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SESQUITERPENE LACTONES FROM *DICORIA CANESCENS*

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Key Word Index—*Dicoria canescens*, Asteraceae, xanthanolides, ivalbatin, dicorin, dihydrodicorin

Abstract—*Dicoria canescens* from Baja California, Mexico afforded the known monoterpene glucoside, (−)-*cis*-chrysanthenol *O*-β-D-glucopyranoside, ivalbatin, a xanthanolide and two new sesquiterpene lactones, 4-dihydroivalbatin (dicorin) and 11β,13-tetrahydrovalbatin (dihydrocorin)

INTRODUCTION

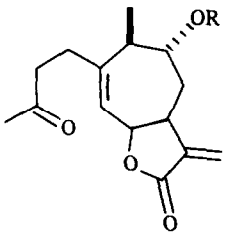
In a continuation of our phytochemical investigations of desert plants exhibiting biological activity, we have identified ivalbatin (**1a**), a xanthanolide previously identified from *Iva dealbata* [1], and two new xanthanolide derivatives, dicorin (**2a**) and dihydrodicorin (**3a**) in an ether extract of the aerial parts of *Dicoria canescens* A Gray from Baja California, Mexico and Southern California Also present in significant quantities was the previously isolated monoterpene glucoside, (−)-*cis*-chrysanthenol *O*-β-D-glucopyranoside [2]

RESULTS AND DISCUSSION

Ivalbatin (**1a**) was obtained from the ether extract and readily polymerized on standing Its ^{1}H NMR spectrum was identical in all respects to those previously published for ivalbatin [1] Acetylation of ivalbatin (**1a**) yielded ivalbatin monoacetate (**1b**) which on comparison with a known sample of ivalbatin monoacetate was found to be identical

Dicorin (**2a**), the major xanthanolide in the ether extract, also polymerized on standing Its molecular formula, $C_{15}H_{22}O_4$, was determined by mass spectrometry, *m/z* 267 $[M+1]^+$, major fragment ions observed at *m/z* 249 $[267-H_2O]$ and 231 $[249-H_2O]$ Its IR spectrum contained one carbonyl absorption band at 1770 cm^{-1}, and a medium strong band at 1670 cm^{-1} for a C=C conjugated to a carbonyl function Its ^{1}H NMR spectrum exhibited two doublets at δ5 57 and 6 26 (*J* = 3 Hz), characteristic for exocyclic methylene protons of the α- methylene-γ-lactone grouping common for sesquiterpene lactones Signals for two methyl groups at δ1 18 (*d*, *J* = 7 Hz) and 1 26 (*d*, *J* = 7 Hz) were assigned to the C-10 and C-4 methyls, respectively A broad signal at δ2 92 integrating for two protons, which disappeared on the addition of deuterium oxide, was assigned to two aliphatic hydroxyl groups Acetylation of **2a** yielded a crystalline diacetate (**2b**) which exhibited an $[M+1]^+$ peak at *m/z* 351 and major fragment ions at 291 $[M-59]^+$, 273 and 231 The ^{1}H NMR spectrum of dicorin diacetate (**2b**) exhibited three proton signals at δ2 03 and 2 10, respectively, characteristic of the two acetyl groups Signals at δ4 84 (1H, *dt*, *J* = 2 5 Hz) and 4 95 (1H, *d(br)*, *J* = 6 Hz) were assigned to H-9 and H-4 signals that had been shifted downfield following acetylation Signals at δ1 15 (3H, *d*, *J* = 7 Hz) and 1 30 (3H, *d*, *J* = 7 Hz) were assigned to the secondary methyls at C-10 and C-4 A broad signal at δ5 45 was assigned to a H-5 trisubstituted olefinic proton Signals at δ5 62 (1H, *d*, *J* = 3 Hz) and 6 38 (1H, *d*, *J* = 3 Hz) were characteristic of the H-13 methylene protons Signals at δ5 56 (1H, *dd*, *J* = 2 5 Hz), 3 35 (1H, *m*) and 2 56 (1H, *sx*, *J* = 7 Hz) were assigned to H-6, H-7 and H-10, respectively Double resonance decoupling experiments established a *cis* configuration for protons at C-5, C-6 and C-7 Irradiation at δ5 45 (H-5) affected H-6 at δ5 56 and irradiation at H-6 resulted in a sharp singlet for H-5 and the H-7 at δ3 35 was changed to a simpler signal Conversely, decoupling at δ5 45 (H-5) also resulted in a less complex signal The decoupling experiments established a *cis*-configuration at the C-6 and C-7 positions That H-9 and H-10 had a *trans*-configuration similar to that of acetylivalbatin (**1b**) was determined by double resonance experiments Irradiation at δ4 84 (H-9) affected the signal at δ2 56 (H-10) which was changed to a simpler signal Decoupling of the H-10 signal resulted in two singlets at δ4 84 (H-9) and 1 15 (C-10-Me) Acetylation of dicorin (**2a**) followed by reduction with sodium borohydride yielded dihydrodicorin diacetate (**3b**) and isodihydrodicorin diacetate (**3c**), compounds previously prepared from ivalbatin (**1a**) and found to be identical (see structures) The structure of dihydrocorin (**3a**), a naturally occurring xanthanolide that is also unstable, was determined by correlation with ivalbatin (**1a**) Acetylation of **3a** yielded **3b** which was previously derived from acetylation of ivalbatin (**1a**) and dicorin (**2a**)

Previously, it had been reported that the genus *Dicoria* did not elaborate sesquiterpene lactones [2] and therefore its chemotaxonomic relationship with other genera of the Heliantheae was not clear Our studies of *D canescens* from Baja California have established the presence of xanthanolides similar to those present in taxa of *Iva*, *Parthenice* and *Parthenium*, which are members of the subtribe Ambrosiinae [3] Furthermore, preliminary studies on the biological activity of xanthanolides suggest that ivalbatin acetate (**1b**) and derivatives of dihydrodicorin are cytotoxic and phototoxic to human erythrocytes at concentrations at 100 μg/ml Results on cytotoxic–activity relationships of xanthanolides will be published elsewhere



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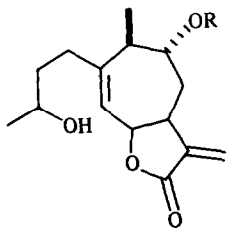

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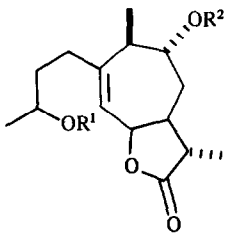
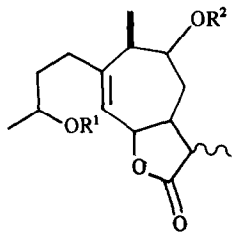


3a R^1 = H, R^2 = H
3b R^1 = Ac, R^2 = Ac



3c R^1 = Ac, R^2 = Ac
4 R^1 = H, R^2 = Ac

EXPERIMENTAL

Mps uncorr

Preparation of plant extract Dried material (833 g) of the leaves of *D canescens* were collected from sandy and disturbed localities in northeastern Baja California and the Colorado desert of California Herbarium specimens were deposited at the UCR Herbarium The ground material was soaked in C_6H_{14} at room temp for 5 days and extracted (×2) for 24 hr Plant material remaining after extraction with C_6H_{14} was soaked in Et_2O for 2 days The Et_2O extract was concentrated *in vacuo* to yield 33 g of a gummy material (4% yield)

Isolation procedures The Et_2O extract (33 g) was dissolved in a small vol of Et_2O or $CHCl_3$ and then 60 g of silica gel was added The solvent mixture was evaporated at room temp and the residue was dried and placed at the top of a silica gel column (105 g, 5 5 cm × 11 cm) and eluted with petrol and Et_2O The column fractions were collected and combined according to TLC

Dihydrodicorin diacetate (**3b**) Fractions 3–5, eluted with 200 ml petrol–Et_2O (1 2) and 800 ml Et_2O, were combined and the solvent removed The extract (2 17 g) was acetylated (Ac_2O, C_5H_5N) at room temp overnight to yield 2 81 g of the crude acetate The mixture was dissolved in a small vol of Et_2O and 3 5 g silica gel were added The solvent was evaporated and the residue placed on the top of silica gel column (63 g, particle size 0 040–0 063 mm, 3 cm × 24 cm) and eluted with petrol and Et_2O The column fractions were collected and combined according to analytical TLC results Fraction 5 (0 15 g), eluted with 80 ml petrol–Et_2O (1 2), was crystallized from a mixture of petrol–Et_2O (1 1) to yield 5 mg dihydrodicorin diacetate (**2b**), mp 140–141°, as white needles Fraction 6 (0 13 g), eluted with 70 ml of the same eluent, was crystallized from the same solvent to yield crystals (40 mg) which were a mixture of dihydrodicorin diacetate (**3b**) and dicorin diacetate (**2b**) as shown by ^{1}H NMR spectroscopy, but which gave one spot on TLC This mixture was further purified by prep TLC using petrol–Et_2O (1 2) to yield 15 mg dihydrodicorin diacetate (**3b**) Recrystallization from petrol and Et_2O yielded 10 9 mg of **3b**, mp 140–141° Fractions 6 and 7, eluted with 320 ml Et_2O, gave a syrup (2 06 g) which was acetylated (8 ml Ac_2O plus and 1 ml C_5H_5N) at room temp overnight The acetylated material was dissolved in Et_2O and the Et_2O soln dried (Na_2SO_4) The crude product (3 1 g) was rechromatographed by flash CC (70 g silica gel, particle size 0 040–0 063 mm) to give fraction 7 which was eluted with 270 ml petrol–Et_2O (1 1) The residue was crystallized from the same solvent to yield crystals (380 mg) as white needles, mp 124–126° The mixture was shown to be of dicorin diacetate (**2b**) and dihydrodicorin diacetate (**3b**) by ^{1}H NMR The mixture was dissolved in 5 ml MeOH and an excess of CH_2N_2 in Et_2O (fresh preparation) was added and the resultant soln left in the refrigerator for 3 hr and then taken to dryness *in vacuo* The residue was dissolved and a small vol of Et_2O and 0 5 g silica gel was added The mixture was shaken, the solvent evaporated and the residue placed on the top of a column (1 5 cm × 16 cm, 10 g silica gel, particle size 0 040–0 063 mm) and eluted with petrol–Et_2O (2 3) Fraction 2, eluted with 37 ml of the eluent, was crystallized from petrol and Et_2O to yield very pure dihydrodicorin diacetate (**3b**), mp 140–141°, 97 mg as white needles, $C_{19}H_{28}O_6$, $[\alpha]_D^{24}$ −95° (*c* 0 49, $CHCl_3$) UV λ_{max}^{MeOH} 224 nm (ε6336), IR ν_{max}^{KBr} cm^{-1} 1760 (q-lactone), 1730, 1725, 1230 and 1020 (acetate) and 1650 (C=C) (partial), ^{1}H NMR (90 MHz, $CDCl_3$) δ1 10 (3H, *d*, *J* = 7 0 Hz, Me-11), 1 23 (6H, *d*, *J* = 6 Hz, Me-4 and Me-10), 2 03 (3H, *s*, Ac), 2 07 (3H, *s*, Ac), 4 70 (1H, *dt*, *J* = 2 5 and 10 Hz, H-9), 4 81 (1H, *sx*, *J* = 6 Hz, H-4) and 5 35 (2H, *d*, *J* = 3 Hz, H-5 and H-6), MS (CI) *m/z* 353 $[M+1]^+$, 293 $[M-59]^+$, 233 $[M-2\times59]^+$

Ivalbatin (**1a**) *and its monoacetate* (**1b**) Fraction 8 from the chromatography of the Et_2O extract was eluted with 150 ml Et_2O and was separated by prep TLC (silica gel, MeOH–$CHCl_3$, 1 19) to give **1a** ^{1}H NMR δ1 20 (3H, *d*, *J* = 7 Hz, Me-10), 2 18 (3H, *s*, Me-4), 3 40 (OH, disappeared on addition of D_2O, OH-9), 3 64 (1H, *d*(*br*), *J* = 10 Hz, H-9), 5 18 (1H, *s*, H-5), 5 44 (1H, *dd*, *J* = 2 5 and 10 Hz, H-6), 5 66 and 6 23 (1H each, exocyclic methylene, *d*, *J* = 3 Hz each) It readily polymerized on standing without solvent, and therefore satisfactory physical data could not be obtained

Fraction 9 (2 03 g) from the Et_2O extract was eluted with 300 ml Et_2O and was acetylated (5 5 ml Ac_2O plus 1 5 ml C_5H_5N) to give 2 57 g of crude acetate after washing with cold H_2O Rechromatography by flash CC with a mixture of petrol–Et_2O (1 2) afforded 0 34 g ivalbatin monoacetate (**1b**) (0 06% from dried leaves of *D canesens*) Recrystallization from petrol and Et_2O, mp 125–127°, $C_{17}H_{22}O_5$, $[\alpha]_D^{24}$ −136° (*c* 0 46, $CHCl_3$) UV λ_{max}^{MeOH} 230 nm (ε6938), IR ν_{max}^{KBr} cm^{-1} 1755 (q-lactone), 1730, 1240 and 1015 (acetate), 1720 (C=O) and 1655 (C=C), ^{1}H NMR (90 MHz, $CDCl_3$) δ1 10 (3H, *d*, *J* = 7 Hz, Me-10), 2 10 (3H, *s*, Ac), 2 15 (3H, *s*, Me-4), 3 30 (1H, *m*, H-7), 5 22 (1H, *s*, H-5), 5 44 (1H, *dd*, *J* = 2 5 Hz and 10 Hz, H-6), 4 75 (1H, *dt*, *J* = 2 5 and 10 Hz, H-10), 5 50 (1H, *d*, *J* = 2 5 Hz, H_a-13) and 6 21 (1H, *d*, *J* = 2 5 Hz, H_b-13), MS (CI) *m/z* 247, 229, 201, 189, 175 and 147

Reduction of ivalbatin monoacetate (**1b**) *with* $NaBH_4$ *to* **3b** *and* **3c** To a soln of 0 11 g **1b** in 3 ml MeOH was added (with stirring) 20 mg $NaBH_4$ over 10 min at 0°, after which stirring was continued for 50 min The reaction mixture was acidified with HOAc (pH 5) and taken to dryness under red pres Iced H_2O was added and stirred to yield a solid which was recovered by filtration, washed with H_2O and dissolved in Et_2O The Et_2O soln was dried (Na_2SO_4) and concd to yield 78 mg of **4**, which polymerized on standing without solvent and was therefore acetylated (1 ml Ac_2O plus 0 3 ml C_5H_5N) to yield 61 9 mg crude

acetates **3b** and **3c** Rechromatography by flash CC with petrol–Et_2O (3 2 and then 1 1) gave dihydrodicorin diacetate (**3b**), 15 mg Recrystallized from Et_2O and petrol, mp 140–141°, as white needles UV λ_{max}^{MeOH} 229 nm (ε6272), IR ν_{max}^{KBr} cm^{-1} 1760 (q-lactone), 1730, 1725, 1230 and 1020 (acetate) and 1650 (C=C) (partial), MS (CI) *m/z* 353 $[M+1]^+$ ($C_{19}H_{28}O_6+1$) 293, 279 and 233 Its TLC, UV, IR, MS and ^1H NMR properties were identical with those of dihydrodicorin diacetate (**3b**) from *D canescens* and its mp was not depressed on admixture with dihydrodicorin diacetate (**3b**)

The mixture was eluted with petrol–Et_2O (1 1) to give isodihydrodicorin diacetate (**3c**), 27 mg Recrystallization from the same solvent, mp 67–70° yielded **3c** as needles, $[\alpha]_D^{24}$ −58° (*c* 0 6, $CHCl_3$) UV, IR and mass spectra identical to those of **3c** ^1H NMR (different from **3b**) δ1 06 (3H, *d*, *J* = 7 Hz, Me-10), 1 17 (3H, *d*, *J* = 7 Hz, Me-11), 1 24 (3H, *d*, *J* = 7 Hz, Me-4), 2 03 (3H, *s*, Ac), 2 07 (3H, *s*, Ac), 4 83 (2H, *m*, H-4 and H-9), 5 20 (1H, *m*, H-5) and 5 38 (1H, *d*, *J* = 3 Hz, H-6)

Dicorin (**2a**) Fractions 13, 14 and 15 (3 4 g) from the Et_2O extract were eluted with 960 ml Et_2O Rechromatography by flash CC with MeOH–$CHCl_3$ (1 9) yielded pure dicorin (**2a**), $C_{15}H_{22}O_4$, 2 2 g as a gummy syrup (0 7% from the dried leaves of *D canesens*), which did not yield any crystals after numerous attempts $[\alpha]_D^{24}$ −117° (*c* 1 3, $CHCl_3$), UV λ_{max}^{MeOH} 228 nm (ε6571), IR ν_{max}^{film} cm^{-1} 3560 (OH), 1770 (q-lactone) and 1670 (C=C), ^1H NMR (90 MHz, $CDCl_3$) δ1 18 (6H, *d*, *J* = 7 Hz, Me-4 and Me-10), 2 94 (2H, *br*, disappeared on addition of D_2O), 3 40 (1H, *m*, H-7), 3 61 (1H, *d*(*br*), *J* = 10 Hz, H-9), 3 82 (1H, *sx*, *J* = 6 Hz, H-4), 5 30 (1H, *s*, H-5), 5 42 (1H, *dd*, *J* = 2 5 and 10 Hz, H-6), 5 57 and 6 26 (1H each, *d*, *J* = 3 Hz each, exocyclic methylene, H_2-13), MS (CI) *m/z* 267 $[M+1]^+$, 249 $[M-18]^+$ and 231 $[M-36]^+$

Dicorin diacetate (**2b**) Approximately 1 13 g **2a** was acetylated (3 ml Ac_2O plus 0 5 ml C_5H_5N) at room temp overnight, and then ice H_2O was added to hydrolyse the Ac_2O and yield 1 5 g of a white powder Recrystallization from Et_2O and petrol afforded 1 063 g of pure dicorin diacetate (**2b**), mp 118–120°, $C_{19}H_{26}O_6$, $[\alpha]_D^{24}$ −100° (*c* 0 77, $CHCl_3$) UV λ_{max}^{MeOH} 229 nm (ε6205), IR ν_{max}^{KBr} cm^{-1} 1760 (q-lactone), 1725, 1240 and 1015 (acetate) and 1660 (C=C), ^1H NMR (250 MHz, $CDCl_3$) see text

Reduction of **2b** *with* $NaBH_4$ *to* **3b** *and* **3c** To 0 27 g **2b** in 3 ml MeOH was added with stirring 60 mg $NaBH_4$ over 10 min at 0°, and then the stirring and was continued for 20 min The reaction mixture was acidified with HOAc, concd *in vacuo*, and the residue added to ice water and extracted with Et_2O (× 2) The Et_2O soln was dried (Na_2SO_4) and then evaporated to give 0 315 g Rechromatography by flash CC with petrol followed by a gradient of Et_2O–petrol (1 2 to 1 1) gave dihydrodicorin diacetate (**3b**), mp 140–141° and isodihydrodicorin diacetate (**3c**), mp 67–69° Their UV, IR, mass and ^1H NMR spectra were identical to those obtained for the ivalbatin reduction products

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