3,3-Diallyldienestrol diacetate from IIb. To 15 ml. of diethylaniline, 1.91 g. of 3,4-bis(4-allyloxyphenyl)-3,4-hexanediol (IIb) was added and the solution refluxed under nitrogen for 6 hr. The reaction mixture was poured into 200 ml. of 2*N* hydrochloric acid, stirred, and then extracted with ether. The ether extract was washed with water, dried over sodium sulfate, and the solvent evaporated. The residue, a viscous oil, was used without further purification for dehydration to 3,3′-diallyldienestrol diacetate. The dehydration was carried out by refluxing the oil, *meso*-3,4-bis(3-allyl-4-hydroxyphenyl)-3,4-hexanediol, with a mixture of 10 ml. of acetyl chloride and 10 ml. of acetic anhydride for 4 hr. Four hundred milliliters of water was added and the mixture extracted with ether. The ether extract was washed with a sodium bicarbonate solution, then with water, dried over sodium sulfate, and the ether evaporated. The residue was crystallized from ethanol and then recrystallized from an ether–petroleum ether (b.p. 40–60°) mixture. The 3,3′-diallyldienestrol diacetate melted at 145–147° and its yield was 0.25 g.

Anal. Calcd. for $C_{28}H_{30}O_4$: C, 78.11; H, 7.02. Found: C, 78.09; H, 7.18.

3,3′-Diallyldienestrol diacetate from dienestrol. Dienestrol was treated with allyl bromide in the same manner as described for the preparation of compound IIb. Dienestrol diallyl ether was rearranged in diethylaniline to form 3,3′-diallyldienestrol, which was then crystallized from an ether–petroleum ether (b.p. 40–60°) mixture and recrystallized from dilute ethanol. The 3,3′-diallyldienestrol melted at 123–125°.

Anal. Calcd. for $C_{24}H_{26}O_2$: C, 83.21; H, 7.28. Found: C, 83.12; H, 7.35.

The 3,3′-diallyldienestrol quickly discolored upon exposure to air but remained colorless when the hydroxyl groups were acetylated. Acetylation with acetic anhydride or a pyridine–acetic anhydride mixture gave very poor yields but acetylation of the sodium salt of the 3,3′-diallyldienestrol with acetic anhydride produced the diacetate in good yields. Six grams of 3,3′-diallyldienestrol was dissolved in 100 ml. of 30% aqueous ethanol containing 1.9 g. of sodium hydroxide. The solution was evaporated to dryness under reduced pressure and the residue refluxed for 7 hr. with 150 ml. of acetic anhydride. The acetic anhydride was decomposed with water and the remaining solid was collected on a filter and crystallized from ethanol. A 5.1-g. sample of 3,3′-diallyldienestrol diacetate was obtained. The compound was identical in melting point and infrared spectrum with the product obtained from the *meso*-3,4-bis(4-allyloxyphenyl)-3,4-hexanediol (IIb) through Claisen rearrangement, acetylation, and dehydration.

CHICAGO 9, ILL.

[CONTRIBUTION FROM THE EASTERN REGIONAL RESEARCH LABORATORY AND THE DEPARTMENT OF CHEMISTRY, ST. JOSEPHS COLLEGE[1]]

Steroidal Sapogenins. LXIII. Chiapagenin, a New Normal (25 L) Δ^5-12β-Hydroxysapogenin[2]

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A new normal Δ^5-12β-hydroxysapogenin, for which the name chiapagenin is proposed, has been isolated from the tubers of Dioscorea chiapasensis Matuda.[4] Unequivocal proof of structure was obtained by catalytic hydrogenation of chiapagenin followed by acidic isomerization at C_{25} to give the known 5α,12β-hydroxysapogenin rockogenin. Reduction of the known normal Δ^5-12-ketosapogenin correllogenin with lithium in liquid ammonia containing methyl alcohol gave chiapagenin. Therefore the latter must be 12β-hydroxyyamogenin.

During the course of investigations of the plant kingdom for steroidal sapogenins, a new sapogenin was isolated from *Dioscorea chiapasensis* Matuda[4] and identified. The two sapogenins present in this plant were separated by chromatography. The first product eluted was identified as the Δ^5-normal (25 L) sapogenin, yamogenin, by direct comparison with an authentic sample. Further elution gave a more polar, obviously polyhydroxy, sapogenin. The melting points of the latter compound and its acetate did not correspond to any previously known sapogenin. The structure of the new sapogenin, for which the name chiapagenin is proposed, was established in the following manner. Analysis of the new sapogenin and its acetate showed two hydroxyl groups present and the typical C_{27} skeleton found in steroidal sapogenins. The infrared spectrum of chiapagenin diacetate showed this sapogenin to have the normal spiroketal side chain[5,6] with bands characteristic of Δ^5-unsaturation.[7,8] This latter feature of the molecule

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(2) Previous paper in this series, Steroidal Sapogenins. LXII, M. E. Wall and S. Serota, *Tetrahedron*.

(3) (a) Eastern Utilization Research and Development Division, Philadelphia 18, Pennsylvania. Abstracted from a thesis submitted to the Department of Chemistry, St. Joseph's College, Philadelphia 31, Pennsylvania, in partial fulfillment of the requirements for the degree of Master of Science. (b) Professor of Chemistry, St. Joseph's College, Philadelphia 31, Pennsylvania. (c) Eastern Utilization Research and Development Division, Philadelphia 18, Pennsylvania.

(4) Identified by Dr. E. Matuda, Instituto Biologico, University of Mexico, Villa Obregón, Mexico, D. F.

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(8) C. R. Eddy, M. E. Wall, and M. K. Scott, *Anal. Chem.*, **25**, 266 (1953).

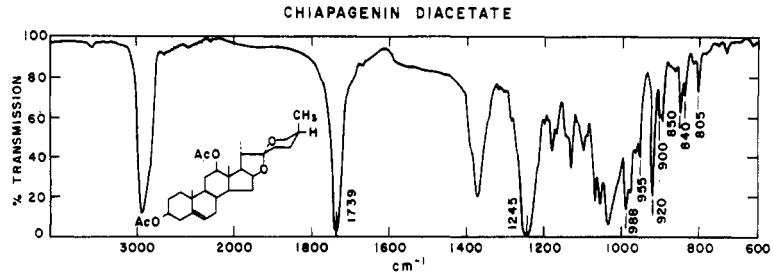

Figure 1

was confirmed by the high negative optical rotation.[9]

The entire structure of chiapagenin was established by two independent paths. Catalytic hydrogenation of chiapagenin gave a normal sapogenin

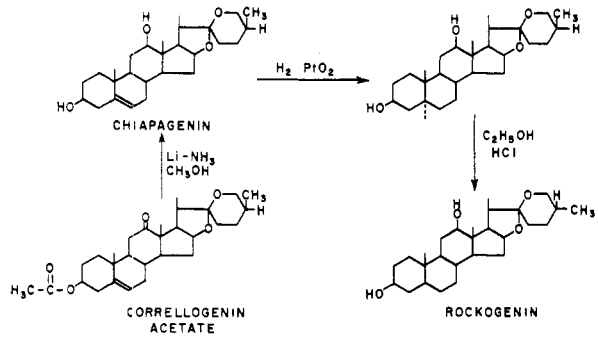

Figure 2

with the 5α-ring A/B fusion. This compound was not further characterized but was subjected to prolonged heating at reflux temperature in ethanolic hydrochloric acid. This is the well known iso reaction which converts normal sapogenins with the axial methyl group to the corresponding isosapogenin (25 D) with the more stable equatorial methyl group.[10] The crude reaction product was chromatographed and the major product, after acetylation, was shown to be rockogenin diacetate,[11,12] identical with an authentic specimen. As the iso reaction inverts only the asymmetric center at C_{25},[10] and as under the experimental conditions used catalytic hydrogenation affects only the Δ^5 moiety,[13] chiapagenin must be 12β-hydroxyyamogenin.

The second independent proof of structure involved the normal Δ^5-12-keto sapogenin correllogenin,[14] which was reduced with lithium metal in liquid ammonia in the presence of methanol.[15] This reaction when applied to hindered ketones gives the most stable hydroxyl[15] which, in the case of the 12-ketone, should be the equatorial 12β-hydroxyl group. In accordance with this prediction, correllogenin acetate reduced in the above manner gave chiapagenin in excellent yield.

Steroidal sapogenins with a 12-hydroxyl group are of rare occurrence in nature. The only examples to date are rockogenin[11] and agavogenin,[11] which occur in a limited group of *Agave* species and are 5α,12β-hydroxysapogenins with the iso side chain. The isolation from *D. chiapasensis* Matuda of chiapagenin represents the first case of a 12-hydroxy sapogenin to be found in this genus. Oxygenation at C_{12} of any type is rare in the *Dioscorea*. In contrast to the *Agave* genus, where many species contain the 12-keto sapogenins hecogenin and/or manogenin,[11] the 12-ketones gentrogenin and correllogenin have to date been found only in *D. spiculiflora*.[14]

EXPERIMENTAL

Melting points were obtained with a Kofler[16] micro melting-point apparatus. Optical rotations were determined in chloroform solution. Infrared spectra were obtained with a Perkin-Elmer Model 21 double beam spectrophotometer with sodium chloride prism and cells.

Isolation of chiapagenin. Tubers of *D. chiapasensis* Matuda were collected[17] at the Finca Monte Bella of Sr. Cristobal above San Sebastian, Guatemala. The sample was received dry and was ground to give 5.2 kg. of dry material. This material was boiled, with stirring, for 1 hr. with two separate 5-gallon portions of 70% isopropyl alcohol, the extract being drawn off each time; 5 gallons of 90% isopropyl alcohol was used for the third extraction. These alcoholic extracts were combined and concentrated by distillation to 8.6 l. which was refluxed 4 hr. with 1.3 l. of concd. hydrochloric acid. The crude precipitate was filtered, washed with 10 l. of 5% sodium bicarbonate solution followed by 6 l. of water, and the solids dried in a forced-air oven. This material was extracted in a soxhlet extractor with 4 l. of heptane for 60 hr. The heptane solution of crude sapogenins was concentrated by distillation to 700 ml. and after standing at room tem-

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(16) Specification of brand names of equipment and material used does not imply endorsement over similar commercial products.

(17) Tubers were collected by Dr. Ernest P. Imle, present address, Director of Research, American Cocoa Research Institute, 1741 K Street, N.W., Washington, D. C.

perature for several days 3.8 g. of crude crystalline material was deposited. This was chromatographed on a Florisil column using benzene as solvent. The benzene eluates gave 1.4 g. of a monohydroxy sapogenin. This material (0.2 g.) was refluxed for 30 min. in 5 ml. of acetic anhydride in pyridine (1:1). The solution was evaporated to dryness *in vacuo* and the residue recrystallized from methyl alcohol to give needles, m.p. 194–197°. These crystals were identified as yamogenin acetate by comparison of its infrared spectra with that of an authentic specimen. Elution with benzene-chloroform (1:1) and chloroform gave 1.8 g. of chiapagenin which, after recrystallization from methyl alcohol, melted at 249–251°, $[\alpha]_D^{24°}$ −126°.

Anal. Calcd. for $C_{27}H_{42}O_4$: C, 75.31; H, 9.83. Found: C, 75.25 H, 10.06.

Acetylation of 0.2 g. of chiapagenin, as in the previously described manner, and recrystallization of the product from methyl alcohol yielded 0.14 g. rectangular needles, m.p. 191–193°, $[\alpha]_D^{24°}$ −127°. The infrared spectrum shows strong bands at 1739 and 1245 cm.$^{-1}$ (acetate carbonyl and —C—O—C— stretching respectively) and bands at 988 and 920 (strong), 900 and 850 (weak) cm.$^{-1}$, all four bands attributed to the normal spiroketal system. Bands at 840 and 805 cm.$^{-1}$ are due to Δ^5 unsaturation.

Anal. Calcd. for $C_{31}H_{46}O_6$: C, 72.34; H, 9.01. Found: C, 72.65; H, 9.22.

Conversion of chiapagenin to rockogenin. A 0.272-g. sample of chiapagenin was dissolved in a solution of 100 ml. of a 5% solution of glacial acetic acid in ethyl alcohol to which was then added 0.272 g. of platinum oxide. The mixture was hydrogenated at 3 atm. for 21 hr. at room temperature. The platinum was filtered and all the solvent removed *in vacuo*. Infrared analysis showed the double bond had been saturated (absence of 805 and 840 cm.$^{-1}$ bands). The white amorphous looking material which resulted was dissolved in 47 ml. of absolute ethyl alcohol. Concentrated hydrochloric acid (7 ml.) was then added and the resulting solution was refluxed 48 hr. Solvent was removed *in vacuo* and the residue chromatographed on a Florisil column using benzene as solvent. Elution with chloroform gave a glass which on crystallization from acetone yielded 60 mg. of crystals. This crystalline fraction was acetylated in the usual manner and gave crystals from methyl alcohol, m.p. 200–203. The infrared spectrum was identical with that of authentic rockogenin diacetate.

Conversion of correllogenin to chiapagenin. A 0.33-g. sample of correllogenin acetate was dissolved in a mixture of 5 ml. of anhydrous ether and 5 ml. of anhydrous tetrahydrofuran in a 100 ml. two-necked round bottomed flask. The solution was added, with continued stirring, to a solution of liquid ammonia (25 ml.) containing 5 ml. of methyl alcohol. Lithium wire, 0.2 g., cut in small pieces, was added rapidly. After 5 min. 2.0 g. of ammonium chloride was added and the evaporation of ammonia was hastened by a warm water bath. Water (50 ml.) was then added and the aqueous slurry extracted twice with 20-ml. portions of methylene chloride. The solvents were evaporated and the residue acetylated, using the procedure previously described, to yield 0.21 g. of short rectangular needles from methyl alcohol, m.p. 191–193°, and an infrared spectrum identical with that of natural chiapagenin diacetate.

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[CONTRIBUTION FROM THE NORTHERN REGIONAL RESEARCH LABORATORY[1]]

Unique Fatty Acids from *Limnanthes douglasii* Seed Oil: The C_{20}- and C_{22}-Monoenes[2]

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The principal fatty acids of *Limnanthes douglasii* seed oil are shown to include two previously unknown components: *cis*-5-eicosenoic (65%) and *cis*-5-docosenoic acid (7%). The oil also contains *cis*-13-docosenoic (erucic) acid (13%) and 10% of an unknown C_{22}-acid.

Limnanthes douglasii or meadow-foam (fam. Limnanthaceae) is an annual herb native to coastal California and presently cultivated as an ornamental.[3] A recent paper from this laboratory[4] indicated that the seed oil of this species is highly unusual in containing 94% of fatty acids longer than C-18. The present paper will report isolation and characterization of three of the four principal fatty acids of *Limnanthes douglasii* seed oil.

Isolation of pure acids. Gas chromatographic analysis of the methyl esters of the mixed acids from *Limnanthes* oil indicated that the principal components were a C_{20}-monoene, a C_{22}-monoene

(1) One of the laboratories of the Northern Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

(2) Presented before the Division of Organic Chemistry, 137th Meeting, American Chemical Society, Cleveland, Ohio, April 5–14, 1960.

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