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SHORT COMMUNICATION

SESQUITERPENE LACTONES: CONSTITUENTS OF AN F_1 HYBRID *ENCELIA FARINOSA* × *ENCELIA CALIFORNICA*

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Abstract—The principal component of *Encelia californica* Nutt., encecalin, is not present in *Encelia farinosa* Gray, and the principal components of the latter, encelin and farinosin, are absent from the former. An F_1 hybrid of the two species contains encecalin and farinosin and, in addition, a new lactone, structurally related to farinosin, which is absent from the parental species.

INTRODUCTION

THE CHEMISTRY of interspecific hybrids *vis-à-vis* the constitution of the parental species has been the subject of numerous studies with respect both to primary constituents (e.g. by serology) and to secondary metabolites.[1] While the presence in the hybrid of constituents occurring in both parents has often been observed, the presence of hybrid-specific secondary metabolites is relatively uncommon. The present study has disclosed the presence in a hybrid of the characteristic parental constituents and, in addition, a compound not present in either progenitor.

RESULTS AND DISCUSSION

Encelia farinosa Gray is a shrubby perennial widespread in the lower deserts of southern California and Arizona. *Encelia californica* Nutt. is also an abundant perennial found chiefly in the higher deserts and coastal regions. The two plants are clearly distinguishable in morphology and habit. In certain areas between the desert habitat of *E. farinosa* and the coastal habitat of *E. californica* are found hybrids which possess some of the morphological features of both parents. Plants identified† as F_1 hybrids of the two species were collected near Riverside, California and examined for their chemical constituents. Several individual hybrid plants, collected in a localized area, showed identical patterns of spots on thin-layer chromatograms, and were combined for study.

Extraction of the plant material in the usual manner yielded encecalin[2] and farinosin[3,4] in amounts of 0·10% and 0·06%, respectively. A new lactone, hybrifarin was isolated in 0·01% yield.

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† Mr Albert Hill, Department of Botany, U.C.L.A., who has produced F_1 hybrids in laboratory experiments. authenticated the hybrid character of the native plants used in this study. Specimen voucher No. LB-01669-EH.

[1] R. E. ALSTON and B. L. TURNER, *Biochemical Systematics*, Chapter 5, Prentice-Hall, Englewood Cliffs (1963).

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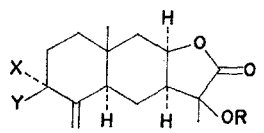
[3] T. A. GEISSMAN and R. MUKHERJEE, *J. Org. Chem.* **33**, 656 (1968).

[4] W. HERZ, P. S. SUBRAMANIAN and T. A. GEISSMAN, *J. Org. Chem.* **33**, 3743 (1968).

Hybrifarin, $C_{15}H_{22}O_4$, showed the molecular ion peak at *m/e* 266 in the mass spectrum, along with ions at *m/e* 251 (M-15), 222 (M-44) and, in common with most other lactones of the class, at 91 ($C_7H_7^+$) and 105 ($C_8H_9^+$). Its i.r. spectrum showed absorption at 3240 (OH) and 1770 (γ-lactone) cm^{-1}. The NMR spectrum contains signals for an exocyclic methylene group (broadened singlets, δ 4·86 and 5·75, one proton each) and two three-proton singlets at δ 0·91 and δ 1·70. A broad one-proton multiplet at δ 5·26 is nearly the same in position and coupling pattern as the C-8 proton in farinosin, and indicates that the lactone is closed C-7/C-8. A one-proton multiplet ($W_{\frac{1}{2}}$ = 13 Hz) at δ 4·20, shifting to 5·10 in the acetate, corresponds to that reported[4] for the C-3 proton of —CHOH of epiisotelekin, and is assigned to the same position in hybrifarin. The degree of coupling of this proton with the 2-CH_2 group indicates that the 3-OH group is β. These data are indicative of the structure I for hybrifarin.



I, X = H; Y = OH; R = H
II, X,Y = O; R = H
III, X = H; Y = OAc; R = Ac

It will be noted that many of the principal skeletal features of I are similar to those of farinosin. In particular, the absence of the very common α-methylene-γ-lactone grouping, and the presence of a methyl group singlet at δ 1·70, indicating the presence of the α-hydroxy-α-methyl-γ-lactone, are distinctive.

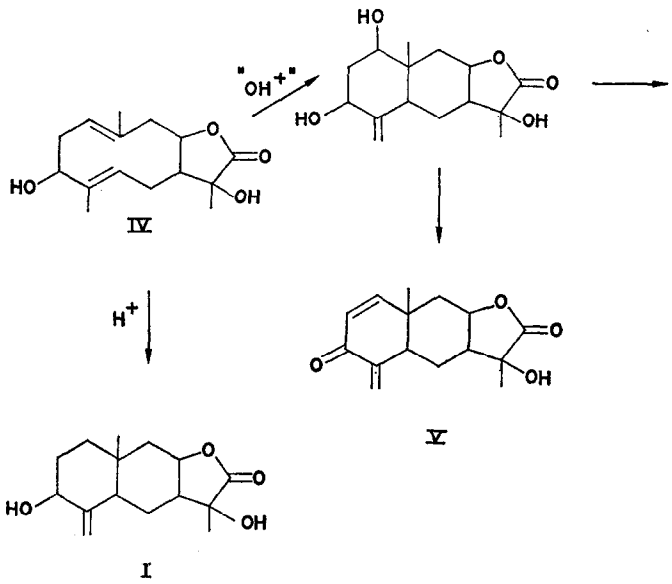
Oxidation of hybrifarin transformed it into the corresponding ketone (II), the absorption maximum (226 nm) and extinction coefficient (4920) of which are in complete agreement with the structure II. Finally the formation of the diacetate III is in accord with the assigned structure, for the acetylation of the tertiary hydroxyl group indicates its position in the position adjacent to the lactone carbonyl group. Farinosin[3,4] forms a corresponding tertiary acetate. The downfield shift of the signal for the C-11 methyl group upon acetylation is consistent with the structure assigned.

The results of this examination of the hybrid are in accord with the generalizations stated by Alston and Turner.[1] Compounds present in minor amounts in the parental species—euparin and euparone methyl ether in *E. californica*, and encelin in *E. farinosa*—could not be detected in the hybrid. Moreover, encecalin is absent from *E. farinosa*, a fact established by vapor phase chromatographic examination of extracts of the latter. Although no hybrifarin could be detected in *E. farinosa*, there is, of course, the possibility of its presence in amounts below the level of recognition.

The close structural resemblance of hybrifarin and farinosin clearly suggests their derivation by related biosynthetic pathways. A precursor such as IV could, it will be apparent, cyclize oxidatively (for example, by epoxidation at the C-1 (10) double bond), or by proton catalysis, to give in the first case the precursor of farinosin (V), and in the second hybrifarin.

EXPERIMENTAL

Extraction of plant material. Plants consisting of the F_1 progeny of *Encelia farinosa* Gray × *E. californica* Nutt. were collected near Riverside, California in late June, 1969. The dried and powdered leaf material (3·47 kg) was extracted with $CHCl_3$ at room temp. and the residue (300 g) remaining after removal of the



solvent was shaken with a mixture of 2 l. of hexane and 2 l. of 3:1 methanol–water. Of the compounds present in the whole extract (by TLC), only encecalin was present in the hexane layer. Removal of the hexane and distillation under reduced pressure yielded 2·8 g of encecalin, which was identified by its i.r. and NMR spectra and by formation of the oxime, m.p. 139–140° (reported,[2] m.p. 140°).

Extraction of the aqueous layer with $CHCl_3$ yielded an oily mixture which was chromatographed over silica gel (1·36 kg), elution being carried out with $CHCl_3$ containing increasing proportions of acetone. Fractions of 1 l. were collected, fraction 25 being eluted with pure acetone.

Fraction 4 yielded 0·5 g of encecalin. Fractions 10–14 yielded crystalline residues which, after recrystallization, afforded 1·7 g of farinosin, identified by comparison with an authentic specimen.

Hybrifarin (*I*). Fractions 18–19 yielded a residue which was recrystallized from $CHCl_3$–pentane to give 35 mg of hybrifarin, m.p. 224–226°. *Anal.* Calc. for $C_{15}H_{22}O_4$: C, 67·64; H, 8·33. Found; C, 67·56; H, 8·41. The spectral properties have been described in the Discussion.

Hybrifarin diacetate (*III*). Acetylation of hybrifarin with Ac_2O and pyridine afforded the diacetate, colorless spears from benzene–hexane, m.p. 188–189°. The mass spectrum showed the molecular ion at *m/e* 350, and prominent peaks at *m/e* 308 (M-42), 290 (M-60), 248 (M-42-60) and 230 (M-60-60).

The NMR spectrum showed two three-proton singlets for the acetyl methyl groups at δ 2·02 and 2·12. The three-proton singlet for the C-11 methyl group appears at δ 1·77, shifted downfield from its position (δ 1·70) in hybrifarin. *Anal.* Calc. for $C_{19}H_{20}O_6$: C, 65·12; H, 7·48. Found: C, 65·28; H, 7·59.

Hybrifarone (*II*). A solution of 10 mg of hybrifarin in 2 ml of acetone was treated with 0·2 ml of Jones' reagent. After 5 min the solution was diluted with water and extracted with $CHCl_3$. Removal of the solvent and recrystallization of the residual solid from $CHCl_3$–hexane yielded colorless prisms, m.p. 273–275° *Anal.* Calc. for $C_{15}H_{20}O_4$: C, 68·16; H, 7·63. Found: C, 67·93; H, 7·59.

The mass spectrum showed the molecular ion at *m/e* 264, along with prominent ions at *m/e* 220 (M-44) and 205 (M-44-15). The i.r. spectrum showed peaks at 3380 (OH), 1770 (γ-lactone) and 1700 (ketone) cm^{-1}, and the u.v. spectrum showed a maximum at 226 nm (ε 4920).

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