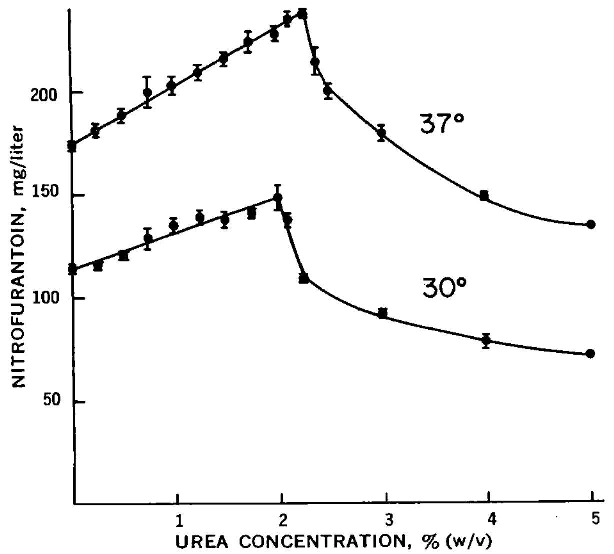


Figure 1—*Nitrofurantoin solubility in aqueous urea solutions at 30 and 37°. Bars mark off 1* SD *on either side of the average.*

not possible now. Preliminary spectral studies suggest some type of interaction between nitrofurantoin and urea molecules. This possible interaction may account for the increased solubility at low urea concentrations. The breakdown of water structure might account for the increase in solubility at lower urea concentrations; however, disruption of the water structure might not occur at the low urea concentrations used. The rapid decrease in nitrofurantoin solubility at higher urea concentrations may be due to a salting-out effect. However, the formation of an insoluble complex at higher urea concentrations might account for the abrupt decrease in solubility.

Further investigations are being conducted in these laboratories to determine the effect of urea and another urine component, creatinine, on nitrofurantoin solubility at various temperature and pH conditions and to elucidate the mechanism(s) of action of the solubilization phenomena.

(1) H. E. Paul and M. F. Paul, in "Experimental Chemotherapy," vol. II, R. J. Schnitzer and F. Hawking, Eds., Academic, New York, N.Y., 1964, p. 334.

(2) M. F. Paul, R. C. Bender, and E. G. Nohle, *Amer. J. Physiol.*, **197,** 580(1959).

(3) T. R. Bates, J. M. Young, C. M. Wu, and H. A. Rosenberg, *J. Pharm. Sci.*, **63,** 643(1974).

(4) W. A. Richards, E. Riss, E. H. Kass, and M. Finland, *AMA Arch. Intern. Med.*, **96,** 437(1955).

(5) E. H. Beutner, J. J. Petronio, H. E. Lind, H. M. Trafton, and M. Correia-Branco, in "Antibiotics Annual 1954–1955," H. Welch and F. Marti-Ibanez, Eds., Medica Encyclopedia Inc., New York, N.Y., 1955, p. 988.

(6) M. F. Paul, C. Harrington, R. C. Bender, E. Nohle, and M. J. Bryson, *Proc. Soc. Exp. Biol. Med.*, **125,** 941(1967).

(7) J. D. Conklin and R. D. Hollifield, *Clin. Chem.*, **11,** 925(1965).

(8) H. A. Harper, "Review of Physiological Chemistry," 14th ed., Lange Medical Publications, Los Altos, Calif., 1973, p. 398.

(9) A. White, P. H. Handler, and E. L. Smith, "Principle of Biochemistry," 3rd ed., McGraw-Hill, New York, N.Y., 1964, p. 737.

(10) K. Sekiguchi, N. Obi, and Y. Ueda, *Chem. Pharm. Bull.*, **12,** 134(1964).

(11) A. H. Goldberg, M. Gibaldi, J. L. Kanig, and M. Mayersohn, *J. Pharm. Sci.*, **55,** 581(1966).

(12) A. H. Goldberg, M. Gibaldi, and J. L. Kanig, *ibid.*, **55,** 482(1966).

(13) K. Sekiguchi and N. Obi, *Chem. Pharm. Bull.*, **9,** 866(1961).

(14) P. Mukerjee and A. K. Ghosh, *J. Phys. Chem.*, **67,** 193(1963).

(15) C. McDonald and E. Lindstrom, *J. Pharm. Pharmacol.*, **26,** 39(1974).

(16) S. Bolton, *J. Pharm. Sci.*, **52,** 1071(1963).

(17) D. M. Altwein, J. N. Delgado, and F. P. Cosgrove, *ibid.*, **54,** 603(1965).

(18) S. Feldman and M. Gibaldi, *ibid.*, **56,** 370(1967).

(19) D. B. Wetlaufer, S. K. Malik, L. Stoller, and R. L. Coffin, *J. Amer. Chem. Soc.*, **86,** 509(1964).

Donald E. Cadwallader ˣ
Hung Won Jun
Lian-Kaun Chen

Department of Pharmacy
School of Pharmacy
University of Georgia
Athens, GA 30602

Received December 20, 1974.
Accepted for publication February 2, 1975.

ˣ To whom inquiries should be directed.

Ambrosin, Tumor Inhibitory Agent from *Hymenoclea salsola* (Asteraceae)

Keyphrases □ Ambrosin—tumor inhibitory agent from *Hymenoclea salsola,* isolation and identification □ *Hymenoclea salsola* (Asteraceae)—isolation and identification of ambrosin, a tumor inhibitory agent □ Antitumor agents—isolation and identification of ambrosin from *Hymenoclea salsola*

To the Editor:

As a result of the continuing search for plants having tumor inhibitory constituents, it was found that the chloroform extract of the leaves and stems of *Hymenoclea salsola* Torr. and Gray (Asteraceae)[1] showed inhibitory activity toward the P-388 lymphocytic leukemia test system (3PS)[2].

One of the two major constituents of the chloroform extract was shown to be ambrosin, previously isolated from many plants of the Asteraceae family including *H. salsola* (1). Isolation was effected by column chromatography, recrystallization, and preparative TLC[3]. Identification was achieved by IR, NMR, mass spectrometry, elemental analysis, and comparison with an authentic specimen[4].

Ambrosin demonstrated activities of 180, 158, 130, and 132% test/control (T/C) at 35, 22, 14, and 9.6 mg/kg, respectively, in the 3PS system. Activity in

[1] Identification was confirmed by Dr. Robert E. Perdue, Medicinal Plant Resources Laboratory, Plant Genetics and Germ Plasm Institute, Beltsville, Md. A reference specimen was deposited in that herbarium. The plant was collected in California in December 1973.

[2] Division of Cancer Treatment, National Cancer Institute, National Institutes of Health, Bethesda, Md.

[3] In our plant extract, ambrosin crystallized as a 1:1 mixture with its 11,13-dihydro derivative, so preparative TLC was required to isolate pure ambrosin. Geissman and Toribio (1) detected none of the dihydro material in their plant extract.

[4] We are indebted to Professor Werner Herz, Florida State University, Tallahassee, Fla., for providing the authentic sample of ambrosin.

the 3PS test system is defined as an increase in the survival of treated animals over that of control animals resulting in a $T/C \geqq 125\%$ (2).

(1) T. A. Geissman and F. P. Toribio, *Phytochemistry*, **6**, 1563(1967).
(2) R. I. Geran, N. H. Greenberg, M. N. MacDonald, A. M. Schumacher, and B. J. Abbott, *Cancer Chemother. Rep.*, **3** (3), 9(1972).

S. J. Torrance
R. M. Wiedhopf
J. R. Cole ˣ

Division of Pharmaceutical Chemistry
College of Pharmacy
University of Arizona
Tucson, AZ 85721

Received January 27, 1975.
Accepted for publication February 19, 1975.
Supported by Contract NO1-CM-33750 from the Division of Cancer Treatment, National Cancer Institute, National Institutes of Health.
ˣ To whom inquiries should be directed.

Chemical Constituents of Gentianaceae XVI: Antitubercular Activity of Xanthones of *Canscora decussata* Schult

Keyphrases □ *Canscora decussata* Schult—antitubercular activity of polyoxygenated xanthones □ Xanthones, polyoxygenated—activity against *Mycobacterium tuberculosis* □ Mangiferin—activity against *Mycobacterium tuberculosis* □ Medicinal plants—antitubercular activity of xanthones of *Canscora decussata* Schult

To the Editor:

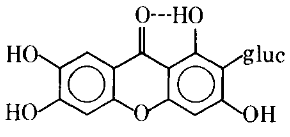
The extract of *Canscora decussata* Schult (Gentianaceae) is used in the treatment of certain mental disorders and of tuberculosis in the Indian system of medicine (1). Previously, the isolation, characterization, and pharmacological screening of about two dozen polyoxygenated xanthones of *C. decussata* were reported from this laboratory (2–5). The pharmacological profile of activities of the total xanthones (excluding mangiferin) and of the major glycoxanthone, mangiferin, was consistent with the reported uses of the plant extract in the treatment of affective disorders. We now wish to report the significant antitubercular activity of the free polyoxygenated xanthones of this medicinal plant.

After concentration, the ethanolic extract of the defatted plant material yielded mangiferin in high yield. The alcoholic mother liquor, after the separation of mangiferin, gave a syrupy mass with further concentration. The nitrogenous constituents were separated from it by aqueous acetic acid treatment in the usual way (2). The chloroform-soluble fraction of the acidic aqueous suspension gave a mixture of about a dozen polyoxygenated xanthones ("total xanthones"); their number and relative abundance were monitored by TLC and mass spectrometry of the mixture, its methyl ethers, and the acetates.

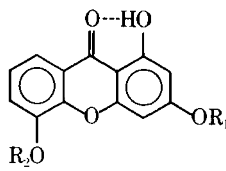
The major xanthonic constituent was mangiferin (I). The remaining total xanthones consisted of three 1,3,5-trioxygenated xanthones (II*a*–II*c*, relative proportion about 12%), four 1,3,5,6-tetraoxygenated xanthones (III*a*–III*d*, 32%), and four 1,3,5,6,7-pentaoxygenated xanthones (IV*a*–IV*d*, 56%), plus some unidentified minor xanthones. All of these compounds were screened for activity against *Mycobacterium tuberculosis* H37 RV using Youman's medium. The tube dilution method was used. The test compounds were added in different concentrations to Youman's medium tubes containing horse serum (10%). The tubes were then inoculated with the organism (about 10^6 million/ml). Thereafter, the tubes were incubated at 37° for 21 days.

After the incubation, the tubes were observed for growth of the microorganism. The minimum concentration of the compounds required for preventing the growth was recorded as the minimum inhibitory concentration (MIC). The total xanthones (II–IV) were found to be more active than mangiferin. The MIC (10 μg/ml) of the total xanthones was comparable to that of streptomycin. Mangiferin showed only weak inhibitory activity, its MIC being 200 μg/ml.

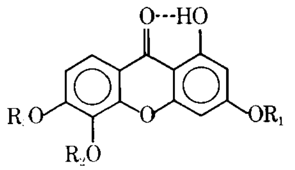
The activity of this class of compounds against *M. tuberculosis* was reported twice before in the literature. 1,3,8-Trihydroxyxanthone, a degradation product of sterigmatocystin (occurring in *Aspergillus versicolor*), was reported (6) to be active at a dilution of 1 in 80,000. Sterigmatocystin had virtually no tuberculostatic effect *per se*. Norswertianolin (3,5,8-trihydroxyxanthone-1-*O*-glucoside), occurring in a



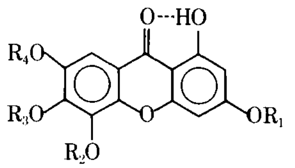
I



II*a*: $R_1 = H$, $R_2 = H$
II*b*: $R_1 = CH_3$, $R_2 = H$
II*c*: $R_1 = CH_3$, $R_2 = CH_3$



III*a*: $R_1 = H$, $R_2 = H$, $R_3 = H$
III*b*: $R_1 = H$, $R_2 = CH_3$, $R_3 = H$
III*c*: $R_1 = H$, $R_2 = H$, $R_3 = CH_3$
III*d*: $R_1 = H$, $R_2 = CH_3$, $R_3 = CH_3$



IV*a*: $R_1 = H$, $R_2 = H$, $R_3 = H$, $R_4 = H$
IV*b*: $R_1 = CH_3$, $R_2 = CH_3$, $R_3 = H$, $R_4 = H$
IV*c*: $R_1 = H$, $R_2 = CH_3$, $R_3 = CH_3$, $R_4 = H$
IV*d*: $R_1 = CH_3$, $R_2 = CH_3$, $R_3 = CH_3$, $R_4 = H$