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Mexicanin E. A Norsesquiterpenoid Lactone¹

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Analytical and degradative evidence has been obtained which indicates that mexicanin E is an azulogenic lactone that represents the first member of a new type of sesquiterpenoid lactones with only fourteen carbon atoms.

Recently, we have described the isolation of an interesting new lactone² from *Helenium mexicanum* collected in the neighborhood of Tepexpan and San Cristóbal Ecatepec (Estado de México) which was named mexicanin E. It is interesting to note that *Helenium mexicanum* collected near Nochistlán³ (Estado de Oaxaca) does not contain mexicanin E; only helenalin could be isolated.

Mexicanin E (C₁₄H₁₆O₃), as already reported,² has m.p. 100.5–101.5°, [α] –47°, and gave analysis for C₁₄H₁₆O₃; the infrared and ultraviolet spectra show that the three oxygen atoms as in ambrosin⁴ are distributed as a trisubstituted α,β -unsaturated cyclopentenone and an α,β -unsaturated lactone. We shall now record the salient experiments which led to the elucidation of the gross structural features of this substance and which indicate that mexicanin E is the first member of a new class of C₁₄-azulogenic lactones.^{2,5} The molecular weight determined by mass spectrography through the courtesy of Dr. Biemann⁶ affords a value of 232 which is in full accord with the C₁₄H₁₆O₃ formulation.

Mexicanin E forms a red dinitrophenylhydrazone, whose ultraviolet maximum at 386 m μ corresponds to that of an α,β -unsaturated dinitrophenylhydrazone.⁷

Mexicanin E adds two bromine atoms to the double bond conjugated with the ketone as in the case of helenalin.⁸ The dibromo derivative thus obtained shows only an ultraviolet maximum at 210 m μ (ϵ 12000) corresponding to an exocyclic double bond conjugated with the γ -lactone. The presence of an exocyclic methylene group was confirmed when mexicanin E gave rise to formaldehyde on ozonolysis. The dibromo derivative in boiling ethanol lost hydrobromic acid, forming a monobromolactone. Its ultraviolet spectrum (λ_{\max} 244 m μ , ϵ 8100) showed a bathochromic shift due to the bromine atom attached to the double bond conjugated with the ketone, analogous to those ex-

hibited by bromohelenalin and bromoambrosin.^{4,9}

Partial hydrogenation of mexicanin E with palladium-on-charcoal catalyst until one equivalent of hydrogen was absorbed afforded dihydromexicanin E (C₁₄H₁₈O₃), whose infrared spectrum showed bands at 1760 cm.⁻¹ (γ -lactone), at 1700 and 1690 cm.⁻¹ (cyclopentenone), the ultraviolet spectrum (λ_{\max} 224, 316–318 m μ ; ϵ 9550, 52) confirms the presence of a trisubstituted α,β -unsaturated cyclopentenone chromophore like that present in tenulin and dihydrohelenalin.^{10–12} Dihydromexicanin E forms an orange-red dinitrophenylhydrazone. On further hydrogenation dihydromexicanin E consumes one equivalent of hydrogen yielding tetrahydromexicanin E (C₁₄H₂₀O₃); the same derivative is obtained by hydrogenation of mexicanin E until two equivalents of hydrogen are absorbed. The tetrahydro-lactone does not show in the ultraviolet spectrum the maximum of the α,β -unsaturated ketone chromophore. The infrared spectrum had bands at 1755 cm.⁻¹ (γ -lactone) and at 1728 cm.⁻¹ (cyclopentanone). Tetrahydromexicanin E readily forms an orange dinitrophenylhydrazone and a semicarbazone. Raney nickel desulfurization of its oily cycloethylenemercaptol yielded the nicely crystalline desoxotetrahydromexicanin E, which only contains the lactone as functional group. Its infrared spectrum showed the band of a five-membered lactone at 1760 cm.⁻¹.

The above experiments indicate a close relationship of the chemistry of mexicanin E with that of helenalin⁵ and ambrosin⁴; furthermore the rotatory dispersion curve of mexicanin E¹³ shows a negative Cotton effect similar to that found in helenalin and dihydrohelenalin,¹⁴ whereas the curve of tetrahydromexicanin E shows a positive Cotton effect of the same type of tetrahydrohelenalin. This fact indicates that mexicanin E and helenalin and also tetrahydromexicanin E and tetrahydrohelenalin have respectively the same functional groups and identical stereochemistry in the cyclopentane moiety.

We have confirmed that mexicanin E is an azulogenic lactone by dehydrogenating a suitable derivative in the customary manner.¹⁵

(1) Taken in part from a thesis to be submitted by A. Romo de Vivar to the Universidad Nacional Autónoma de México in partial fulfillment of the requirements to obtain the degree of Doctor en Ciencias Químicas.

(2) A. Romo de Vivar and J. Romo, *Rev. Ciencia*, in press

(3) We are grateful to Dr. Faustino Miranda of the Instituto de Biología de la Universidad Nacional Autónoma de México for the identification of the plants.

(4) H. Abu-Shady and T. O. Soine, *J. Amer. Pharm. Assn.*, **42**, 387 (1953); L. Bernardi and G. Büchi, *Experientia*, **13**, 466 (1957).

(5) The elemental analysis of mexicanin F (see ref. 2) isolated in small amounts from the same plant corresponds also to a C₁₄H₁₆O₃ formula.

(6) We are indebted to Dr. K. Biemann of the Massachusetts Institute of Technology for the mass spectroscopy analyses and molecular weights reported in this paper (*vide infra*).

(7) E. A. Braude and E. R. H. Jones, *J. Chem. Soc.*, 498 (1945); J. D. Roberts and C. Green, *J. Am. Chem. Soc.*, **68**, 214 (1946); C. Djerassi and E. Ryan, *ibid.*, **71**, 1000 (1949).

(8) R. Adams and W. Herz, *ibid.*, **71**, 2546 (1949).

(9) See ref. 8 and references cited therein; see also A. L. Nussbaum, O. Mancera, R. Daniels, G. Rosenkranz and C. Djerassi, *ibid.*, **73**, 3263 (1951).

(10) D. H. R. Barton and P. de Mayo, *J. Chem. Soc.*, 142 (1956).

(11) B. H. Braun, W. Herz and K. Rabiindran, *J. Am. Chem. Soc.*, **78**, 4423 (1956).

(12) G. Büchi and D. Rosenthal, *ibid.*, **78**, 3860 (1956).

(13) We are indebted to Dr. Carl Djerassi and to Syntex, S. A., for the determination of the rotatory dispersions.

(14) C. Djerassi, J. Osiecki and W. Herz, *J. Org. Chem.*, **22**, 1361 (1957).

(15) J. Simonsen and D. H. R. Barton, "The Terpenes." Vol. III, Cambridge University Press, 1952, and A. J. Haagen-Smith in L.

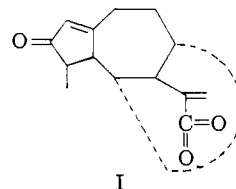
When tetrahydromexicanin E was reduced with sodium borohydride and then heated with palladium-on-charcoal a small amount of a blue oil was obtained. It showed an ultraviolet spectrum typical of azulenes. The mass spectrogram of the blue oil indicated that it is a mixture of about 2/3 of a compound with molecular weight 170 which corresponds to a $C_{13}H_{14}$ azulene, which may be isopropyl or a methylethylazulene but definitely not a trimethylazulene. The other component (about one third) had mol. wt. 184 and is obviously a $C_{14}H_{16}$ azulene. The mixture also contains a trace of a compound with mol. wt. 198 in which oxygen is present and corresponds to a $C_{14}H_{14}O$ azulene. From the oily mixture a crystalline trinitrobenzene adduct could be prepared. Its elemental analysis corresponds to a $C_{14}H_{16}$ azulene moiety.

Therefore we have the location of thirteen carbon atoms, *i.e.*, ten carbons distributed in the perhydroazulene nucleus, three involved in the five-membered lactone and the exocyclic methylene group conjugated with the carbonyl of the lactone. If mexicanin E is biogenetically related to other azulogenic lactones as it appears to be, then this lactone might have a methyl group at C_4 or at C_{10} . Indeed, C-methyl determination indicates that mexicanin E possesses only one methyl group, whereas tetrahydromexicanin E afforded two methyl groups (the second methyl group in the tetrahydro derivative is formed by the hydrogenation of the exocyclic methylene group conjugated with the lactone) and we could find evidence that this methyl group is located at C_4 in the following way: When the ultraviolet spectrum of dihydromexicanin E is carried out in alkaline medium a bathochromic shift of the original maximum (*vide supra*) is produced. The new maximum appears at 236 $m\mu$, and corresponds to a tetrasubstituted double bond conjugated with the ketone. We can rationalize this shift as involving a migration of the C_1 - C_2 double bond to a C_4 - C_5 conjugation with the ketone and a methyl group at C_4 must be present in order to explain the bathochromic shift produced. A similar shift is observed in the alkaline treatment of isotenulin which gives rise to desacetylneotenulin.¹⁰ Furthermore, when dihydromexicanin E was treated with methanolic potassium hydroxide an oily lactone (λ_{max} 236 $m\mu$, ϵ 11200) was obtained which showed infrared bands at 1760 cm^{-1} (γ -lactone), at 1690 and 1635 cm^{-1} (cyclopentenone). The strong band at 1635 cm^{-1} is of the same type as that present in geigerin¹⁶ and desacetylneotenulin¹⁰ which have a conjugated double bond at C_4 - C_5 . From the oily lactone a red dinitrophenylhydrazone (λ_{max} 386 $m\mu$) was prepared.

Alkaline treatment of tetrahydromexicanin E yielded also an oily lactone whose crystalline dinitrophenylhydrazone and semicarbazone proved to be isomeric with those obtained from tetrahydromexicanin E. We attribute this change to the epimerization of the asymmetric center vicinal to the lactonic carbonyl group. Herz and Mitra¹⁷

have reported a similar reaction in the helenalin series which very probably resulted in the epimerization of the C_{11} asymmetric center.

From the analytical and degradative evidence outlined above we tentatively assign to mexicanin E the partial structure I. The isopropyl chain which constitutes the lactone presumably is attached at C_7 on biogenetical grounds. Further work aimed to obtain evidence about the position and orientation of the lactone is now in progress.



Experimental¹⁸

Mexicanin E was obtained by chromatographic fractionation of the chloroformic extract of the plant. As adsorbent, in order to avoid undesirable rearrangements of the lactones,² alumina washed with ethyl acetate and dried at 90° *in vacuo* was used.

Mexicanin E showed m.p. 100.5–101.5° (prisms from ether-hexane), $[\alpha]_D -47^\circ$, gives positive Legal and Tollens reactions; λ_{max} 218, 318 $m\mu$; ϵ 18200, 77; ν_{max} 1760, 1700 and 1588 cm^{-1} .

Anal. Calcd. for $C_{14}H_{16}O_2$: C, 72.39; H, 6.94; O, 20.67; C-methyl, 6.45. Found: C, 72.28; H, 7.10; O, 21.02; C-methyl, 6.07.

Mexicanin E dinitrophenylhydrazone had m.p. 219–221° (deep-red prisms from chloroform-methanol), $\lambda_{max}^{CHCl_3}$ 386 $m\mu$, ϵ 23300.

Anal. Calcd. for $C_{20}H_{20}O_6N_4$: C, 58.25; H, 4.89; O, 23.28; N, 13.58. Found: C, 58.11; H, 4.76; O, 23.02; N, 13.86.

Ozonolysis of Mexicanin E.—A solution of the lactone (0.2 g.) in tetrahydrofuran was ozonized for 30 min. at 0° (a large excess of ozone was passed through the solution), water (5 ml.) was added and most of the solution was distilled into an aqueous solution of dimedone (0.14 g.); upon concentration of this solution to a small volume, there crystallized 25 mg. of dimedone-formaldehyde, m.p. 183–187°. One crystallization from aqueous methanol raised the m.p. to 187–189°, which showed no depression in m.p. on admixture with an authentic specimen.

Mexicanin E Dibromide.—A solution of the lactone (0.5 g.) in acetic acid (10 ml.) was treated with a 10% solution of bromine in acetic acid until a light yellow color persisted. The dibromo derivative crystallized upon dilution with water (0.75 g.); m.p. 130–131°. The analytical sample showed m.p. 131–132° (needles from acetone-hexane), $[\alpha]_D -30^\circ$, λ_{max} 210 $m\mu$, ϵ 12000, ν_{max} 1755 and 1660 cm^{-1} .

Anal. Calcd. for $C_{14}H_{16}O_2Br_2$: C, 42.88; H, 4.11; O, 12.24; Br, 40.76. Found: C, 43.20; H, 4.11; O, 12.28; Br, 40.28.

Bromomexicanin E.—The above dibromo derivative (0.5 g.) in ethanol (50 ml.) was refluxed 5 hr. The solution was evaporated to a small volume *in vacuo*, diluted with water and extracted with chloroform; evaporation of the solvent left an oily residue which crystallized from ether yielding prisms (140 mg.), m.p. 146–149°; further crystallizations from acetone-hexane raised the m.p. to 153–154°, $[\alpha]_D -33^\circ$, λ_{max} 244 $m\mu$, ϵ 8100; ν_{max} 1760, 1715, 1660, 1588 cm^{-1} .

(17) W. Herz and R. B. Mitra, *J. Am. Chem. Soc.*, **80**, 4876 (1958).

(18) Melting points are corrected in the Kofler block; rotations were determined at 22° in chloroform. The ultraviolet absorption spectra were determined in 95% ethanol solution in a Beckman DK 2 spectrophotometer, unless noted otherwise. The infrared spectra were run in chloroform solution on a Perkin-Elmer double beam spectrophotometer (unless stated otherwise). The microanalyses were performed by Dr. Franz Pascher, Bonn, Germany, and the C-methyl group analyses by Dr. J. M. L. Cameron, Department of Chemistry, Glasgow University, England.

Zechmeister, "Progress in the Chemistry of Organic Natural Products," Vol. XII, Springer Verlag, Vienna, 1955, pp. 1–43.

(16) G. W. Perold, *J. S. African Chem. and Ind.*, **8**, 12 (1955); D. H. R. Barton and J. E. D. Levisalles, *J. Chem. Soc.*, 4518 (1958).

Anal. Calcd. for $C_{14}H_{18}O_3Br$: C, 54.03; H, 4.85; O, 15.42; Br, 25.68. Found: C, 53.25; H, 5.27; O, 15.19; Br, 25.96.

Dihydromexicanin E.—A solution of mexicanin E (2 g.) in ethyl acetate (50 ml.) was hydrogenated with Adams catalyst (0.1 g.) until one equivalent of hydrogen was absorbed. The catalyst was filtered off and the solvent evaporated to dryness. The oily residue crystallized from ether, yielding prisms (0.595 g.), m.p. 105–108°; further crystallizations from acetone-hexane raised the m.p. to 130–131°, $[\alpha]_D^{25} -175^\circ$, λ_{max} 224, 316–318 m μ ; ϵ 9550, 52 [in alkaline medium (addition of 5 drops of 10% sodium hydroxide)], γ_{max} 236 m μ , ϵ 10800; ν_{max} 1760, 1700 and 1590 cm^{-1} .

Anal. Calcd. for $C_{14}H_{18}O_3$: C, 71.77; H, 7.74; O, 20.49. Found: C, 71.48; H, 7.53; O, 20.80.

Dihydromexicanin E dinitrophenylhydrazone showed m.p. 236–237° (orange-red needles from chloroform-methanol), $\lambda_{max}^{CHCl_3}$ 384 m μ , ϵ 22000.

Anal. Calcd. for $C_{20}H_{22}O_6N_4$: C, 57.96; H, 5.35; O, 23.17; N, 12.52. Found: C, 58.26; H, 5.58; O, 22.69; N, 13.19.

Alkaline Treatment of Dihydromexicanin E.—To a solution of dihydromexicanin E (0.4 g.) in methanol (20 ml.) a solution of 0.4 g. of sodium hydroxide in 3 ml. of water was added. The mixture was refluxed for 1 hr. It was acidified with acetic acid and concentrated *in vacuo* to a small volume, diluted then with water and extracted with chloroform. The chloroformic extract was evaporated to dryness, the oily residue (0.36 g.) showed λ_{max} 236 m μ , ϵ 11200; ν_{max} 1760, 1690 and 1635 cm^{-1} . The oil could not be crystallized even after chromatographic fractionation. The crystalline dinitrophenylhydrazones obtained from the oily lactone showed m.p. 210–213° (deep-red prisms from chloroform-methanol), $\lambda_{max}^{CHCl_3}$ 386 m μ , ϵ 21758.

Anal. Calcd. for $C_{20}H_{22}O_6N_4$: C, 57.96; H, 5.35; O, 23.17; N, 13.52. Found: C, 58.19; H, 5.90; O, 23.14; N, 13.58.

Tetrahydromexicanin E.—A similar hydrogenation of mexicanin E (2 g.) was carried out as reported above until 2 equivalents of hydrogen was absorbed. The catalyst was filtered and the solvent evaporated to dryness, the oily residue was crystallized from ether affording prisms (1.07 g.), m.p. 98–100°. Chromatography of the mother liquors on alumina yielded 420 mg. more, m.p. 97–99°. The analytical sample showed m.p. 101–102°, $[\alpha]_D^{25} +67^\circ$; ν_{max} 1755, 1728 cm^{-1} .

Anal. Calcd. for $C_{14}H_{20}O_3$: C, 71.17; H, 8.53; O, 20.31; 2C-methyl, 12.69. Found: C, 70.85; H, 8.52; O, 20.50; C-methyl, 11.46.

Hydrogenation of Dihydromexicanin E.—Under the same conditions used above, dihydromexicanin E (1 g.) was hydrogenated, and the tetrahydro derivative was obtained as prisms (0.78 g.), m.p. 100–102°, showing no depression in m.p. on admixture with the above specimen; the infrared spectra were identical.

Tetrahydromexicanin E dinitrophenylhydrazone was obtained as orange needles from methanol; m.p. 233–234°, $\lambda_{max}^{CHCl_3}$ 362 m μ , ϵ 18000. (The position of this maximum corresponds to a dinitrophenylhydrazone of a saturated ketone; see ref. 6.)

Anal. Calcd. for $C_{20}H_{24}O_6N_4$: C, 57.68; H, 5.81; O, 23.05; N, 13.46. Found: C, 57.41; H, 5.83; O, 22.82; N, 13.15.

Tetrahydromexicanin E semicarbazone showed m.p. 231–233° dec. (prisms from methanol).

Anal. Calcd. for $C_{15}H_{23}O_3N_3$: C, 61.41; H, 7.9; O, 16.36; N, 14.33. Found: C, 61.29; H, 7.85; O, 16.41; N, 14.15.

Desoxotetrahydromexicanin E.—The tetrahydro lactone (1.5 g.) was dissolved in 12 ml. of acetic acid, ethane dithiol (1.5 ml.) and 70% perchloric acid (1 ml.) were added, the

mixture was left at room temperature for 3 hr. The solution was diluted with chloroform, washed with water, 5% sodium hydroxide solution and water again, and the extract was then evaporated to dryness. The oily residue (1.55 g.) was dissolved in ethanol (100 ml.), Raney nickel (15 g.) was added and the mixture refluxed with mechanical stirring for 17 hr. The nickel was filtered off and the solution evaporated to dryness. Crystallization of the oily residue from hexane afforded prismatic needles (0.95 g.), m.p. 65–68°. Further crystallizations from hexane raised the m.p. to 73°, $[\alpha]_D^{25} +51^\circ$, ν_{max} 1760 cm^{-1} .

Anal. Calcd. for $C_{14}H_{22}O_2$: C, 75.63; H, 9.97; O, 14.39. Found: C, 75.42; H, 9.93; O, 15.00.

Alkaline Treatment of Tetrahydromexicanin E.—A solution of tetrahydromexicanin E (0.4 g.) in methanol (30 ml.) was mixed with a solution of sodium hydroxide (0.4 g.) in water (3 ml.) and refluxed for 1 hr. It then was worked up as in the alkaline treatment of the dihydro lactone. The oily residue (0.32 g.) showed max. at 1755 cm^{-1} . The dinitrophenylhydrazone prepared from the oily lactone showed m.p. 199–200° (yellow needles from chloroform-methanol), $\lambda_{max}^{CHCl_3}$ 362 m μ , ϵ 16770.

Anal. Calcd. for $C_{20}H_{24}O_6N_4$: C, 57.68; H, 5.81; O, 23.05; N, 13.46. Found: C, 57.52; H, 5.95; O, 23.31; N, 13.11. The semicarbazone showed m.p. 227–228° (prisms from methanol).

Anal. Calcd. for $C_{15}H_{23}O_3N_3$: C, 61.41; H, 7.90; O, 16.36; N, 14.33. Found: C, 61.14; H, 7.89; O, 16.73; N, 14.12.

Aromatization of Tetrahydromexicanin E.—The tetrahydro lactone (3 g.) was dissolved in tetrahydrofuran (40 ml.); sodium borohydride (3 g.) and water (0.5 ml.) were added. The mixture was refluxed for 5 hr., concentrated then to a small volume, diluted with water and acidified with acetic acid. The oily precipitate was extracted with chloroform, the extract washed with water and evaporated to dryness; the gummy residue (2.73 g.) showed in the infrared spectrum only the maximum of the γ -lactone at 1750 cm^{-1} . The residue was dehydrogenated in 200-mg. portions by 10% palladized charcoal (200 mg.) under nitrogen at 320–325° for 20 min. The combined dehydrogenated product was dissolved in hexane and passed through deactivated alumina (20 g.); the blue fraction was extracted with 88% phosphoric acid, diluted then with cold water and extracted with hexane. The extract was washed with water and evaporated to dryness. The blue oil (35 mg.) was dissolved in ethanol and trinitrobenzene (50 mg.) was added. The solution crystallized upon concentration to a small volume furnishing brownish-black needles with metallic luster, m.p. 97–99°. Several crystallizations from methanol raised the m.p. to 107–109°.

Anal. Calcd. for $C_{20}H_{18}O_6N_3$: C, 60.45; H, 4.82; O, 24.16; N, 10.58. Found: C, 60.05; H, 5.19; O, 23.74; N, 10.44.

Isolation of Helenalin.—The *Helenium mexicanum* was collected in the neighborhood of Nochistlán (Oaxaca) in May when the plant was in blossom; it was dried and ground without the roots in a mill. The chloroformic extract of the plant (1 kg.) was processed following the method of Herz, *et al.*¹⁹ The residue (38 g.) after treatment with lead acetate, crystallized from benzene, affording helenalin (7.3 g.), m.p. 167–169°, which showed no depression on admixture with an authentic specimen; the infrared spectra were identical.

The mother liquors were chromatographed on alumina (washed with ethyl acetate). The crystalline fractions obtained with benzene were combined and recrystallized from benzene to give 9.2 g. of helenalin, m.p. 168–170°. No other lactone could be isolated from this plant.

(19) W. Herz, R. B. Mitra, K. Rabindran and W. A. Rohde, *J. Am. Chem. Soc.*, **81**, 1481 (1959).