strate little activity against *E. coli* (1), suggesting that only small amounts of furazolidone are excreted in dog and human urine. This also indicates that the urinary drug-related metabolites observed in the present study are not significantly active against *E. coli*.

In summation, when urines collected from dogs and humans administered furazolidone orally were analyzed by a new analytical procedure, furazolidone was not detected. Chromatographic examination of these urine samples verified this conclusion and also revealed the presence of two drug-related metabolites in both dog and human urine.

REFERENCES

(1) Paul, H. E., and Paul, M. F., in "Experimental Chemotherapy," vol. II, Academic Press Inc., New York, N. Y., 1964, pp. 307–370.
(2) *Ibid.*, vol. IV, 1966, pp. 521–536.
(3) Nakamura, N., and Inoue, S., *Ann. Rept. Takeda Res. Lab.*, **12**, 16(1961).
(4) Herrett, R. J., and Buzard, J. A., *Anal. Chem.*, **32**, 1676(1960).
(5) Buzard, J. A., Vrablic, D. M., and Paul, M. F., *Antibiot. Chemotherapy*, **6**, 702(1956).
(6) Paul, H. E., Hayes, K. J., Paul, M. F., and Borgmann, A. R., *J. Pharm. Sci.*, **56**, 882(1967).
(7) Paul, H. E., Ellis, V. R., Kopko, F., and Bender, R. C., *J. Med. Pharm. Chem.*, **2**, 563(1960).
(8) Olivard, J., Valenti, S., and Buzard, J. A., *ibid.*, **5**, 524(1962).
(9) Tennent, D. M., and Ray, W. H., *Federation Proc.*, **22**, 367(1963).

Keyphrases

Furazolidone
Urine–furazolidone analysis procedure
Metabolites, furazolidone–analysis in urine
Colorimetry–analysis
Paper chromatography–analysis

Fractionation of Fatty Acids of *Cucurbita maxima* Seed Oil With Urea

By J. P. TEWARI and M. C. SRIVASTAVA

The mixed fatty acids of *Cucurbita maxima* seed oil have been fractionated by liquid–solid countercurrent distribution with urea. The percentage fatty acid composition of oil is: palmitic, 21.5; stearic, 8.4; oleic, 27.0; and linoleic, 43.10.

UNDER the Indian Council of Medical Research inquiry on anthelmintic activity of *Cucurbita maxima* seeds (family, *Cucurbitaceae*), the anthelmintic activity and the chemotherapeutic actions of aqueous, alcoholic, and ethereal extracts of the decorticated seeds of *C. maxima* have been reported (1, 2).

A large amount of the oil was obtained as a by-product during the defatting of the seeds prior to the isolation of cucurbitin. In view of the importance of this oil in the Indian system of medicine (3, 4) the mixed fatty acids of *C. maxima* oil were fractionated by the liquid–solid countercurrent distribution of fatty acids with urea employing the method of Sumerwell (5).

The results of fractionation have been recorded in Table I and agree closely with those obtained by Chowdhury *et al.* spectrophotometrically (6).

TABLE I—LIQUID–SOLID COUNTERCURRENT DISTRIBUTION OF *C. maxima* FATTY ACIDS WITH UREA

S. No. of Fraction	Wt. of Fraction	S.E.	I.V.	Palmitic Acid	Stearic Acid	Oleic Acid	Linoleic Acid
1	3.28	205.2	0.82	1.20	2.04	0.03	...
2	2.61	215.8	2.42	2.26	0.28	0.07	...
3	2.08	215.2	2.52	1.72	0.19	0.18	...
4	1.56	216.2	6.92	1.38	0.06	0.12	...
5	...	...	...	...	...	...	...
6	0.61	195.4	90.1	...	...	0.61	...
7	...	...	...	...	...	...	...
8, 9, 10	0.91	196.4	90.2	...	...	0.91	...
11	4.85	197.8	121.2	...	...	3.20	1.65
Raffinate	14.60	198.5	162.0	...	...	3.10	11.50
Total	30.50	...	...	6.56	2.57	8.22	13.15
Percentage of acids	...	...	...	21.5	8.4	27.0	43.1
Percentage of acids by Chowdhury (6)	...	...	...	←——29.9——→		26.4	43.7

EXPERIMENTAL

The oil from the seeds of *C. maxima* was saponified and fatty acids were obtained from the soap after removing the unsaponifiable matter. The mixed fatty acids (30.5 Gm. I.V., 103.2; N.V., 198.2) were fractionated by liquid–solid countercurrent distribution of fatty acid with urea employing the method

Received April 26, 1967, from the Department of Pharmacology, G.S.V.M. Medical College, Kanpur, India.
Accepted for publication August 18, 1967.
This work was generously supported by a grant from I.C.M.R., New Delhi, India.

of Sumerwell (5) as described by Tewari and coworkers (8). Each fraction was analyzed for iodine values (I.V.) and saponification equivalent (S.E.). The composition of fractions was calculated by the method of Hilditch (7) and the result recorded in Table I.

REFERENCES

(1) Srivastava, M. C., Tewari, J. P., Singh, S. W., Gupta, M. L., and Mishra, K. C., *Labdev J. Sci. Tech.*, **5**, 64(1967).
(2) Srivastava, M. C., and Singh, S. W., *Ind. J. Med. Res.*, **55**, 629(1967).
(3) Chopra, R. N., Nayar, S. L., and Chopra, I. C., "Glossary of Indian Medicinal Plants," C.S.I.R., New Delhi, India, 1956, p. 83.
(4) Nadkarni, A. K., and Nadkarni, K. M., "Indian Materia Medica," Dhootpapeswar Prakasha Ltd., Bombay, India, 1954, p. 407.
(5) Sumerwell, W. N., *J. Am. Chem. Soc.*, **79**, 3411(1957).
(6) Chowdhury, D. K., Chakrabarty, M. M., and Mukherji, B. K., *Science and Culture (India)*, **19**, 163(1953).
(7) Hilditch, T. P., "The Chemical Constitution of Natural Fats," 3rd. ed. Chapman and Hall Ltd., London, England, 1956, p. 611.
(8) Tewari, J. P., Dixit, B. B., and Mishra, S. S., *J. Pharm. Sci.*, **54**, 923(1965).

Keyphrases

Cucurbita maxima seed oil
Fatty acid fractionation
Countercurrent distribution with urea
Iodine values
Neutralization value
Saponification equivalents

Isolation of Candicine Chloride, Laurifoline Chloride, and Xanthoxyletin from the Bark of *Zanthoxylum elephantiasis*

By JOZEF TOMKO*, ALBERT T. AWAD†, JACK L. BEAL, and RAYMOND W. DOSKOTCH

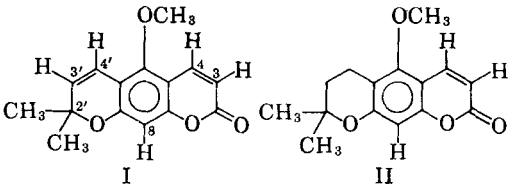
A study of the quaternary alkaloids of the bark of *Zanthoxylum elephantiasis* was made. Utilizing a silicic acid and diatomaceous earth (4:1) chromatographic column, the quaternary alkaloids laurifoline chloride and candicine chloride were separated and identified. In addition, xanthoxyletin was isolated from a petroleum ether extract of the bark. Its identification was made by physical and chemical data.

In a previous publication (1) the presence of quaternary bases in the bark of *Zanthoxylum elephantiasis* Macf. was reported. From the mixture of quaternary bases, only laurifoline chloride was isolated in small quantity by a rather unsatisfactory procedure. An improved procedure for the separation of laurifoline chloride from the mixture and, in addition, the isolation and identification of candicine chloride from the same mixture are reported here.

The petroleum ether extract of the powdered bark when chromatographed on acid alumina with benzene as eluting agent yielded xanthoxyletin (I) previously reported in a species of *Zanthoxylum* (2). Its identity was established by comparision of its physical properties with those reported in the literature and the preparation of the dihydroxanthoxyletin (II).

The NMR spectra of xanthoxyletin is consistent with its structure showing a singlet at δ = 1.47 (6H) for the two methyl groups at C-2′, a singlet at 3.88 (3H) for the methoxy group, and one at 6.52 (1H) for the aromatic proton at C-8. The protons at C-3 and C-4 appeared as a pair of doublets at δ = 6.20 and 7.87 (J = 9.5 c.p.s.), respectively, and those at C-3′ and C-4′ also as a pair of doublets were found at δ = 5.73 and 6.61 (J = 10 c.p.s.), respectively.

In dihydroxanthoxyletin the singlet for the two methyl groups at C-2′ was shifted upfield to δ = 1.38 and the pair of doublets for C-3′ and C-4′ were lost and now appeared as a pair of triplets centered at δ = 1.82 (2H) and 2.79 (2H), respectively. The other peaks of the spectrum remained essentially unchanged.



EXPERIMENTAL[1]

Separation of Candicine Chloride and Laurifoline Chloride—A quantity of 550 mg. of the mixture of quaternary alkaloid chloride salts obtained by chromatography on alumina as described in the previous publication (1) was dissolved in methanol and adsorbed on 5 Gm. of silicic acid and diatoma-

Received July 13, 1967 from the College of Pharmacy Ohio State University, Columbus, OH 43210
Accepted for publication September 22, 1967.
This investigation was supported by research grant GM-05640 from the National Institutes of Health, U. S. Public Health Service, Bethesda, Md.

* Present address: Institute of Chemistry of the Slovak Academy of Sciences, Bratislava, Czechoslovakia.
† Present address: College of Pharmacy, Ohio Northern University, Ada, OH 45810

[1] The melting points are uncorrected and were determined using a Thomas-Hoover Unit-Melt capillary melting point apparatus. The infrared spectra were taken in KBr pellets using a Perkin-Elmer model 237 infrared spectrophotometer. The ultraviolet spectra were determined using a Cary model 15 spectrophotometer. Optical rotations were measured on a Zeiss-Winkel polarimeter in a 2-decimeter polarographic tube. The NMR spectra were determined on a Varian A-60 spectrometer in deuterochloroform with tetramethylsilane as internal standard. Chemical shifts are reported in δ (p.p.m.) values. Thin-layer chromatography utilized Silica Gel G (Merck) as the adsorbent and the following solvent systems: *1*, methanol–water–ammonium hydroxide solution (27%) (2:1:1), and *2*, methanol–water–ammonium hydroxide solution (27%) (1:2:2). Dragendorff's test solution was used for the detection of the alkaloids. Silicic acid was obtained from Mallinckrodt Corp., St. Louis, Mo.