tinction was poor since the spots ranged from yellow to brown in color and soon became brown. Anisidine phosphate[39] likewise gave tan colors with variations toward yellow and pink, but both the sheets and the spots became dark gray on standing. In contrast to the wide applicability, considerable color distinction and ease of use of the aryl amine sprays, phenolic sprays were found to be less satisfactory.[40]

The unknown sugar solution was chromatographed by the descending method on Whatman No. 1 filter paper with butanol–acetic acid–water as the developing agent and *m*-phenylenediamine as the spray. Two, four, six and ten applications of the solution were made to separate areas 5 mm. in diameter,[41] and the chromatogram was run for 14 hours after the chromatography box had been saturated overnight. The spots, after development of the color, were clear and well-defined, showing that the anion exchange resin had been effective in removing the acid. The results listed in Table IV were obtained, proving that the sugar was glucose.

TABLE IV

CHROMATOGRAPHY OF UNKNOWN SUGAR

Sugar[a]	R_f[b]	Color
Unknown	0.205[c]	Yellow-tan
Glucose	.206	Yellow-tan
Galactose	.189	Yellow-tan
Mannose	.241	Yellow-tan
Fructose	.243	Pale yellow

[a] These sugars were used for comparison since the unknown was known from analysis to be a hexose and since it appeared to yield glucosazone.[3] [b] Average of 4 values. [c] Average of 12 values.

(39) S. Mukherjee and H. C. Srivastava, *ibid.*, **169**, 330 (1952).

(40) They were, however, more selective for certain types of sugars. Those tried included orcinol and trichloroacetic acid, A. Benvenue and K. T. Williams, *Arch. Biochem. Biophys.*, **34**, 225 (1951); resorcinol, V. V. Rachinskii and E. I. Knyazyatova, *Doklady. Akad. Nauk S.S.S.R.*, **85**, 1119 (1952) [*C. A.*, **47**, 448 (1953)] and W. G. Forsyth, *Nature*, **161**, 240 (1948); and naphthoresorcinol, W. G. Forsyth, above, and footnote 7.

(41) Each 5 mm. spot was approximately equivalent to 3–5 μl. of a 1% sugar solution or 30–50 μg. of sugar. Extra applications were made since the strength of the unknown solution was undoubtedly less than 1%.

WELLESLEY 81, MASS.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF WAYNE UNIVERSITY]

Terpenoids. XI.[1] Investigation of Nine Cactus Species. Isolation of Two New Triterpenes, Stellatogenin and Machaeric Acid[2]

BY CARL DJERASSI, L. H. LIU, E. FARKAS, A. E. LIPPMAN, A. J. LEMIN, L. E. GELLER, R. N. MCDONALD AND B. J. TAYLOR

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The chemical examination of nine cacti from Mexico, Costa Rica and Peru is described. In addition to the known triterpenes oleanolic acid (I) and betulinic acid (III), two new triterpenes were encountered. From *L. stellatus* and *M. eruca* there was isolated a new dihydroxy lactone, stellatogenin, which was shown to possess one secondary and one tertiary hydroxyl function by direct correlation with the cactus triterpene thurberogenin. The isolation of a second new triterpene, machaeric acid (21-keto-oleanolic acid) (II), from *M. gummosus* is recorded.

Our initial observation[1,3] of the occurrence of triterpene glycosides in certain giant cacti has prompted us to undertake a more extensive study of this plant family. We should now like to report on nine cactus species which range, as far as their natural habitats are concerned, from Northern Mexico to Peru.

The genus *Lemaireocereus* has so far been the one most extensively studied in our laboratory[1,3] since it appears to be particularly rich in triterpene glycosides and it has now been possible to obtain four additional species.

L. pruinosus[4] occurs rather widely in Central and Southern Mexico[5] where it is known as "Pitayo" and the presently employed specimens were obtained from the gardens of Mr. Howard E. Gates of Corona, California.[6] The plant contained glycosidic material and upon hydrolysis yielded a single triterpene identified as oleanolic acid (I).

L. stellatus[6,7] is quite common in Southern Mexico, principally in the States of Oaxaca and Puebla and is generally referred to as "Xoconochtle" or "Pitayo." This cactus has so far proved to be the richest source of triterpenes, approximately 3.5% of a triterpene mixture being obtained after acid hydrolysis. The acidic fraction was separated only with difficulty after chromatography of the methyl esters, whereupon oleanolic (I) and betulinic (III) acids[8] could be identified. This represents the second isolation of a member of the lupeol group of triterpenes from cacti[1]; as will become apparent from subsequent publications several new triterpenes encountered in our cactus studies also belong to this group.

The neutral fraction was processed by chromatography of the acetates which resulted in the clean separation of two components. The earlier eluted material (*ca.* 15%) was identical with the acetate of thurberogenin, a new triterpene lactone ($C_{30}H_{46}O_3$) isolated recently[3a] from the cactus *Lemaireo-*

(1) Paper X, C. Djerassi and A. E. Lippman, THIS JOURNAL, **76**, 5780 (1954).

(2) We are greatly indebted to the Division of Research Grants (Grant No. G-3863) of the U. S. Public Health Service and to the Rockefeller Foundation for financial support.

(3) (a) C. Djerassi, L. E. Geller and A. J. Lemin, THIS JOURNAL, **75**, 2254 (1953); (b) C. Djerassi, R. N. McDonald and A. J. Lemin, *ibid.*, **75**, 5940 (1953); (c) C. Djerassi, E. Farkas, A. J. Lemin, J. C. Collins and F. Walls, *ibid.*, **76**, 2969 (1954).

(4) N. L. Britton and J. N. Rose, "The Cactaceae," Carnegie Institution of Washington, Washington, D. C., 1920, Vol. II, p. 88.

(5) H. Bravo, "Las Cactaceas de Mexico," Mexico, D. F., 1937, p. 256.

(6) We are greatly indebted to Mr. Howard E. Gates for his cooperation in supplying specimens for our work.

(7) Reference 4, p. 92; ref. 5, p. 261.

(8) In contrast to oleanolic acid (see ref. 3a where all plant sources described up to 1952 are summarized), the occurrence of betulinic acid has been reported in only relatively few plants (*cf.* list of plant sources by A. Stabursvik, *Acta Chem. Scand.*, **7**, 446 (1953)). It is quite probable that the presence of this acid has been overlooked in other plants because of the difficult separation from the isomeric oleanolic acid.

reus thurberi. The bulk of the material (80%) was a homogeneous monoacetate, $C_{32}H_{50}O_5$, which upon saponification yielded the parent triterpene, $C_{30}H_{48}O_4$ now named "stellatogenin." Stellatogenin exhibits a strong infrared absorption band at 5.66 μ similar to that observed[3a] with thurberogenin and typical of five-membered lactones, thus accounting for three of the four oxygen atoms. The remaining oxygen atom was characterized as a tertiary hydroxyl group, since stellatogenin acetate still showed a free hydroxyl band in the infrared, but could not be further acetylated or oxidized. The presence of this tertiary hydroxyl group was proved unequivocally by the smooth dehydration of stellatogenin acetate by means of phosphorus oxychloride in pyridine to thurberogenin acetate.[9] In order to exclude the possibility that this dehydration was accompanied by skeletal rearrangement, a further correlation between stellatogenin and thurberogenin was accomplished in the following manner. Thurberogenin readily forms an oxide with perbenzoic acid and lithium aluminum hydride reduction of this oxide led to a tetrol, which was shown to be identical with the tetrol obtained directly by similar reduction of stellatogenin. This appears to be the first time that a triterpene possessing a tertiary hydroxyl group has been encountered in nature.

In view of the fact that stellatogenin can be dehydrated so readily to thurberogenin, it remained to be established whether thurberogenin is indeed a constituent of *L. stellatus* or formed as an artifact in the isolation procedure, since two steps—acetylation with acetic anhydride or acid hydrolysis of the glycosides—could have resulted in dehydration. Acetylation of pure stellatogenin by the same procedure as was employed for the crude triterpene mixture followed by careful chromatography yielded no trace of thurberogenin. However when stellatogenin was refluxed with methanolic hydrochloric acid under precisely the same conditions used in the hydrolysis of the *L. stellatus* glycosides, a mixture of stellatogenin and thurberogenin was encountered. It appears, therefore, that thurberogenin is not a true constituent of the cactus *L. stellatus*, but rather is formed as an artifact by dehydration of stellatogenin. It should be pointed out, however, that the dehydration with acid occurs only to the extent of about 25% and that thurberogenin isolated[3a] from *L. thurberi* must be a true constituent of that cactus rather than a dehydration product since no stellatogenin was encountered in that plant.

According to Britton and Rose,[10] one *Lemaireocereus* species, *L. aragonii*, is indigenous to Costa Rica although some authors consider this a species of the genus *Cereus*.[11] It is interesting to note that chemically, this cactus was different from the other *Lemaireocereus* species examined by us[1,3] in that only traces of triterpenoid material (apparently a mixture of amyrins) was obtained in both the glycosidic and non-glycosidic fraction. There is also some question whether *Lemaireocereus laetus*,[12] a cactus growing in Southern Ecuador and Central Peru should not be assigned to the genus *Cereus* and our chemical study[13] of this plant indicated the almost total absence of triterpenes or alkaloids; it is premature to state whether this observation is of taxonomic significance.

Machaerocereus is a new genus established by Britton and Rose[14] and only two species, *M. gummosus* and *M. eruca*, both of them indigenous to the Mexican State of Baja California, have been assigned to it. We were interested particularly in studying these two cacti since they are related to the genus *Lemaireocereus* and, in fact, were once referred to it[14] because of the close botanical similarity. A study of the neutral triterpene fraction of *M. gummosus* already has been published earlier and from it there has been isolated a new triterpene "gummosogenin," the structure of which was demonstrated[15] to be Δ^{12}-18β-oleanene-3β,16β-diol-28-al (IV). A sufficient amount of the relatively small acidic fraction now has accumulated so that it has been possible to investigate it also. Chromatography of the crude methyl esters furnished one pure component, $C_{31}H_{48}O_4$, characterized further as the monoacetate and free acid, which we have named "machaeric acid." The fourth oxygen atom is present as a reactive carbonyl group and it has been possible to locate[16] it at position 21 (II).

Machaereocereus eruca[6] ("chirinole" or "creeping devil") proved to have a triterpene composition similar to that mentioned above for *Lemaireocereus stellatus* since both stellatogenin and betulinic acid could be isolated after hydrolysis of the glycosides. Since gummosogenin (IV), the principal triterpene of *M. gummosus*, already has been correlated[15] with longispinogenin (V),[3b] the chief neutral triterpene of *Lemaireocereus longispinus*, the close taxonomical relationship between the two genera *Machaereocereus* and *Lemaireocereus* has now been confirmed in a very striking manner by the chemical study.

In the Experimental section there are recorded briefly the inconclusive or negative results obtained with three Peruvian[13] cacti, the two monotypic genera[17] *Neoraimondia macrostibas*[18] and *Espostoa lanata*, and *Trichocereus peruvianus*, one of three *Trichocereus* species[19] growing in Peru.

(9) As shown in the Experimental section, the dehydration of stellatogenin to thurberogenin represents a better route to this latter triterpene than the isolation from *L. thurberi* (ref. 3a).

(10) Reference 4, p. 92.

(11) We are grateful to Dr. Jorge Leon (Interamerican Institute of Agricultural Sciences, Turrialba, Costa Rica) for this information and for the plant specimens which were collected in the Virilla valley, approximately 10 km. southwest of Alajuela, Costa Rica.

(12) Reference 4, p. 99.

(13) We should like to acknowledge our indebtedness to Dr. Rama Ferreyra (Museo de Historia Natural "Javier Prado," Universidad Nacional Mayor de San Marcos, Lima, Peru) for his continued cooperation in supplying us with Peruvian plant specimens.

(14) Reference 4, p. 114; ref. 5, p. 284.

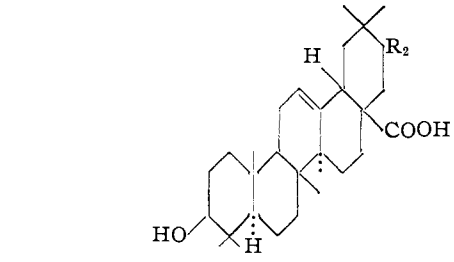
(15) C. Djerassi, L. E. Geller and A. J. Lemin, *Chemistry and Industry*, 161 (1954); THIS JOURNAL, **76**, 4089 (1954).

(16) C. Djerassi and A. R. Lippman, *Chemistry and Industry*, 960 (1954).

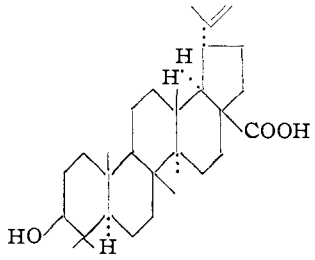
(17) Ref. 4, pp. 61 and 181.

(18) This plant probably contains the longest spines of any cactus and grows quite abundantly along the central highway between Lima and Chosica, Peru.

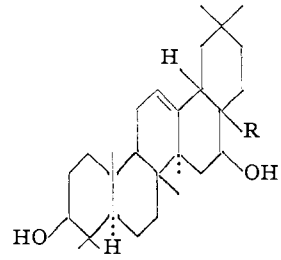
(19) Reference 4, p. 130. The alkaloid composition of several Argentinian *Trichocereus* species has been investigated by L. Reti and others (*cf.* L. Reti in R. H. F. Manske and H. L. Holmes, "The Alkaloids," Academic Press, Inc., New York, 1954, Vol. IV, pp. 23–28).



I, $R_2 = H_2$
II, $R_2 = O$



III



IV, R = CHO
V, R = CH_2OH

Experimental[20]

Isolation of Oleanolic Acid from *Lemaireocereus pruinosus*.—The despined, fresh cactus[6] (3.4 kg.) was dried at 100° furnishing 375 g. of dry material which was extracted exhaustively with ethanol. The ethanolic residue (74 g.) was washed with ether (10 g. oil removed) and then hydrolyzed in the standard manner[3] with 20% methanolic hydrochloric acid. Essentially all of the triterpene fraction was acidic and after crystallization from methanol yielded 7.0 g. of oleanolic acid, m.p. 297–302°, methyl ester, m.p. 196–197°; for further characterization, 1.5 g. of the acid was refluxed with hydrochloric acid–acetic acid furnishing 1.25 g. of 3-acetoxy-18-iso-oleanolic acid lactone, m.p. 350–355°. In each instance, identity was established by mixture melting point determination and infrared comparison.

Isolation of Stellatogenin from *Lemaireocereus stellatus*.—The usual processing of 4.2 kg. of fresh cactus[6] yielded 532 g. of dry material and 119 g. of ethanolic extract. Acid hydrolysis, followed by washing with 10% potassium hydroxide solution (see below for isolation of acids), ether extraction and concentration of the ether to *ca.* 80 cc. furnished 11.8 g. of nearly colorless neutral material, m.p. *ca.* 300°. Purification was best accomplished by acetylation with acetic anhydride–pyridine at room temperature (40 hours) followed by chromatography on 600 g. of neutral alumina. Elution with 1:1 benzene–ether and recrystallization from methanol–chloroform yielded 14% of thurberogenin **acetate**,[3a] m.p. 249–251°, $[\alpha]D$ +22°,[21] identified with authentic material[3a] by infrared analysis.

Anal. Calcd. for $C_{32}H_{48}O_4$: C, 77.37; H, 9.74. Found: C, 77.27; H, 9.66.

Further elution with ether–chloroform (3:2) gave nearly 80% of colorless crystals of **stellatogenin monoacetate**, m.p. 327–330° after recrystallization from chloroform–methanol, $[\alpha]D$ +53°, $\lambda_{max}^{CHCl_3}$ 5.66 (lactone) and 5.82 μ (acetate).

Anal. Calcd. for $C_{32}H_{50}O_5$: C, 74.67; H, 9.79. Found: C, 74.40; H, 9.66.

The acetate was unchanged after treatment with chromium trioxide in sulfuric acid–acetone or acetic acid solution indicating that the fourth oxygen atom is present as a tertiary hydroxyl group.

Saponification of the acetate with 3% methanolic potassium hydroxide (1.5 hours, steam-bath) *followed by acidification* (to permit relactonization) and recrystallization from methanol–chloroform produced in nearly quantitative yield **stellatogenin**, m.p. 317–319°, $[\alpha]D$ +36°, $\lambda_{max}^{CHCl_3}$ 5.66 μ (lactone).

Anal. Calcd. for $C_{30}H_{48}O_4$: C, 76.22; H, 10.24. Found: C, 76.09; H, 9.99.

Acetylation furnished the above monoacetate in 95% yield demonstrating that thurberogenin acetate was not formed by dehydration of stellatogenin during the acetylation process.

Dehydration of Stellatogenin Acetate to Thurberogenin Acetate. (a) **With Phosphorus Oxychloride in Pyridine.**—A solution of 95 mg. of stellatogenin acetate in 11 cc. of pyridine was refluxed for 2 hours with 2.2 cc. of phosphorus oxychloride. Dilution with water, extraction with benzene and chromatography yielded a single homogeneous fraction (85 mg.) identified as thurberogenin acetate by its physical constants, m.p. 247–250°, $[\alpha]D$ +22°, and infrared comparison.

Anal. Calcd. for $C_{32}H_{48}O_4$: C, 77.37; H, 9.74. Found: C, 77.14; H, 9.91.

(b) **Preparative Experiment.**—Since *L. stellatus* is such a rich source of triterpenes, the following method has proved to be superior to the isolation[3a] from *L. thurberi* as far as the accumulation of larger amounts of thurberogenin (required for structure elucidation studies) is concerned.

The crude stellatogenin (35 g.) obtained from 2.1 kg. of dry *L. stellatus* was acetylated with 150 cc. of acetic anhydride and 300 cc. of pyridine overnight at 25°. The crude product, obtained on pouring the acetylation mixture into water, filtering and drying of the solid, was refluxed with 500 cc. of pyridine and 100 cc. of phosphorus oxychloride whereupon 25 g. (1.2% based on dry cactus) of thurberogenin acetate was obtained after chromatographic purification (600 g. of alumina, deactivated with 18 cc. of 10% acetic acid[1] and eluted with benzene).

(c) **With Methanolic Hydrochloric Acid.**—The following experiment demonstrates that the thurberogenin acetate isolated above *directly* from *L. stellatus* most likely arises from partial dehydration of stellatogenin during the glycoside hydrolysis. Stellatogenin (140 mg.) was refluxed for 2 hours with 30 cc. of methanol and 7.5 cc. of concd. hydrochloric acid. Chromatography of the crude product led to 94 mg. of recovered stellatogenin and 40 mg. of thurberogenin.

Thurberogenin Oxide.—Thurberogenin (291 mg.) consumed one equivalent of perbenzoic acid in chloroform solution in 18 hours at room temperature. After washing with carbonate solution and water, the solvent was removed *in vacuo* and the crystalline oxide recrystallized from methanol–chloroform; yield 203 mg. (needles), m.p. 299–302°, $[\alpha]D$ +35°. $\lambda_{max}^{CHCl_3}$ 5.66 μ. The double bond band at 6.03 μ, observable in the infrared spectrum[3a] of thurberogenin, was now absent.

Anal. Calcd. for $C_{30}H_{46}O_4$: C, 76.55; H, 9.85. Found: C, 76.40; H, 9.91.

"**Stellatogenin Tetrol.**" (a) **By Lithium Aluminum Hydride Reduction of Stellatogenin Acetate.**—Stellatogenin acetate (180 mg.) in 15 cc. of tetrahydrofuran was refluxed for 4 hours with a mixture of 1.8 g. of $LiAlH_4$ in 40 cc. of ether. The excess reagent was decomposed with ethyl acetate and then treated with sodium sulfate solution in the manner described[3c] for oleanolic acid lactone. Recrystallization from methanol gave 120 mg. of the tetrol, m.p. 274–277°, $[\alpha]D$ + 22° (pyridine), which showed no infrared absorption in the carbonyl region.

Anal. Calcd. for $C_{30}H_{52}O_4$: C, 75.58; H, 11.00. Found: C, 75.20; H, 11.05.

(b) **By Lithium Aluminum Hydride Reduction of Thurberogenin Oxide.**—Thurberogenin oxide (200 mg.) was reduced exactly as described under (a) furnishing 141 mg. of tetrol, m.p. 273–276°, $[\alpha]D$ +18° (pyridine), undepressed upon admixture with a sample prepared according to method (a).

Isolation of Oleanolic and Betulinic Acids from *L. stellatus*.—The alkaline solution from the glycoside hydrolysis described above was acidified, the precipitate filtered and reprecipitated from alkaline solution yielding 7 g. of crude acids, which were methylated with diazomethane. The

(20) All melting points were determined on the Kofler block. Unless mentioned otherwise, rotations were measured in chloroform solution; infrared spectra were obtained with a Baird recording double beam infrared spectrophotometer. The microanalyses were carried out by Geller Laboratories, Hackensack, N. J.

(21) The earlier reported (ref. 3a) rotation of +45° for thurberogenin acetate involved a mathematical error in which the actual rotation value had been doubled.

crude methyl ester fraction was chromatographed on 280 g. of alumina (deactivated with 5.6 cc. of 10% acetic acid) and eluted with benzene–ether (4:1). The first three crystalline eluates (out of a total of twenty fractions) were combined (2.0 g., m.p. 188–191°) and recrystallized four times from methanol to yield **methyl betulinate**,[22] m.p. 221–223°, $[\alpha]_D$ +5°.

Anal. Calcd. for $C_{31}H_{50}O_3$: C, 79.10; H, 10.71. Found: C, 79.15; H, 10.74.

Saponification of 150 mg. of methyl betulinate was accomplished by refluxing for 2.5 hours with 50 mg. of potassium hydroxide and 12 cc. of diethylene glycol; m.p. 299–303°.

Anal. Calcd. for $C_{30}H_{48}O_3$: C, 78.89; H, 10.59. Found: C, 78.73; H, 10.43.

Methyl betulinate acetate was prepared from the methyl ester in the standard manner, m.p. 201–204°, $[\alpha]_D$ +19°.

Anal. Calcd. for $C_{33}H_{52}O_4$: C, 77.29; H, 10.22. Found: C, 77.54; H, 10.14.

For further characterization, 100 mg. of methyl betulinate was reduced with $LiAlH_4$ leading to 80 mg. of **betulin**, m.p. 251–253°, $[\alpha]_D$ +13°, which was shown to be identical (mixture melting point, infrared comparison) with an authentic specimen isolated in this Laboratory from birch bark.

The later eluates from the original methyl ester chromatogram were combined and rechromatographed to yield eventually 50 mg. of essentially pure **methyl oleanolate**, m.p. 195–197°. The remainder was acetylated and chromatographed whereupon 1.4 g. of methyl **oleanolate acetate**, m.p. 215–217°, could be isolated; comparison with authentic samples[3a] was carried out in the standard manner.

Isolation of Machaeric Acid from *Machaerocereus gummosus*.—Methylation of the acid fraction from the hydrolysis[15] of the glycosides of 984 g. of dry *M. gummosus* yielded 2.8 g. of crude methyl esters which were chromatographed on 75 g. of alumina deactivated with 3 cc. of 10% acetic acid. Combination of the benzene and benzene–ether (2:1) eluates gave 1.88 g. of crystals which furnished 1.23 g. of **methyl machaerate**, m.p. 191–194°, after two recrystallizations from methanol. The analytical sample exhibited m.p. 197–198°, $[\alpha]_D$ +23°.

Anal. Calcd. for $C_{31}H_{48}O_4$: C, 76.80; H, 9.98. Found: C, 76.83; H, 10.08.

Methyl machaerate acetate was recrystallized from methanol–chloroform whereupon it showed m.p. 256–260°, $[\alpha]_D$ +17°.

Anal. Calcd. for $C_{33}H_{50}O_5$: C, 75.24; H, 9.57; acetyl, 8.17. Found: C, 75.17; H, 9.35; acetyl, 7.70.

Machaeric acid (II)[23] was obtained by saponification of 100 mg. of the methyl ester with 5 cc. of 5% methanolic potassium hydroxide (17 hours, steam-bath).[24] The analytical sample was obtained by recrystallization from ether and from acetone–hexane; m.p. 309–312°, $[\alpha]_D$ + 20°.

Anal. Calcd. for $C_{30}H_{46}O_4$: C, 76.55; H, 9.85. Found: C, 76.56; H, 9.94.

Isolation of Stellatogenin and Betulinic Acid from *Machaerocereus eruca*.—The fresh cactus[6] (4.5 kg.) gave 69 g. of ethanolic extract which yielded after hydrolysis 3.8 g. of neutral and 2.1 g. of acidic material. Chromatography of the neutral fraction gave **stellatogenin** (3.1 g., m.p. 317–319°, $[\alpha]_D$ +35°), identified by direct comparison (including infrared comparison) with the triterpene isolated from *L. stellatus*.

Methylation of the acid portion followed by chromatography yielded as the only identifiable fraction slightly impure **methyl betulinate**, m.p. 204–210°, undepressed on admixture with a sample isolated from *L. stellatus*. The infrared spectra were identical.

Anal. Calcd. for $C_{31}H_{50}O_3$: C, 79.10; H, 10.71. Found: C, 79.22; H, 10.98.

Examination of *Lemaireocereus aragonii*.—The fresh cactus (5.4 kg.)[11] was reduced in weight to 471 g. after drying and this material was extracted with ethanol and evaporated to dryness. Extraction of this residue with ether gave no alkaloids, but chromatography of the neutral portion (97% of ether residue) on alumina (activity IV) gave 37 mg. of colorless solid, m.p. 95–105°, yellow color with tetranitromethane, the infrared spectrum of which closely resembles that of α- and β-amyrin. The nearly total absence of glycosides is indicated by the fact that only 13.6 g. of ethanol-soluble, ether-insoluble material was encountered. Acid hydrolysis in the standard manner followed by chromatography of the neutral fraction gave 41 mg. of crystals (m.p. 95–110°) which appeared to be identical (infrared) with the amyrin mixture (?) obtained from the "non-glycosides."

The acid hydrolysis liberated a small amount of oil in the alkaloid fraction. Filtration through alumina and conversion to the picrate followed by a single crystallization from methanol gave a total of 21 mg. of yellow picrate, m.p. 212–216°. Lack of material prevented further purification or characterization.

Anal. Calcd. for $C_{19}H_{19}N_5O_9$: C, 49.46; H, 4.15; N, 15.18; OCH_3, 6.75. Found: C, 49.85; H, 4.05; N, 15.13; OCH_3, 14.01; N-CH_3, 0.

Examination of *Lemaireocereus laetus*.—A total of 36 g. of "non-glycosidic" (ethanol and ether-soluble) and 31 g. of "glycosidic" (ethanol-soluble, ether-insoluble) material was obtained from 5.03 kg. (580 g. dry) of cactus.[13] No alkaloids were detected in either fraction. The "non-glycosidic" portion was essentially all neutral but no crystals could be isolated after chromatography. Acid hydrolysis of the "glycosides" yielded 2.8 g. of neutral oil which also failed to yield crystalline material after chromatography.

Examination of *Espostoa lanata*.—The partially sun-dried cactus[13] (3.7 kg.) was reduced in weight to 254 g. after drying and yielded only 21 g. of ethanol-extractable material (negative Mayer test).

Examination of *Neoraimondia macrostibas*.—The fresh plant (4.95 kg.)[13] gave 700 g. of dry material and 232 g. of ethanolic extract. Ether extraction yielded 119 g. (all neutral, no alkaloids) of oily material, which furnished only oily or amorphous fractions, in spite of extensive chromatographic separation on alumina and silica gel columns. The remaining 106 g. (ethanol-soluble, ether-insoluble) was hydrolyzed in the usual manner giving 0.8 g. of basic (partially crystalline, but insufficient for further characterization since chromatography gave at least six separate fractions) and 73.5 g. of neutral products. This neutral, gummy material behaved like the above "non-glycosidic" portion and yielded no crystals after chromatography.

Examination of *Trichocereus Peruvianus*.—Particular attention was paid in the case of this cactus[13] (269 g. dry weight, 52 g. ethanolic extract) to any alkaloids, since Argentinian *Trichocereus* species[19] are relatively rich in alkaloids. The ether-soluble portion was neutral (1.8 g.) and furnished 0.6 g. of a waxy solid (m.p. 70–74°) after chromatography; this was not investigated further.

The material after acid hydrolysis of the "glycoside" fraction gave a positive test with Mayer reagent but no alkaloids (or quaternary bases) could be isolated. Chromatography of the neutral portion yielded traces of a triterpene lactone, m.p. 320° (dec.) (after crystallization from acetone and high vacuum sublimation), λ_{max}^{nujol} 5.68 μ, which could not be investigated further for lack of material.

Anal. Calcd. for $C_{30}H_{46}O_4$: C, 76.55; H, 9.85. Found: C, 76.66; H, 9.48.

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(22) The physical constants reported by various authors for betulinic acid and its derivatives are summarized by D. H. R. Barton and E. R. H. Jones, *J. Chem. Soc.*, 659 (1944).

(23) The empirical formula $C_{30}H_{48}O_4$ given in ref. 16 should read $C_{30}H_{46}O_4$.

(24) The activating effect of the 21-keto group is quite striking since methyl oleanolate is recovered unchanged under such conditions.