Antineoplastic and Cytotoxic Components of Desert *Baileya*

Keyphrases □ *Baileya multiradiata*—six antineoplastic and cytotoxic components, isolated and evaluated □ Antineoplastic agents, potential—isolated from *Baileya multiradiata*, biological evaluation □ Cytotoxic agents—isolated from *Baileya multiradiata*, biological evaluation

To the Editor:

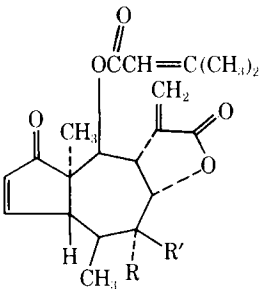
The bright-yellow ray-florets of *Baileya multiradiata* Harv. and Gray (family Compositae)[1] are commonly seen in the southwestern United States from Texas to California. The flowering portion is attractive to sheep, and ingestion has led to the death of many such range animals (1).

In 1966, we collected *B. multiradiata* in Arizona for evaluation in respect to possible antineoplastic components (2). Later, an ethanol extract of the complete plant was found to reach confirmed active status in the National Cancer Institute's murine lymphocytic leukemia P-388 screen. A cell line derived from this PS system suitable for *in vitro* measurements was studied as a bioassay method for isolating the antineoplastic constituents of *B. multiradiata*.

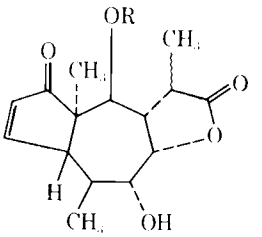
We now wish to report the isolation of six cytotoxic and antineoplastic constituents of *B. multiradiata* and that the new P-388 *in vitro* screening technique[2] proved very efficient for this purpose. The results of this bioassay are expressed as ED_{50}, namely, that concentration of material (in micrograms per milliliter) that produces 50% inhibition of growth in a 48-hr incubation period.

A chloroform-soluble fraction (5.98 g) prepared from *B. multiradiata* Harv. and Gray was partially separated by chromatography on cross-linked dextran gel[3]. The fraction (4.8 g) eluted with methanol–chloroform (6:4) was subjected to careful gradient (benzene to ethyl acetate) elution chromatography on silica gel. The following were obtained (in order of isolation): fastigilin C (I*b*, 0.13 g, ED_{50} 0.004)[4] (4), fastigilin B (II*a*, 0.09 g, ED_{50} 0.078) (4), baileyin (III, 0.05 g, ED_{50} 0.47) (5), and pleniradin (IV*a*, 2.03 g, ED_{50} 5.7), characterized as the monoacetate derivative IV*b* (ED_{50} 2.9) (6).

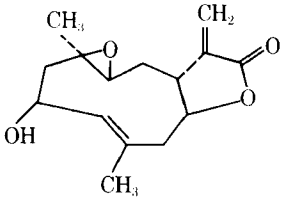
A new pseudoguaianolide, designated multiradiatin (I*a*, ED_{50} 0.02, crystals from acetone–heptane, 0.19 g, mp 226–230°) (4), as well as radiatin (II*b*, 0.67 g, ED_{50} 0.39) (6), was isolated by analogous large-scale chromatography (161 g of chloroform fraction). The structural assignment for multiradiatin was confirmed by the identity of this substance with the ketone obtained by chromic acid (Jones reagent) oxidation of fastigilin C. Both fastigilin C and baileyin (III) were isolated previously from *B. multiradiata* by Waddell and Geissman (5).



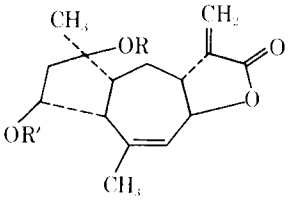
I*a*: R = R′ = O
I*b*: R = OH, R′ = H



II*a*: R = COCH=C(CH$_3$)$_2$
II*b*: R = COC(=CH$_2$)CH$_3$



III



IV*a*: R = R′ = H
IV*b*: R = H, R′ = COCH$_3$

In vivo biological studies against several tumors of the National Cancer Institute's screening systems are now in progress. At present, consistent *in vivo* activity has been obtained, for example, with fastigilin C (I*b*), which in the PS screen leads to a 50% increase in survival time at a dose of 3 mg/kg. Recently, we (7) summarized analogous and encouraging biological activity for the related pseudoguaianolide helenalin. Thus, results of the present study suggest that the pseudoguaianolide lactone of the fastigilin, helenalin, and radiatin type possesses (in addition to the usual cytotoxic properties of such lactones) carrier groups that fall within requirements for *in vivo* antineoplastic activity. Also, the cytotoxicity displayed by the sesquiterpene lactones of *B. multiradiata*, while correlated with the antineoplastic activity, may suggest that these components are responsible for sheep poisonings attributed to this plant.

[1] The plant material was identified by Mrs. E. Lehto, Herbarium curator, Department of Botany, Arizona State University. Voucher specimens are preserved in our Institute and in the University's Herbarium.

[2] For results of a study employing L-1210 cells *in vitro* as a cytotoxicity assay, refer to Ref. 3.

[3] Sephadex LH-20.

[4] Spectral data (mass, IR, and PMR) and elemental analyses were consistent with each structural assignment. Reported yields correspond to the recrystallized (from benzene–ethyl acetate unless otherwise noted) product.

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[x] To whom inquiries should be directed.

Germination and Morphine Content of *Papaver somniferum* Plants Produced from Commercially Available Poppy Seed

Keyphrases □ *Papaver somniferum*—germination and cultivation from commercial poppy seeds, morphine content, TLC, column chromatography, IR and mass spectra □ Morphine—isolated from *Papaver somniferum* germinated and cultivated from commercial poppy seeds, TLC, column chromatography, IR and mass spectra □ Narcotics—morphine, isolated from *Papaver somniferum* germinated and cultivated from commercial poppy seeds, TLC, column chromatography, IR and mass spectra

To the Editor:

There are no legal constraints to the possession of seeds of *Papaver somniferum*, the opium poppy. However, it is illegal to cultivate this plant in the United States. This paradox is interesting since *Cannabis sativa* (marijuana) seeds must be sterilized prior to sale. The narcotic regulations do not require sterilization of *P. somniferum* seeds.

Because *P. somniferum* could be illicitly cultivated in the United States as a source of codeine and morphine, the present study was carried out to answer the following questions. What is the botanical identification of commercially available poppy seeds? What is the germination rate of such seeds? And will plants produced from commercial poppy seeds grow and produce morphine in a temperate climate such as we have in the United States?

Poppy seeds (100-g samples) were obtained from two commercial bakeries in Chicago as well as from three supermarkets in the area. The seeds were soaked for 2 min in a dilute solution of sodium hypochlorite, washed thoroughly with water, and germinated on wet vermiculite. Approximately 60% of the seeds from each of five lots germinated.

After the seedlings developed well-expanded cotyledons, they were transplanted to potting soil. The plants were grown in a greenhouse in 10.2-cm (4-in.) diameter clay pots. Flowering occurred about 8 weeks after germination. Greenhouse plants were similar in appearance to fieldgrown plants, although they were smaller[1].

Immature capsules of one seed lot (I-9786) were pierced with a suitable needle daily, and the dried latex was scraped from the capsules and accumulated. A total of 24 mg of dried latex was collected in this manner from several capsules.

When the plants were fully mature, the entire plants were dried and then subjected to alkaloid extraction. The latex (24 mg), which had been scraped from the immature capsules, was dissolved in 15 ml of 1% HCl and filtered. After the filtrate was made alkaline with ammonium hydroxide, it was extracted three times with 15-ml volumes of chloroform. The combined chloroform extracts were taken to dryness to yield a 10-mg residue, which was used in the TLC analyses.

TLC of the extract on silica gel G plates, when compared with an authentic morphine sample, indicated morphine to be present in the alkaloid extract in addition to several other alkaloids.

Chromatography of the alkaloid extract over a small column (disposable pipet) containing 1.5 g of silica gel PF_{254}, using chloroform–methanol–ammonium hydroxide (90:10:1) as the eluent, resulted in the isolation of a compound (2 mg) whose R_f on silica gel PF_{254} plates, using the same solvent system, was 0.71.

The mass spectrum of the compound indicated a molecular ion M^+ at *m/e* 285 nm, and the fragmentation pattern was consistent with that reported for morphine (1).

To confirm this identification, additional capsules and foliage (8.38 g) were processed for alkaloids as already described. We wanted to determine if a larger quantity of the compound, tentatively identified as morphine, could be isolated and more definitively identified. TLC analysis of the resulting alkaloid extract indicated the same pattern as was observed for the alkaloid extract prepared from the latex.

Column chromatography of this extract, using 10 g of silica gel PF_{254}, resulted in the isolation of 7.6 mg of the compound. The UV spectrum of the sample (in ethanol) showed a maximum absorption at 284 nm,

[1] The plants were identified as *Papaver somniferum* L. (Papaveraceae) by F. A. Crane, and herbarium specimens (I-9786–I-9790) were deposited at the Department of Pharmacognosy and Pharmacology, College of Pharmacy, University of Illinois at the Medical Center, Chicago, IL 60612