H-CH_2-\dot{C}H=O-$  group, a (3H) signal at 7.86  $\tau$  for  $CH_3-COO-$  group, a (2H) signal at 6.72  $\tau$  for  $-CH_2-COO-$  group, a (1H) signal at 5.20  $\tau$  for a  $=CH-O-$  group and a (1H) signal at 4.90  $\tau$  for  $=CH-O-CO-$  group of a lactone. Thus from the foregoing chemical and spectral studies and the negative optical rotation,  $[\alpha]_D^{27} - 12.3^\circ$ , nodolidate is finally assigned the structure of (–)-7-acetoxy-9,10-dimethyl-1,5-octacosanolide (I).



Nodolidate (I)

## EXPERIMENTAL

M.ps. are uncorrected and specific rotations are equal values.

*Isolation and purification of nodolidate.* The benzene extracts of the dried *C. nodosa* flowers (12.5 kg) on concentration yielded a semisolid mass (1.56%) which was resolved into  $Et_2O$  sol and insol fractions. The

<sup>11</sup> HUANG-MINLON, *J. Am. Chem. Soc.* **68**, 2487 (1946).

<sup>12</sup> S. STÄLLBERG-STENHAGEN, *Ark. Kemi* **3**, 267 (1951).

<sup>13</sup> E. RYHAGE and E. STENHAGEN, *Ark. Kemi* **15**, 333 (1960).

Et<sub>2</sub>O insol brown solid was repeatedly extracted with CHCl<sub>3</sub> and the extracts after treatment with animal charcoal and boiling with Fuller's earth yielded a solid. Column chromatography on Al<sub>2</sub>O<sub>3</sub> in CHCl<sub>3</sub> gave a colourless solid, which after several crystallizations from MeOH-CHCl<sub>3</sub> afforded flakes of nodolidate (12.2 g), m.p. 69–70°, homogeneous by TLC in different solvents.

**Nodolidate.** IR spectrum showed  $\nu_{\text{max}}^{\text{KBr}}$  2910, 2840, 1735, 1455, 1170 and 715 cm<sup>-1</sup>;  $[\alpha]_D^{27}$  -12.3° (CHCl<sub>3</sub>) (Found: C, 75.42; H, 11.88; C-methyl, 8.50 (corrected for acetyl %); acetyl, 8.04%; neut equiv., 260. C<sub>32</sub>H<sub>60</sub>O<sub>4</sub> requires: C, 75.59; H, 11.81; C-methyl, 8.86; 1 acetyl, 8.46%, neut equiv., 254). NMR  $\tau$  9.12 (9H), 8.74 (36H), 8.56 (2H), 8.40 (4H), 8.22 (2H), 7.86 (3H), 6.72 (2H) and 4.90 (1H). Saponification of the compound (10.8 g) with alc KOH (2 N) for 20 hr at 100° followed by the distillation of solvent, acidification of the soap and extraction with CHCl<sub>3</sub> resulted in the isolation of nodolidol, purified by column chromatography on Al<sub>2</sub>O<sub>3</sub> in CHCl<sub>3</sub>.

**Characterization of nodolidol.** Several crystallizations from MeOH-CHCl<sub>3</sub> afforded flakes (7.86 g), m.p. 86–87°,  $[\alpha]_D^{29}$  -9.4° (CHCl<sub>3</sub>);  $\nu_{\text{max}}^{\text{KBr}}$  3450 (OH), 2910, 2850, 1725 (—COO—), 1390, 1278, 1120 and 720 cm<sup>-1</sup>; NMR  $\tau$  9.16 (9H), 8.80 (36H), 8.56 (2H), 8.45 (4H), 8.26 (2H), 6.75 (2H), 6.12 (1H), 5.32 (1H) and 4.92 (1H). (Found: C, 77.08; H, 12.52; C-methyl, 9.06%; neut equiv., 470. C<sub>30</sub>H<sub>58</sub>O<sub>3</sub> requires: C, 77.25; H, 12.45; 3 C-methyl, 9.66%; neut equiv., 466.) Acetate prepared by Ac<sub>2</sub>O-pyr method was found to be identical to nodolidate by m.p. and m.m.p. (69–70°).

Oppenauer oxidn of nodolidol (4.5 g) gave a product, after on column chromatography (Al<sub>2</sub>O<sub>3</sub>) and elution with benzene-CHCl<sub>3</sub>, as waxy flakes from MeOH-CHCl<sub>3</sub> of nodolidone (3.38 g), m.p. 58–59°,  $[\alpha]_D^{29}$  -5° (CHCl<sub>3</sub>);  $\nu_{\text{max}}^{\text{KBr}}$  2920, 2840, 1730 (C=O, enhanced), 1460, 1385, 1155 and 718 cm<sup>-1</sup> (Found: C, 77.50; H, 12.28; C-methyl, 9.46%; neut equiv., 468. C<sub>30</sub>H<sub>56</sub>O<sub>3</sub> requires: C, 77.59; H, 12.07; 3C-methyl, 9.70%; neut equiv., 464.) The compound formed a 2,4-dinitrophenylhydrazone, m.p. 130–131° (MeOH) and an oxime, m.p. 79–80° (aq. EtOH), by refluxing it with NH<sub>2</sub>OH.HCl and KOH in 95% EtOH for 20 hr. (Found: C, 75.02; H, 11.95; N, 2.80. C<sub>30</sub>H<sub>57</sub>O<sub>3</sub>N requires: C, 75.16; H, 11.90; N, 2.92%.)

Reduction of nodolidone (~2.5 g) carried out by Huang-Minlon<sup>11</sup> modification of the Wolff Kishner method with hydrazine hydrate and KOH in ethylene glycol yielded a syrupy product which was purified by column chromatography on Al<sub>2</sub>O<sub>3</sub> and eluted with CHCl<sub>3</sub>. Several crystallizations from MeOH-CHCl<sub>3</sub> afforded flakes (1.98 g) of nodolide, m.p. 77–78°,  $[\alpha]_D^{30}$  -6.4° (CHCl<sub>3</sub>);  $\nu_{\text{max}}^{\text{KBr}}$  2925, 2860, 1720 (C=O of  $\delta$  or higher lactone), 1460, 1375, 1180 and 725 cm<sup>-1</sup>; NMR  $\tau$  9.20 (9H), 8.76 (40H), 8.48 (2H), 8.30 (4H), 6.68 (2H, —CH<sub>2</sub>—CO—) and 4.92 (1H, =CH—O—CO— of  $\delta$ -lactone). (Found: C, 79.82; H, 12.96; C-methyl, 9.48; neut equiv., 456. C<sub>30</sub>H<sub>58</sub>O<sub>2</sub> requires: C, 80.00; H, 12.89; 3C-methyl, 10.00%; neut equiv., 450.)

Nodolide (~0.4 g) was treated with Na-Hg in acid soln and the product, after purification on SiO<sub>2</sub>-gel, was crystallized from MeOH as waxy flakes (310 mg) of nodoic acid, m.p. 54–55°,  $[\alpha]_D^{30}$  +12.2° (CHCl<sub>3</sub>);  $\nu_{\text{max}}^{\text{KBr}}$  3250–3200 (broad, OH), 2920, 2850, 2725, 1710 (COOH), 1460, 1380, 1260, 1130 and 720 cm<sup>-1</sup>; NMR  $\tau$  9.04 (9H), 8.72 (44H), 8.38 (2H), 8.16 (2H, —CH<sub>2</sub>—CH<sub>2</sub>—COOH) and -0.96 (1H, —COOH). (Found: C, 79.56; H, 13.20; C-methyl, 9.66%; neut equiv., 454. C<sub>30</sub>H<sub>60</sub>O<sub>2</sub> requires: C, 79.64; H, 13.27; 3C-methyl, 9.96%, neut equiv., 452.) It formed an amide, m.p. 95–96° (aq. EtOH). (Found: C, 79.68; H, 13.80; N, 2.98%. C<sub>30</sub>H<sub>61</sub>ON requires: C, 79.82; H, 13.53; N, 3.10%) and a Me-ester, m.p. 23–24° (Me<sub>2</sub>CO at 0°). MS of Me-ester: *m/e* 74 (CH<sub>3</sub>CO—C(OH<sup>+</sup>)=CH<sub>2</sub>), 87 (CH<sub>3</sub>OCO—CH<sub>2</sub>—CH<sub>2</sub>), 101 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>2</sub>—CH<sub>2</sub>), 115 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>3</sub>—CH<sub>2</sub>), 129 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>4</sub>—CH<sub>2</sub>), 143 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>5</sub>—CH<sub>2</sub>), 157 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>6</sub>—CH<sub>2</sub>), 185 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>7</sub>—CH—CH<sub>3</sub>), 253 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>16</sub>—CH<sub>2</sub>), 281 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>17</sub>—CH—CH<sub>3</sub>), 309 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>17</sub>—CH(CH<sub>3</sub>)—CH—CH<sub>3</sub>), 423 (M<sup>+</sup>-43), 435 (M<sup>+</sup>-31) (acylium ion), 437 (M<sup>+</sup>-29) and 466 (M<sup>+</sup>). The peaks at *m/e* 253, 281 and 309 with the absence of significant peaks at *m/e* 267 and 295 indicate the presence of two methyl side chains, one on each of the two vicinal C atoms. Further, the peaks at *m/e* 157 and 185 with the absence of a significant peak at 171 show the presence of the first methyl side chain at C<sub>9</sub>.

Nodolide (1 g) was oxidized with 50 ml 10% KMnO<sub>4</sub> and 20 ml 12% KOH for ~2 hr at ~50° and the products were isolated in the usual way.<sup>14</sup> Acid I (H<sub>2</sub>O insol) was purified on SiO<sub>2</sub> gel and crystallized from MeOH as flakes (600 mg), m.p. 62–63°,  $[\alpha]_D^{30}$  +16° (CHCl<sub>3</sub>);  $\nu_{\text{max}}^{\text{KBr}}$  3150 (OH) 2910, 2850, 2720, 1700 (COOH), 1460, 1370, 1270, 1135 and 725 cm<sup>-1</sup>, NMR  $\tau$  9.02 (9H), 8.75 (34H), 8.40 (2H), 8.20 (2H, —CH<sub>2</sub>—CH<sub>2</sub>—COOH), 7.58 (2H, —CH<sub>2</sub>—COOH) and -0.86 (1H, —COOH). (Found: C, 78.40; H, 13.31; C-methyl, 11.52%; neut equiv., 378. C<sub>25</sub>H<sub>50</sub>O<sub>2</sub> requires: C, 78.53; H, 13.09; 3 C-methyl, 11.78%; neut equiv., 382.) It formed an amide, m.p. 102–103° (aq. MeOH) (Found: C, 78.65; H, 13.52; N, 3.56%. C<sub>25</sub>H<sub>51</sub>ON requires: C, 78.74; H, 13.39; N, 3.67%) and a Me-ester, m.p. 25–26° (Me<sub>2</sub>CO at 0°). MS of Me-ester showed significant peaks at *m/e* 74, 87 (CH<sub>3</sub>OCO—CH<sub>2</sub>CH<sub>2</sub>), 115 (CH<sub>3</sub>OCO—(CH<sub>2</sub>)<sub>2</sub>—CH—CH<sub>3</sub>), 253 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>16</sub>—CH<sub>2</sub>), 281 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>17</sub>—CH—CH<sub>3</sub>), 309 (CH<sub>3</sub>—(CH<sub>2</sub>)<sub>17</sub>—CH(CH<sub>3</sub>)—CH—CH<sub>3</sub>), 323 (M<sup>+</sup>-73), 339 (M<sup>+</sup>-57), 365 (M<sup>+</sup>-31), 367 (M<sup>+</sup>-29) and 396 (M<sup>+</sup>). The peaks at 367 (M<sup>+</sup>-29) and 339 (M<sup>+</sup>-57) are due to elimination of C<sub>2</sub> and C<sub>3</sub>, and C<sub>2</sub>, C<sub>3</sub> and C<sub>4</sub> with the attached methyl side chain at C<sub>4</sub> respectively

<sup>14</sup> S. C. VARSHNEY, S. A. I. RIZVI and P. C. GUPTA, *Planta Med.* (in press) (1972).

and are evidence for the terminal isopropyl structure. Acid II ( $\text{H}_2\text{O}$  sol) was recovered by  $\text{Et}_2\text{O}$  extraction of the aq. reaction mixture, purified (CC) and crystallized from light petroleum as crystals (160 mg), m.p. 96–97°; amide, m.p. 173–174° (aq. EtOH); anhydride prepared by  $\text{Ac}_2\text{O}$ , m.p. 55° ( $\text{Et}_2\text{O}$ ). (Found: C, 45.33; H, 6.20%; neut equiv., 68. Calc for  $\text{C}_5\text{H}_8\text{O}_4$ : C, 45.46; H, 6.06%; neut equiv., 66.) It was identified as glutaric acid by m.m.p. and co-TLC with an authentic sample.

Reduction of nodolide (0.32 g) with  $\text{LiAlH}_4$  gave a product which after purification on  $\text{Al}_2\text{O}_3$  was crystallized from MeOH (yield 240 mg), m.p. 92–93°,  $[\alpha]_D^{25} -10.2^\circ$  ( $\text{CHCl}_3$ ),  $\nu_{\text{max}}^{\text{KBr}}$  3550 (OH). (Found: C, 79.02; H, 13.70; C-methyl, 9.50%;  $\text{C}_{30}\text{H}_{62}\text{O}_2$  requires: C, 79.30; H, 13.66; 3C-methyl, 9.91%.) It formed an acetate ( $\text{Ac}_2\text{O}$ – $\text{NaOAc}$  method), m.p. 71–72° (MeOH– $\text{CHCl}_3$ ). (Found: C, 75.56; H, 12.36; acetyl, 15.43%.  $\text{C}_{34}\text{H}_{66}\text{O}_4$  requires: C, 75.84; H, 12.27; 3 acetyl, 15.99%.) Alk  $\text{KMnO}_4$  oxidn of the diol (100 mg) gave acids I and II as confirmed by m.m.p.

Nodolidol (1.5 g) was oxidized by alk  $\text{KMnO}_4$  as nodolide and the oxidn products were isolated and purified on  $\text{SiO}_2$  gel columns. Acid III (680 mg) ( $\text{H}_2\text{O}$  insol) was crystallized from EtOH, m.p. 66–67°,  $[\alpha]_D^{25} +20.2^\circ$  ( $\text{CHCl}_3$ ),  $\nu_{\text{max}}^{\text{KBr}}$  1705  $\text{cm}^{-1}$  (COOH). (Found: C, 77.76; H, 13.22; C-methyl, 12.39%; neut equiv., 357.  $\text{C}_{23}\text{H}_{46}\text{O}_2$  requires: C, 77.97; H, 13.00; 3C-methyl, 12.71%; neut equiv., 354.) It formed an amide, m.p. 117–118° (aq. EtOH). (Found: C, 77.97; H, 13.40; N, 3.78%.  $\text{C}_{23}\text{H}_{47}\text{ON}$  requires: C, 78.18; H, 13.32; N, 3.96%) and a Me-ester, m.p. 26–27° (EtOH at 0°). MS of the ester showed significant peaks at:  $m/e$  88 ( $\text{CH}_3\text{O}-\text{C}(\text{OH}^+)=\text{CH}-\text{CH}_3$ ), 281 ( $\text{CH}_3-(\text{CH}_2)_{17}-\text{CH}-\text{CH}_3$ ), 297 ( $\text{M}^+-71$ , elimination<sup>15</sup> of  ${}^2_2\text{CH}(\text{CH}_3)-{}_3\text{CH}(\text{CH}_3)-{}_4\text{CH}_2$ , + H), 309 ( $\text{CH}_3-(\text{CH}_2)_{17}-\text{CH}(\text{CH}_3)-\text{CH}-\text{CH}_3$ ), 339 ( $\text{M}^+-57$ , elimination of  ${}^2_2\text{CH}(\text{CH}_3)-{}_3\text{CH}(\text{CH}_3)+\text{H}$ ), 365 ( $\text{M}^+-31$ ) and 396 ( $\text{M}^+$ ). Thus acid III was characterized as (+)-2,3-dimethylheneicosanoic acid<sup>12,13</sup> although direct comparison could not be made. Acid IV (260 mg) ( $\text{H}_2\text{O}$  sol), m.p. 96° (benzene), was identified as glutaric acid by m.m.p., co-TLC with an authentic sample and through the preparation of amide and anhydride.

Reduction of nodolidol (1 g) with  $\text{LiAlH}_4$  gave a product which was purified by chromatography and crystallized from EtOH– $\text{CHCl}_3$  as needles (630 mg), m.p. 100–101°,  $[\alpha]_D^{30} -15.8^\circ$  ( $\text{CHCl}_3$ ),  $\nu_{\text{max}}^{\text{KBr}}$  3500 (OH), (Found: C, 76.52; H, 13.27; C-methyl, 9.26%.  $\text{C}_{30}\text{H}_{62}\text{O}_3$  requires: C, 76.60; H, 13.19; 3C-methyl, 9.57%.) Acetate prepared by  $\text{Ac}_2\text{O}$ – $\text{NaOAc}$  had m.p. 78–79° (EtOH). (Found: C, 72.26; H, 11.39; acetyl, 21.27%.  $\text{C}_{36}\text{H}_{68}\text{O}_6$  requires: C, 72.48; H, 11.41; 3 acetyl, 21.64%.) The triol (200 mg) was oxidized by alk  $\text{KMnO}_4$  and the oxidn products were isolated as  $\text{H}_2\text{O}$  insol and sol portions, purified by chromatography, crystallized and identified as acids III and IV resp. by m.m.p.

*Examination of  $\text{Et}_2\text{O}$  sol fraction.*  $\text{Et}_2\text{O}$  sol fraction (~166 g) of the benzene extract of *C. nodosa* flowers after necessary purification, was saponified and separated into a mixture of fatty acids and unsaponifiable matter. The mixed acids (~149 g) were studied for the composition by the combined application of liquid–solid countercurrent distribution of urea inclusion complexes<sup>4,16</sup> of (a) mixed acids and (b) solid and liquid acid fractions separated by Pb-salt method, reversed phase PPC<sup>17–19</sup>, TLC<sup>20</sup> and spectrophotometry.<sup>21,22</sup> The fatty acids found were: palmitic, 8.18, 8.64; stearic, 12.34, 12.81; arachidic, 1.63, 1.50; behenic, 1.02, 1.26; lignoceric, 0.72, 0.68; oleic, 48.34, 47.71; linoleic, 27.77, 27.40 and linolenic acid 0.82% ( $\text{E}^{1\%}_{1\text{cm}}$  in EtOH at 2340 Å 7.15).

The unsaponifiable matter (~6 g) was separated into three fractions on  $\text{Al}_2\text{O}_3$  column. Fraction I obtained from light petroleum was crystallized from MeOH as flakes (1.86 g), m.p. 65–66°, resistant towards acetylation and addition of  $\text{Br}_2$ ; IR absorption bands characteristic of saturated hydrocarbon. MS showed it to be a mixture of  $\text{C}_{27}$  ( $m/e$  380),  $\text{C}_{28}$  ( $m/e$  394),  $\text{C}_{30}$  ( $m/e$  422),  $\text{C}_{31}$  ( $m/e$  436),  $\text{C}_{32}$  ( $m/e$  450),  $\text{C}_{38}$  ( $m/e$  534, traces) and  $\text{C}_{40}$  ( $m/e$  562) saturated hydrocarbons.

Fraction II obtained from petrol(60–80°)–benzene (4:1) gave crystals (280 mg) from EtOH– $\text{CHCl}_3$ , m.p. 79–80°,  $\nu_{\text{max}}^{\text{KBr}}$  3300  $\text{cm}^{-1}$  (OH); acetate, m.p. 66°. It was identified as ceryl alcohol by direct comparison (m.m.p., TLC) with an authentic sample.

Fraction III obtained from petrol–benzene (1:1) gave crystals (810 mg), m.p. 147–148° (petrol),  $[\alpha]_D^{30} -40.8^\circ$  ( $\text{CHCl}_3$ ); acetate, m.p. 142–143°,  $[\alpha]_D^{30} -47.6^\circ$  ( $\text{CHCl}_3$ ); benzoate, m.p. 152–153°,  $[\alpha]_D^{30} -24^\circ$  ( $\text{CHCl}_3$ ). It was identified as a 1:1 mixture of campesterol and sitosterol by direct comparison (m.m.p., TLC, IR) with authentic samples.

*Acknowledgements*—The authors thank Dr. S. K. Gandhi and Dr. D. S. Bhakuni for spectra and elemental analyses and also the C.S.I.R., New Delhi, for financial assistance.

<sup>15</sup> R. RYHAGE and E. STENHAGEN, *Ark. Kemi* **15**, 291 (1960).

<sup>16</sup> W. N. SUMERWELL, *J. Am. Chem. Soc.* **79**, 3411 (1957).

<sup>17</sup> V. KOBRLI and R. ZAHRADEK, *Chem. Listy* **48**, 1189 (1954).

<sup>18</sup> D. R. GUPTA and S. K. GARG, *Indian J. Appl. Chem.* **29**, 29 (1966).

<sup>19</sup> Y. INOUE, M. NODA and O. HIRAYAMA, *J. Am. Oil Chem. Soc.* **32**, 132 (1955).

<sup>20</sup> H. K. MANGOLD and R. KAMMERER, *Chem. & Ind.* 1032 (1961).

<sup>21</sup> J. H. MITCHELL, H. R. KRAYBILL and F. P. ZSCHEILE, *Ind. Engng. Chem.* **15**, 1 (1943).

<sup>22</sup> T. P. HILDITCH, C. B. PATEL and J. P. RILEY, *Analyst* **76**, 81 (1951).

*Key Word Index*—*Cassia nodosa*; Leguminosae; nodolidate; (–)-7-acetoxy-9,10-dimethyl-1,5-octacosanolide; fatty acids.