determine the antifertility effects of 5-[4-(substituted thiocarbamido)-phenoxymethyl]-2-oxazolidinethiones is warranted.

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Cytotoxic Agents from *Bursera klugii* (Burseraceae) I: Isolation of Sapelins A and B

S. D. JOLAD, R. M. WIEDHOPF, and J. R. COLE ˣ

Abstract □ A crude chloroform-soluble fraction of the ethanol extract of the leaves of *Bursera klugii* showed activity against two test systems, the P-388 lymphocytic leukemia (3PS) and the human epidermoid carcinoma of the nasopharynx (9KB). The PS activity was due to two constituents, sapelins A and B.

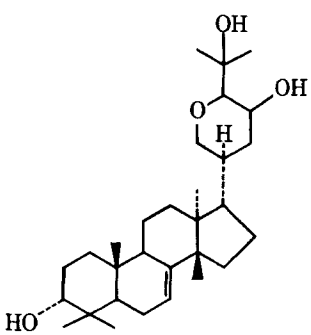
Keyphrases □ Sapelins A and B—isolated from ethanol extract of leaves of *Bursera klugii*, cytotoxic activity evaluated □ *Bursera klugii*—sapelins A and B isolated from ethanol extract of leaves, cytotoxic activity evaluated □ Cytotoxic activity—sapelins A and B evaluated

During a continuing search for plants having tumor inhibitory constituents, the chloroform-soluble fraction of the ethanol extract of the leaves of *Bursera klugii* Macbr (Burseraceae[1]) was shown to be active against the P-388 lymphocytic leukemia test system (3PS) and the human epidermoid carcinoma of the nasopharynx test system[2] (9KB). The activity against the PS test system was due to two constituents of the crude chloroform-soluble fraction.

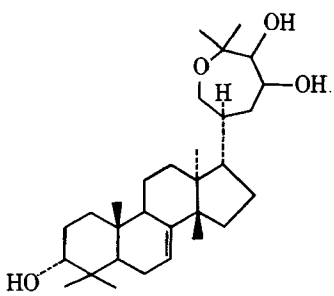
DISCUSSION

The two major constituents of the chloroform-soluble fraction were triterpenes, sapelin A (I) and sapelin B (II). These compounds were isolated previously from commercial sapele, *Entandrophragma cylindricum* Sprague (1). Isolation of these triterpenes was effected by solvent extraction followed by column chromatography and crystallization. Identification was made by elemental analysis; IR, NMR, and mass spectral data; and direct comparison with an authentic specimen.

Sapelin A demonstrated activities of 130, 136, 127, and 130% test/control (T/C) at 10.0, 5.0, 2.5, and 1.25 mg/kg, respectively. Sapelin B demonstrated activities of 136, 136, 130, and 138% T/C at the same doses, respectively. Activity in the PS system is defined as an increase in the survival of treated animals over that of control animals resulting in a T/C ≥ 125% (2).



I



II

EXPERIMENTAL[3]

The leaves of *B. klugii*, collected in Peru during August 1975, were ground and stored at −10° prior to extraction.

The ground material (7.72 kg) was extracted exhaustively in a Lloyd-type extractor with petroleum ether followed by 95% ethanol. The air-dried ethanol extract was subjected to partitioning between chloroform and water. The chloroform phase was first air dried and then vacuum dried. A 300-g portion of the residue was extracted with 5 liters of *n*-hexane in three portions by mechanical stirring. The hexane-insoluble residue was then extracted with 5 liters of ether in three portions using a magnetic stirrer.

The combined ether-soluble fraction, on concentration under vacuum followed by cooling in a freezer overnight, yielded a residue. TLC of this residue showed two spots as major constituents, one corresponding to sapelin A and the other to sapelin B. A 4-g portion of this residue was subjected to alumina (grade III, 160 g) dry column chromatography. Elution with ethyl acetate–benzene (1:1) afforded nearly pure sapelins

[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.

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

[3] Carbon and hydrogen analyses were carried out by Chemalytics, Inc., Tempe, Ariz. Melting points were determined on a Kofler hot-stage apparatus and are uncorrected. IR spectra were run on a Beckman IR-33. NMR and mass spectra were run using a Varian T-60 spectrometer and a Hewlett-Packard quadrupole spectrometer (model 5930), respectively.

A and B, which were further purified by decolorization followed by crystallization.

Sapelin A was obtained as long colorless thick needles from methanol, mp 219–220° with initial sweating. It was identical in all respects with the authentic specimen[3].

Anal.—Calc. for $C_{30}H_{50}O_4$: C, 75.9; H, 10.6; mol. wt. 474. Found: C, 75.31; H, 10.10; *m/e* 474 (M^+).

Sapelin B was obtained as tiny colorless needles from ethyl acetate–petroleum ether, mp 174–176° with initial sweating. It was identical in all respects with the authentic specimen.

Anal.—Calc. for $C_{30}H_{50}O_4$: C, 75.9; H, 10.6; mol. wt. 474. Found: C, 75.32; H, 10.13; *m/e* 474 (M^+).

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[x] To whom inquiries should be directed.

Quinuclidine Chemistry: Autocondensation Reactions of 3-Quinuclidinone

R. A. SANDMANN [x] and W. C. McHUGH, Jr.

Abstract □ During the synthesis of 3-hydroxy-3-ethinylquinuclidine (I), two additional products were isolated and identified as (*E*)-3-[2-(3-oxoquinuclidine)]quinuclidylidene (III) and (*E*)-3-[2-(3-hydroxy-3-ethinylquinuclidine)]quinuclidylidene (V). The base-catalyzed autocondensation of 3-quinuclidinone resulted in the α,β-unsaturated ketone dimer (III) as a single isomer. The geometric configuration was deduced by examination of the NMR spectra of the methyl iodide salt. Compound V was thus the result of attack on the carbonyl carbon of III by the acetylide anion. The isolation and identification of these compounds clarified the reported differences in the physical properties of I and its analogs.

Keyphrases □ Quinuclidines, substituted—synthesized, structures, stereoisomers, and physical properties identified □ Autocondensation—3-quinuclidinone, products identified

It was necessary to synthesize significant quantities of 3-hydroxy-3-ethinylquinuclidine (I) as an intermediate in preparing a series of potential antidepressant agents. However, there was a disparity in the physical properties of I prepared by two different methods (1, 2). The synthetic method employed by Ernest (2) gave a compound melting at 192°, and that of Clemo and Hoggarth (1) gave a compound melting at 159°.

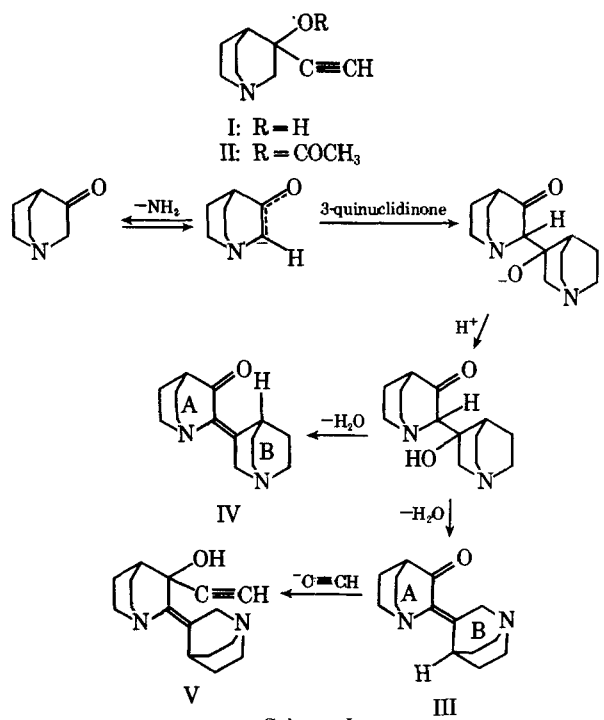
A clarification of these discrepancies by the synthesis of I and (*E*)-3-[2-(3-oxoquinuclidine)]quinuclidylidene (III) (mp 159°) is now reported.

DISCUSSION

The preparation of I involved dispersing acetylene gas through a tetrahydrofuran solution of 3-quinuclidinone and sodium amide. Three products were isolated, the major compound being I and having the same characteristics as demonstrated by Ernest (2). Mass spectra of the remaining two compounds indicated masses of 258 and 232. The difference in masses was 26, the molecular weight of acetylene. IR analysis of the 258 compound indicated a terminal acetylene (ν_{max} at 3240 and 2095 cm^{-1}). The IR spectrum for the 232 compound showed an α,β-unsaturated ketone (ν_{max} 1685 and 1615 cm^{-1}).

3-Quinuclidinone condenses with aldehydes under basic conditions (3–5); autocondensation under similar conditions and by the same aldol-type mechanism can now be demonstrated (Scheme I). A base such as sodium amide generates the enolate, which condenses by nucleophilic attack with another molecule of 3-quinuclidinone. The resultant dimer alkoxide is protonated, and dehydration of the ketol should result in a mixture of α,β-unsaturated ketone isomers, III and IV, its (*Z*)-isomer.

Conventional synthesis of III employed the use of potassium *tert*-butoxide in *tert*-butyl alcohol as the condensing agent for 3-quinuclidinone. The product (mol. wt. 232) proved to be identical with the one isolated from the acetylene–sodium amide reaction. Thus, the reaction between III and the acetylide ion gave (*E*)-3-[2-(3-hydroxy-3-ethinylquinuclidine)]quinuclidylidene (V, mol. wt. 258).



Scheme I