

Lawsone Derivatives II

Sulfonium Mustard Precursors and 1-Naphthylisocyanate Complexes

By H. MACHATZKE and W. R. VAUGHAN

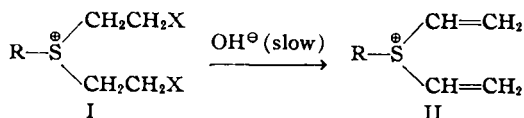
An approach to synthesis of derivatives of 2-hydroxy-1,4-naphthoquinone (lawsone) bearing the bis- β -chloroethylsulfonium group in a 3-alkyl side chain is described along with incidental formation of a related hemi sulfur mustard. In addition, the unexpected formation of what appear to be charge-transfer complexes of lawsone derivatives with 1-naphthylisocyanate is reported for the first time. The free 2-hydroxyl of lawsone appears to be essential for complexation, since in its absence insoluble complexes do not form. The presence of a 3-alkyl substituent also appears to be necessary since lawsone itself does not afford an insoluble complex.

ALTHOUGH IT IS not the purpose of this paper to report on or suggest the existence of significant physiological properties in the various compounds described but rather to delineate problems encountered in the course of a specific type of synthesis which may be of use in other contexts, it nevertheless seems appropriate to describe the rationale for undertaking the synthesis considered here.

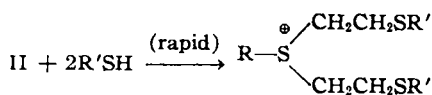
A previous paper concerned with the potential use of lawsone derivatives bearing "alkylating" functions in a 3-alkyl side chain, pointed out that initial attempts afforded only very unstable products owing to facile reversal of the Mannich reaction whereby they were produced. Hence, it was desirable to prepare higher homologs wherein the alkylating group was farther removed from the quinone nucleus. To this end the preparation of ω -substituted 3-polymethylene lawsones was investigated with the replacement of the terminal substituent by the bis- β -chloroethylamino group as the objective. However, the chemistry of such compounds proved to be unexpectedly complex owing to a surprising tendency for even decamethylene groups with terminal ester or halogen substituents to cyclize both inter- and intramolecularly under the influence of diethanolamine (1).

Since these anomalous reactions appear to depend upon the basic character of diethanolamine, attention was directed toward introduction of the sulfonium mustard system. Sulfonium salts of structure I are known to undergo a characteristic elimination in a basic medium (Scheme I) (2).

The reactivity of bis-vinyl sulfonium ions such as II toward nucleophiles, especially mercaptans at physiological pH (Scheme II) has been demonstrated (3), and sulfonium salts such as I ($X = \text{bromine or chlorine}$) have been shown as a consequence to be



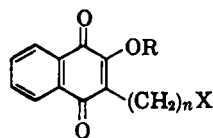
Scheme I



Scheme II

cytostatically active (3-5). Thus, synthesis of a number of compounds of type I, wherein $\text{R} = (2\text{-hydroxy-1,4-naphthoquinon-3-yl})\text{polymethylenyl}$, was undertaken.

In each case a 3- ω -halopolymethylenelawsone (III) was used to alkylate bis- β -hydroxyethylsulfide (IV) in refluxing ethanol.

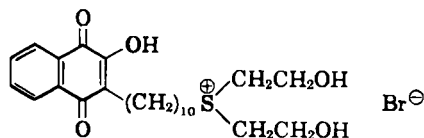


III

- (a) $\text{R} = \text{H}$, $n = 3$, $\text{X} = \text{Cl}$
- (b) $\text{R} = \text{CH}_3$, $n = 3$, $\text{X} = \text{Cl}$
- (c) $\text{R} = \text{H}$, $n = 10$, $\text{X} = \text{Br}$

The reaction of IIIa (6) with IV was abortive, with only starting materials being recovered, and the reaction of IIIb afforded cyclized materials (6) as in the diethanolamine reactions cited above. Consequently, attention was focussed on IIIc (7) as the alkylating agent for IV.

This reaction was successful; 60% of the desired product, bis- β -hydroxyethyl-10(2-hydroxy-1,4-naphthoquinon-3-yl) decylsulfonium bromide (V), was obtained.

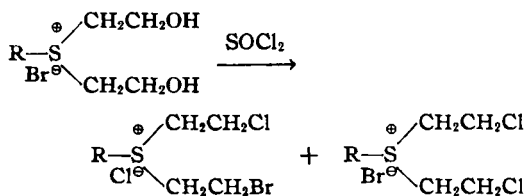


In addition, a by-product, 2-hydroxy-3(10-hydroxydecyl)-1,4-naphthoquinone (VI), was obtained, presumably from V by an S_N reaction with adequate precedence (8).

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Scheme III

Since previous work (9) had shown that thionyl chloride reacts with compounds such as I ($\text{R} = \text{OH}$, anion = Br^{\ominus}) with appreciable exchange of ionic bromide and covalent chloride in the product (Scheme III) and since V may well exist, at least in part, as the thetine and hydrogen bromide owing to the pronounced acidity of the 2-hydroxyl of the lawsone system, a model compound, bis- β -hydroxyethyl-10-carboxydecylsulfonium bromide (VII), was prepared for study by alkylating IV with 10-bromoundecanoic acid (10), a reaction which produced tris- β -hydroxyethylsulfonium bromide as a by-product. Treatment of VII with thionyl chloride at 0–5° in chloroform, followed by treatment of the resultant colorless oil with perchloric acid afforded an oily product (VIII) whose composition most nearly corresponds to that of a 3:1 mixture of the thetine, β -bromoethyl- β -chloroethyldecylsulfonium-10-carboxylate and its hydroperchlorate.

This result was unpromising for a similar conversion of V, but since the reaction represented by Scheme I may also involve $\text{X} = \text{OCOCH}_3$, conversion of VII to the diacetyl fluoborate (IX) and bromide (X) by treatment with boron trifluoride etherate in acetic anhydride (11) (for IX) followed by ethereal hydrogen bromide addition to methanolic IX (for X) was carried out. Unlike the colorless crystalline IX, bis- β -acetoxyethyl-10(2-hydroxy-1,4-naphthoquinon-3-yl)decylsulfonium fluoborate prepared analogously from V, was a brown-yellow oil.

The reaction of V with thionyl chloride proved as complex as expected, and in only one instance was a readily characterizable unique product obtained; at 60° the expected mixed bis- β -haloethylsulfonium halide (XII) decomposed to give a small quantity of β -chloroethyl-10(2-hydroxy-1,4-naph-

thoquinon-3-yl)decyl sulfide (XIII). At the recommended lower reaction temperature (4) XII showed considerable instability; hence conversion to a perchlorate was attempted (XIV). The infrared spectrum of XIV does not show the characteristic hydroxyl of the lawsone system, but the microanalysis is not far out of line for $\text{C}_{24}\text{H}_{32}\text{Cl}_2\text{O}_5\text{S}\cdot\text{HClO}_4$.

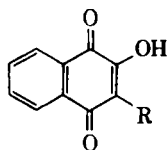
Since the initial reaction product of V with thionyl chloride was too unstable to permit analysis, an attempt was made to effect reaction of its 2-hydroxyl group with 1-naphthylisocyanate (XV) to provide a more tractable and crystalline product. However, on treatment of XII with XV in chloroform, another unstable oil was obtained (XVI). Examination of the infrared spectrum of XVI showed a strong band at 2260 cm^{-1} due to isocyanate as well as a broad band at 3220 cm^{-1} , neither of which could be removed by repeated treatment with chloroform. Furthermore, the carbonyl band at 1640 cm^{-1} was appreciably less intense than in the spectrum of XII, and there was a slightly greater than 10% increase in absorbance for the ultraviolet band at 3270 Å.

These observations led us to suspect the formation of a complex with XV rather than urethane formation. And since both XII and XIV appeared to be too unstable for satisfactory study, V (the precursor of XII) was investigated. Similar treatment with XV afforded a yellow crystalline precipitate (XVII) as did IIIc (XVIII) and 6-(2-hydroxy-1,4-naphthoquinon-3-yl)hexanoic acid (XIX) which afforded XX.

The acetyl derivatives of XIX (XXI) as well as lawsone itself, 2-chloro-1,4-naphthoquinone, and 2-methoxy-3(3-chloropropyl)-1,4-naphthoquinone do not afford insoluble complexes with XV under similar conditions.

Analyses indicate that the complexes (XVII, XVIII, XX) comprise two molecules of the lawsone moiety and one of 1-naphthylisocyanate. Each of them shows the characteristic isocyanate band (2260 cm^{-1}) in the infrared spectrum in addition to one hydroxyl band, which is shifted from 3310–3315 cm^{-1} (sharp) to lower frequency and broadened (3220 cm^{-1}). Table I presents the significant infrared data, and except for the bands listed, the spectra are essentially unchanged from

TABLE I.—PRINCIPAL INFRARED BANDS IN FREE LAWSONE DERIVATIVES AND THEIR 1-NAPHTHYLISOCYANATE COMPLEXES



R		OH (cm^{-1})	C=O (cm^{-1})		—N=C=O (cm^{-1})
			α	β	
$(\text{CH}_2)_6\text{CO}_2\text{H}$	Free	3310 (Sharp)	1660 (sh.)	1640	2260
	Complex	3220 (Broad)	1670	1645 ^b	2260
$(\text{CH}_2)_{10}\text{Br}$	Free	3315 (Sharp)	1660 (sh.)	1645	...
	Complex	3220 (Broad)	1670	1645 ^b	2260
$(\text{CH}_2)_{10}\text{S}^{\oplus}(\text{CH}_2\text{CH}_2\text{OH})_2\text{Br}^{\ominus}$	Free	3220 ^a	1670	1640	...
	Complex	...	1670	1640 ^b	2260
1-Naphthylisocyanate	2260

^a The shift of the hydroxyl frequency is not visible because of the additional hydroxyl groups in the side chain. ^b Less intense than in the free quinone.

those of the constituent parts (12) which are characteristic for the quinone system and the 1-naphthylisocyanate.

Infrared studies have shown two carbonyl bands: one (α) "normal" at about 1670 cm^{-1} , and one shifted and weakened due to hydrogen bonding (β) (13-15). In the lawsone complexes a decrease in absorption band β and an increase in absorption band α is apparent. Consequently, it would appear that there may be less hydrogen bonding to the carbonyl groups of the quinone moieties, in spite of an increase in total hydrogen bonding shown by the more pronounced shift in the hydroxyl absorption. Failure of lawsone derivatives lacking the free hydroxyl to form complexes suggests that this hydrogen bonding is essential but not necessarily sufficient for complex formation.

The visible-ultraviolet spectra of the complexes compared with those of the quinone constituents show changes characteristic of charge-transfer or π -complexes (16). In acetone solution two of the complexes show a strong absorption at 3270 Å, compared to absorption at about 3310 Å for the free quinones, with marked increase in the absorbances of the former. Pertinent data are collected in Table II.

Whether the complexes are of a "sandwich" type with one lawsone derivative bonded to the isocyanate nitrogen by a hydrogen bridge and the other to the opposite face of the isocyanate by charge-transfer attraction cannot be stated with the evidence at hand; but the possibility is attractive and accounts for the spectrographic phenomena as well as for the failure to detect complexes where the lawsone hydroxyl is masked. The role of the 3-substituent is not clear, but failure to obtain a complex with lawsone itself implies a contributory factor which is not associated with the ability of the system to hydrogen bond, since all of the lawsone derivatives examined are necessarily weaker acids than lawsone itself. It should be noted that whatever the structure of the complex may prove to be, the charge-transfer character has but a single precedent in which the quinone is the electron donor; phloroglucinol with two benzoquinones (17).

EXPERIMENTAL¹

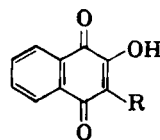
2 - Methoxy - 3 - (3 - chloropropyl) - 1,4 - naphthoquinone (IIIb).—The diacyl peroxide of 4-chlorobutyric acid was obtained in 46% yield by the hydrogen peroxide method (18). The alkylation of lawsone was effected by the "flash-off" method (18). Fractional crystallization from methanol or methanol-water afforded the quinone (IIIb) in 45% yield (based on recovered lawsone), m.p. 67-68°.

Anal.—Calcd. for $\text{C}_{14}\text{H}_{13}\text{ClO}_3$: C, 63.50; H, 4.95; Cl, 13.40. Found: C, 63.72; H, 4.91; Cl, 13.29.

Bis - β - hydroxyethyl - 10(2 - hydroxy - 1,4-naphthoquinon-3-yl)decylsulfonium Bromide (V).—A solution of 5.1 Gm. (0.013 mole) of 2-hydroxy-3(10 - bromodecyl) - 1,4 - naphthoquinone (IIIc)

¹ Melting points are uncorrected; Kofler block indicated by superscript *k* where used. Microanalyses by Spang Microanalytical Laboratory, Ann Arbor, Mich. Infrared spectra of Nujol mulls obtained with Perkin-Elmer model 21 double beam recording spectrometer. Ultraviolet spectra obtained with Cary model II double beam spectrometer. The authors are indebted to Mr. B. E. Wenzel for preparing certain of these spectra.

TABLE II.—SIGNIFICANT ULTRAVIOLET ABSORPTION BANDS IN FREE LAWSONE DERIVATIVES AND THEIR 1-NAPHTHYLSOCYANATE COMPLEXES IN ACETONE SOLUTION



R	Free Quinone		Complex	
	λ max.	ϵ^a	λ max.	ϵ^a
$(\text{CH}_2)_6\text{CO}_2\text{H}$	3310	2800	3270	5900
$(\text{CH}_2)_{10}\text{Br}$	3310	2900	3270	5400
$(\text{CH}_2)_{10}\text{S}^{\oplus}$ $(\text{CH}_2\text{CH}_2\text{OH})_2\text{Br}^{\ominus}$	3270	3000	3270	3400

^a Molecular absorbance. Complex calculated for two quinones plus one isocyanate.

(8) and 6.0 Gm. (0.05 mole) of bis- β -hydroxyethyl sulfide (IV) in 50 ml. of 90% ethanol was refluxed for 2 days. Next the solvent was removed on the steam bath and ether was added; the resultant oily precipitate was repeatedly recrystallized by solution in methanol or methanol-acetone followed by addition of ether. The product was a yellow, crystalline substance which dissolves in water to give a red solution; 3.5 Gm. (53%), m.p. 87-88°^k.

Anal.—Calcd. for $\text{C}_{24}\text{H}_{36}\text{BrO}_5\text{S}$: C, 56.00; H, 6.83; Br, 15.50; S, 6.23. Found: C, 55.74; H, 6.84; Br, 16.05; S, 6.45.

2 - Hydroxy - 3(10 - hydroxydecyl) - 1,4 - naphthoquinone (VI).—The ether-soluble residues from several preparations of V were triturated with petroleum ether (40-60°), and upon evaporation of the solvent there remained a yellow crystalline product which was recrystallized once from methanol and twice from acetone, m.p. 80-81°^k. Approximately 10-20% (based on IIIc) was recovered in this manner.

Anal.—Calcd. for $\text{C}_{20}\text{H}_{24}\text{O}_4$: C, 72.70; H, 7.93. Found: C, 72.80; H, 8.26.

Bis - β - Hydroxyethyl - 10 - carboxydecylsulfonium Bromide (VII).—A solution of 7.0 Gm. (0.026 mole) of 10-bromoundecanoic acid (10) and 3.7 Gm. (0.03 mole) of bis- β -hydroxyethyl sulfide (IV) in 50 ml. of 80% ethanol was refluxed for 3 days. The solvent was then removed *in vacuo* and the oily residue taken up in methanol. Upon addition of ether there precipitated white crystals which were recrystallized three times from methanol to which ether was added to effect reprecipitation; 4.5 Gm. (45%), m.p. 110-111°^k.

Anal.—Calcd. for $\text{C}_{18}\text{H}_{31}\text{BrO}_5\text{S}$: C, 46.60; H, 8.08. Found: C, 46.87; H, 8.11.

tris - β - Hydroxyethylsulfonium Bromide.—When a large excess of IV was used in the preceding preparation, the product was rather difficult to purify and under these circumstances fractional recrystallization afforded *tris*- β -hydroxyethylsulfonium bromide (19) in substantial amounts, m.p. 96-97°^k.

Anal.—Calcd. for $\text{C}_6\text{H}_{15}\text{BrO}_3\text{S}$: C, 29.20; H, 6.13; Br, 32.40; S, 12.93. Found: C, 29.70; H, 6.20; Br, 31.73; S, 12.98.

Reaction of VII with Thionyl Chloride Followed with Perchloric Acid (VIII).—To a solution of 3.9

TABLE III.—ANALYTICAL DATA FOR 1-NAPHTHYLISOCYANATE COMPLEXES

Complex No.	M.p., °C.	Quinone	Compn. (2Q·XV)		Anal.				
					C	H	Br	N	S
XVI	Oil	XII							
XVII	106°	V	C ₁₉ H ₁₇ Br ₂ NO ₁₁ S ₂	Calcd.	59.10	6.47	13.20	1.17	5.35
				Found	59.02	6.50	13.30	1.40	5.44
XVIII	70°	IIIc	C ₂₁ H ₂₇ Br ₂ NO ₇	Calcd.	64.10	6.02	8.40	1.46	...
				Found	64.01	6.18	8.69	1.40	...
XX	117°	XIX	C ₂₂ H ₂₉ NO ₁₁	Calcd.	69.20	5.27	...	1.88	...
				Found	69.50	5.54	...	1.74	...

Gm. (0.01 mole) of VII in 50 ml. of chloroform at 0° (ice-bath) was added slowly and with magnetic stirring 6.0 Gm. (0.05 mole) of purified thionyl chloride. After 2 hours the reaction was allowed to come to room temperature and stirring was continued for 3 additional hours. Addition of dry ether caused a colorless oil to separate, but the latter could not be crystallized. Consequently, an attempt was made to convert it to the perchlorate by solution in water and dropwise addition of perchloric acid until no further precipitate was formed. The resultant oil was dried *in vacuo* for analysis (2.5 Gm.).

Anal.—Calcd. for C₁₈H₂₃BrClO₅S (A) (thetine): C, 46.40; H, 7.27; Hal (Cl), 18.30; S, 8.25.

Calcd. for C₁₈H₂₃BrClO₅S (B) (thetine hydroperchlorate): C, 37.00; H, 6.00; Hal (Cl), 21.80; S, 6.57.

Calcd. for 3A + 1B: C, 43.50; H, 6.90; Hal (Cl), 19.40; S, 7.75. Found: C, 43.10; H, 7.12; Hal (Cl), 20.80; S, 7.40.

Bis - acetoxyethyl - 10 - carboxydecylsulfonium Fluoborate (IX).—A 0.01-mole sample of VII in 10 ml. of acetic anhydride was treated dropwise with 4 ml. of redistilled boron trifluoride etherate (11) and then allowed to stand at room temperature for 24 hours. Addition of dry ether afforded a colorless crystalline substance which was recrystallized from absolute methanol, precipitation being effected by addition of dry ether; m.p. 85–86°, yield 62%.

Anal.—Calcd. for C₁₉H₂₅BF₄O₈S: C, 48.60; H, 7.55; S, 6.85. Found: C, 48.38; H, 7.45; S, 6.56.

Bis - β - acetoxyethyl - 10 - carboxydecylsulfonium Bromide (X).—A 2-Gm. sample of IX was dissolved in 5 ml. of absolute methanol, and a dry ether solution of hydrogen bromide was added. The treatment was repeated, after which the product was repeatedly washed with dry ether, then dried *in vacuo* over both phosphorus pentoxide and sodium hydroxide; 76%, m.p. 85–86°.

Anal.—Calcd. for C₁₉H₂₅BrO₈S: C, 48.40; H, 7.50; Br, 16.80. Found: C, 48.51; H, 7.63; Br, 16.40.

Bis - β - acetoxyethyl - 10(2 - acetoxy - 1,4-naphthoquinon - 3 - yl)decylsulfonium Fluoborate (XI).—A 2.5-Gm. (0.01 mole) sample of V was treated in acetic acid with redistilled boron trifluoride etherate as for VII. The solution became red, but addition of the dry ether produced a brown-yellow oil which was "recrystallized" as for IX; 2.5 Gm. (92%).

Anal.—Calcd. for C₂₀H₂₁BF₄O₈S: C, 55.50; H, 6.38; S, 4.96. Found: C, 55.32; H, 6.33; S, 4.96.

Reaction of V with Thionyl Chloride (XII).—

A 2.6-Gm. sample of analytically pure V in 20 ml. of chloroform was cooled in an ice-bath and treated dropwise with purified thionyl chloride while being magnetically stirred. Subsequent work-up was the same as for VIII. The product, a yellow oil, was reprecipitated three times from absolute methanol by addition of dry ether, but upon drying *in vacuo* it was apparent that decomposition was setting in. Consequently, conversion to the perchlorate (XIV below) was attempted. Infrared and ultraviolet spectra were obtained on freshly prepared samples.

β - Chloroethyl - 10 - (2 - hydroxy - 1,4 - naphthoquinon - 3 - yl)decylsulfide (XIII).—In a similar reaction to the preceding the temperature was allowed to reach 60°. The product was extracted with petroleum ether and the extract then recrystallized three times from petroleum ether to give yellow crystals, m.p. 64–65°.

Anal.—Calcd. for C₂₂H₂₉ClO₅S: C, 64.50; H, 7.13; Cl, 8.68; S, 7.82. Found: C, 64.63; H, 7.03; Cl, 8.96; S, 7.56.

The infrared spectrum showed the characteristic hydroxyl band of the lawsone system.

Reaction of XII with Perchloric Acid (XIV).—The brown-yellow oil (XII, above) was dissolved in 20 ml. of water and treated dropwise with concentrated perchloric acid until no further precipitation occurred. After refrigeration the water was decanted and the oily residue washed twice with distilled water and then twice "recrystallized" from methanol-ether. The resultant very viscous oil was dried *in vacuo* over phosphorus pentoxide.

Anal.—Found: C, 49.40; H, 5.60; Hal (Cl), 22.30; S, 6.25.

Ethyl 6 - (2 - Hydroxy - 1,4 - naphthoquinon - 3 - yl)hexanoate (XIX Ethyl Ester).—This compound was prepared from lawsone and the peroxide obtained from the acid chloride of ethyl hydrogen pimelate by the Fieser procedure (20). The product was taken up in ether and washed with potassium bicarbonate until the extracts were nearly colorless; recrystallized from petroleum ether (90–100°), 69%, m.p. 72–74°.

Anal.—Calcd. for C₁₈H₂₀O₅: C, 68.40; H, 6.33. Found: C, 68.34; H, 6.24.

6 - (2 - Hydroxy - 1,4 - naphthoquinon - 3 - yl)hexanoic Acid (XIX).—A solution of 16 Gm. of the ethyl ester of XIX in 10 ml. of methanol was dropped into 300 ml. of a 30% aqueous potassium hydroxide solution containing 20 Gm. of sodium hydrosulfite (20), and the resultant solution was refluxed for 2 hours, then allowed to stand overnight at room temperature, after which it was acidified with hydrochloric acid and extracted with ether. The ether extract was then washed thoroughly with water and dried over magnesium sulfate. Evaporation afforded the quinone which recrystal-

lized from methanol-water (three times); 9.5 Gm. (65%), m.p. 113-114°.

Anal.—Calcd. for $C_{16}H_{16}O_5$: C, 66.65; H, 5.61. Found: C, 66.50; H, 5.80.

6-(2-Acetoxy-1,4-naphthoquinon-3-yl)-hexanoic Acid (XXI).—The boron trifluoride procedure described for IX and XI (11) was applied to 15 Gm. of XIX in 150 ml. of glacial acetic acid. After 2 hours at room temperature, water was carefully added and the yellow crystalline precipitate was recrystallized from methanol-water; 9.0 Gm. (53%), m.p. 85-86°.

Anal.—Calcd. for $C_{18}H_{18}O_6$: C, 65.50; H, 5.50. Found: C, 65.58; H, 5.31.

Complexes with 1-Naphthylisocyanate (XV): XVI, XVII, XVIII, XX.—The following procedure (XV + V) was also applied to XV and IIIc, XII, and XIX. A solution of 2 Gm. of V in 10 ml. of absolute chloroform was prepared by gentle warming, and 2-3 ml. of XV was added dropwise, whereupon yellow crystals immediately precipitated. Upon cooling the product was filtered off and an infrared spectrum taken. It was then washed 10 times with absolute ether and dried over phosphorus pentoxide. No change in the intensity of the 2260 cm^{-1} band in the spectrum was observed after washing; 1.8 Gm. (77%), m.p. 106°.

The same complex was obtained by refluxing a chloroform solution of V and XV for 5 hours. Neither the lawsone nor the primary alcohol groups in the side chain were affected.

When the complex was dissolved in absolute ethanol, the ethyl urethane was readily formed,

and the quinone moiety could be recovered by addition of ether to the reaction mixture.

Analytical data for the complexes are collected in Table III.

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Comparative Chromatographic Identification of the Alkaloids of Ten *Aquilegia* Species

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The results of a comparative study of the alkaloid constituents in the roots of ten species of *Aquilegia* are reported. Magnoflorine was present in all of the species, while in eight of the species berberine and aquilegenine were also present. A fourth alkaloid, unidentified, is believed to be present in the roots of *A. canadensis*.

THE GENUS *Aquilegia* is classified botanically among the *Ranunculaceae*. The *Ranunculaceae*, like the closely related (1) *Berberidaceae*, constitute a family among which a number of alkaloid-containing genera are found (2, 3). In this paper the results of a comparative study of the alkaloid constituents of ten species of *Aquilegia* are reported.

A literature search revealed a paucity of phytochemical information concerning the genus *Aquilegia*. Indeed, the only report of a

constituent analysis of an *Aquilegia* species dates from 1897, when De Rochebrune (4) described the isolation of an alkaloid, which he called aquilegine, from the seed of *A. vulgaris*.

Munz recognizes 67 species of *Aquilegia* (5) distributed throughout the world. Of these, there are approximately 24 species in North America.

It is readily apparent when examining monographs on *Aquilegia* (5, 6) that most species have several synonymous names which would lead one to believe that a larger number of species must have been considered to exist prior to the thorough study of the genus. *Aquilegias* are found throughout the entire United States and were considered at one time for the title of

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