

can be related to other physical-chemical data. The analysis indicates that the hydroxyl group contribution to the solubility is variable and depends significantly on the properties of the pure liquid alcohols. Aliphatic alcohols of varying structure are conveniently handled without the need to introduce additional parameters for branching, cyclization, etc. The method appears to be general enough to be extended to other functional groups as well as more complex organic molecules with limited solubility in water.

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Vanadyl Complexes of Antiamebic Drugs: Electronic Absorption Spectra

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Abstract □ The complexing properties of the antiamebic drugs iodochlorhydroxyquin and diiodohydroxyquin with vanadium as VO²⁺ were investigated using spectrophotometric methods. The composition of the complexes and their stability constants were determined by more than one procedure. When ethanol was the solvent and the pH of the solution was 2.0–6.0, the 1:2 complexes were predominant.

Keyphrases □ Iodochlorhydroxyquin—complexing properties, electronic absorption spectra of vanadyl complexes □ Diiodohydroxyquin—complexing properties, electronic absorption spectra of vanadyl complexes □ Vanadyl complexes—iodochlorhydroxyquin and diiodohydroxyquin, complexing properties investigated using electronic absorption spectra □ Complexes—vanadium with iodochlorhydroxyquin and diiodohydroxyquin, complexing properties investigated using electronic absorption spectra

Quinoline derivatives were among the earliest synthetic compounds found active in amebiasis. Greatest success was achieved with iodinated 8-hydroxyquino-

lines. Among the commonly used drugs are iodochlorhydroxyquin, chiniofon, and diiodohydroxyquin; the early work on these drugs was reviewed previously (1).

Testing of these drugs *in vitro* is complicated by their low solubility (2). Bradner and Rawson (3) reported that iodochlorhydroxyquin is active at 125 µg/ml in *Entameba histolytica*-*T. cruzi* cultures. It appears, therefore, that these drugs have a direct amebicidal activity of a relatively low order (3) and are effective to some degree when given orally to cyst-passing patients but less effective in acute amoebic dysentery. Iodochlorhydroxyquin and diiodohydroxyquin are effective only in intestinal amebiasis (4).

Quinoline derivatives are strong chelating agents. At pH 2.5, the 1:2 iron (III)-chiniofon was obtained with log *K_f* 7.5 (5). The same ligand forms different complexes with Co⁺², Ni⁺², Mo⁺⁴ (6, 7), and V⁺⁵

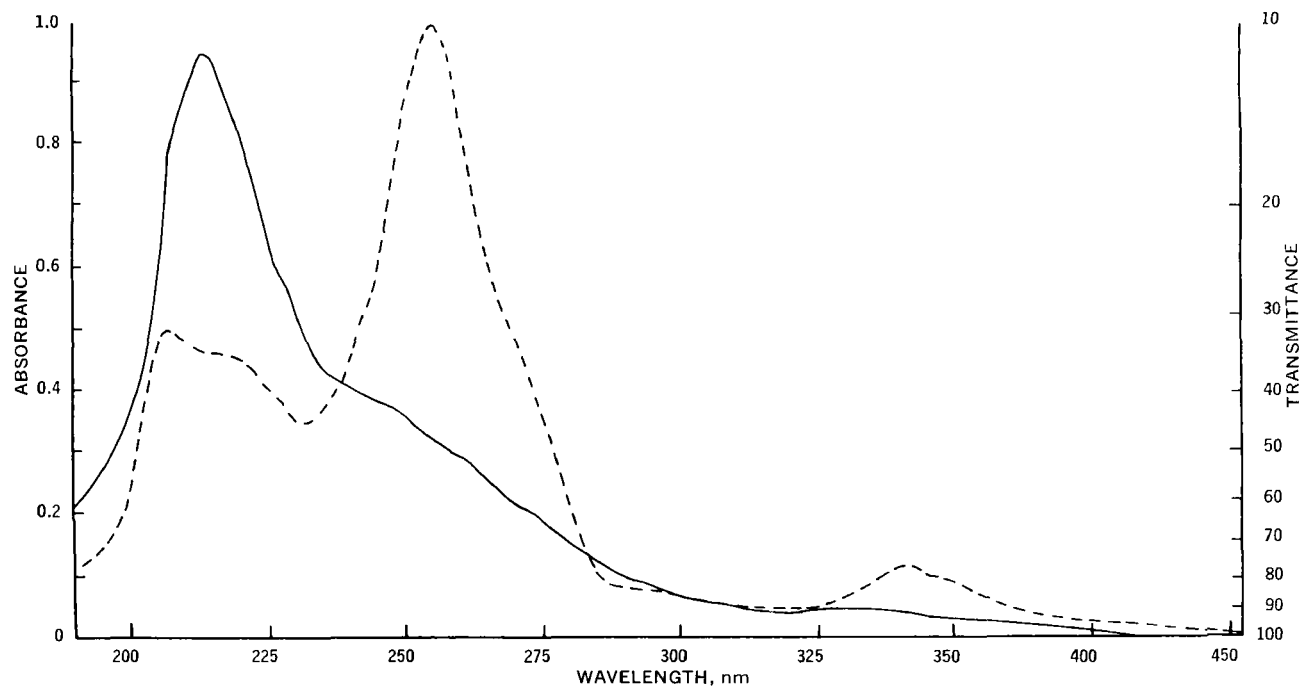


Figure 1—Electronic absorption spectra of vanadyl sulfate (—) and iodochlorhydroxyquin (---) in ethanol.

With Mo^{+4} , a yellow complex was obtained and extracted quantitatively with cyclohexane. Complexation of some substituted 8-hydroxyquinolines with transition metal ions has been investigated using different methods (8–15).

Vanadium as VO^{+2} was determined spectrophotometrically using ferron (chinoxon) (16). Vanadium ions are thought to have toxic effects, and 8-hydroxyquinolines have tuberculostatic, antibacterial, and fungitoxic effects. In this work the complexing properties of VO^{+2} with some 8-hydroxyquinolines were investigated using 96% ethanol as a solvent. Vanadyl quinolates may have tuberculostatic and fungitoxic activities as have copper quinolates (17, 18).

EXPERIMENTAL

Materials—Iodochlorhydroxyquin¹ (5-chloro-7-iodo-8-quinolinol) and diiodohydroxyquin¹ (5,7-diiodo-8-quinolinol) were purified by repetitive crystallization from purified ethanol. Vanadyl sulfate² solution was prepared in 96% ethanol² and standardized potentiometrically (19).

Apparatus and Method—Electronic absorption spectra were scanned³ using 1.0-cm fused silica cells. To determine the composition and stability constant of the complexes, solutions of the metal ion and ligands (in 96% ethanol) were mixed just before scanning the spectra.

In the slope ratio method, two sets of experiments were carried out. In the first, the concentration of VO^{+2} was kept constant at $1.0 \times 10^{-3} M$ and in excess of the iodochlorhydroxyquin concentration, which was varied from 4.0×10^{-5} to $1.0 \times 10^{-4} M$. In the second set the iodochlorhydroxyquin concentration was kept constant at $1.0 \times 10^{-3} M$ and in excess of that of vanadyl sulfate, which was varied from 4.0×10^{-5} to $1.0 \times 10^{-4} M$. The spectra of the two sets of solutions were scanned. In the molar ratio method, the [vanadyl]–[reagent] ratios varied from 1:1 to 1:10.

The acid dissociation constant of iodochlorhydroxyquin was de-

termined potentiometrically by titrating $2 \times 10^{-3} M$ solution (in ethanol) with $2 \times 10^{-2} M$ alcoholic potassium hydroxide (20).

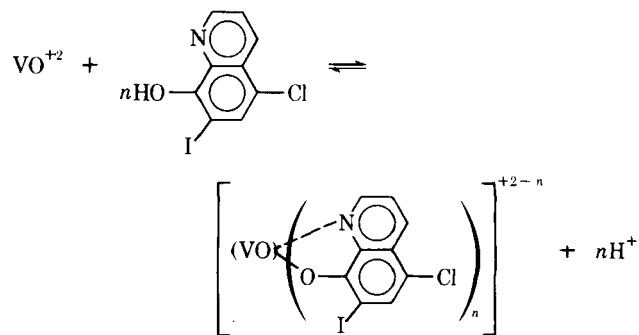
The electronic absorption spectra of the precipitated complexes were scanned in 96% ethanol and found to be similar to those obtained on mixing ligands–vanadyl sulfate solutions. IR spectra⁴ of the dried precipitated complexes were investigated.

The solution pH was adjusted using Britton (21) and Robinson modified universal buffer and was measured on a precision pH meter⁵, using the millivolt scale and calculating the corresponding pH values.

RESULTS AND DISCUSSION

Iodochlorhydroxyquin–Vanadium (IV) Complexes—*Absorption Spectra in Ethanol*—The spectrum of vanadyl sulfate (VOSO_4) (Fig. 1) shows a shoulder at ≈ 250 nm and an intense band at 215 nm. Iodochlorhydroxyquin shows no absorbance in the visible region (Fig. 1) but has some bands in the UV region (λ_{max} at 340, 255, and less than 220 nm). Dilute solutions of iodochlorhydroxyquin are pale yellow, and those of vanadyl sulfate are blue. When both are mixed, a yellow to red color is obtained and a new band appears in the spectra of the mixture (λ_{max} 410 nm) in addition to a long wavelength shoulder in the region of 500 nm (Fig. 2).

The reaction between iodochlorhydroxyquin and vanadyl ion possibly proceeds as shown in Scheme 1. To investigate the nature



Scheme 1

¹ RP grade reagents, obtained from Rhone-Poulenc.

² Prolabo grade reagent.

³ Unicam SP8000 spectrophotometer.

⁴ As KBr pellets with a Spektromom 2000.

⁵ Radelkis type OP-205.

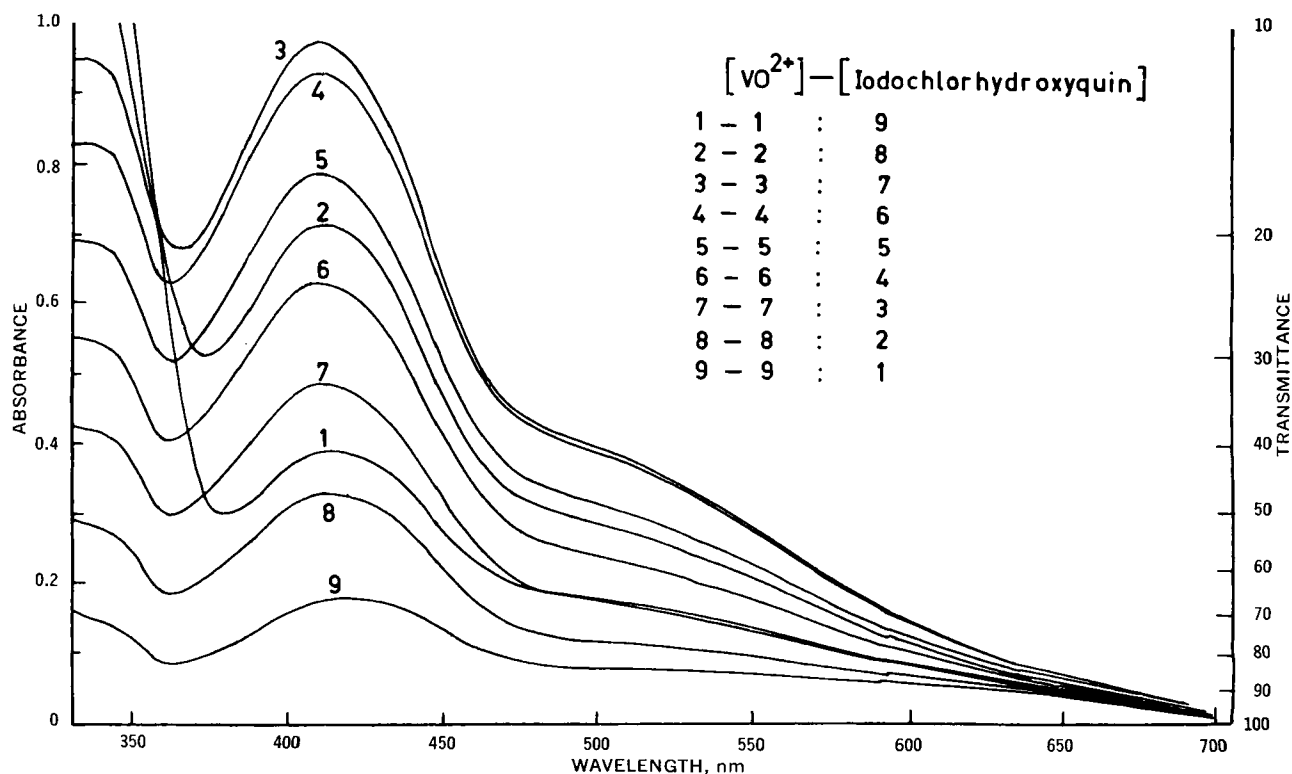


Figure 2—Electronic absorption spectra of vanadyl sulfate-iodochlorhydroxyquin solutions of constant total concentration.

of the chelate, Job's (22) method was followed. The spectra of vanadyl sulfate-iodochlorhydroxyquin solutions of total constant concentration were scanned (Fig. 2). The intensity of the 410-nm band was dependent on both vanadyl and ligand concentrations. The absorbance at 410 nm was plotted *versus* the mole fraction of the ligand (Fig. 3), and the results indicate that the predominant complex is the 1:2 vanadyl sulfate-iodochlorhydroxyquin chelate.

Job's (22) method also was carried out at different pH values (Fig. 4); a small shift of the complex band maximum and an in-

crease in its intensity appeared. The results (Fig. 4) indicate that the 1:2 complex is predominant within the pH range of 2.0–6.0.

To confirm these results, the Harvey and Manning (23) slope ratio method was used to determine the composition of the iodochlorhydroxyquin-vanadyl complex (Table I).

Plots of absorbance *versus* the concentration of the variable gave straight lines (Fig. 5a) with slope ratios of 1:2.1, indicating that the predominant complex at the concentrations used is 1:2. The mole ratio (24) method was also used to confirm the composi-

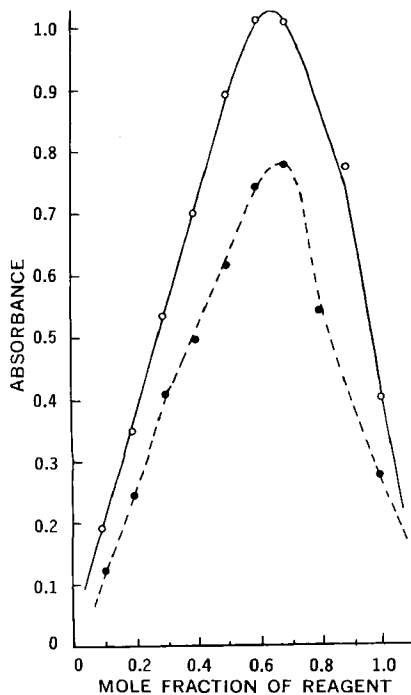


Figure 3—Application of the continuous variation method to vanadyl sulfate complexes. Key: O, reagent is diiodohydroxyquin; and ●, reagent is iodochlorhydroxyquin.

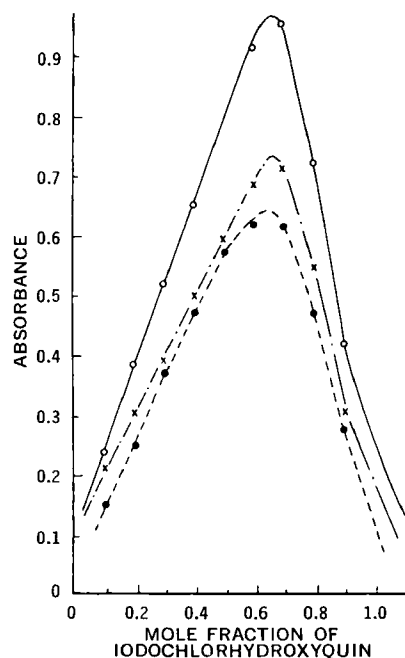


Figure 4—Application of the continuous variation method to vanadyl sulfate-iodochlorhydroxyquin solutions at different pH values. Key: O, pH 6.0, λ_{max} 395 nm; X, pH 4.1, λ_{max} 398 nm; and ●, pH 2.6, λ_{max} 400 nm.

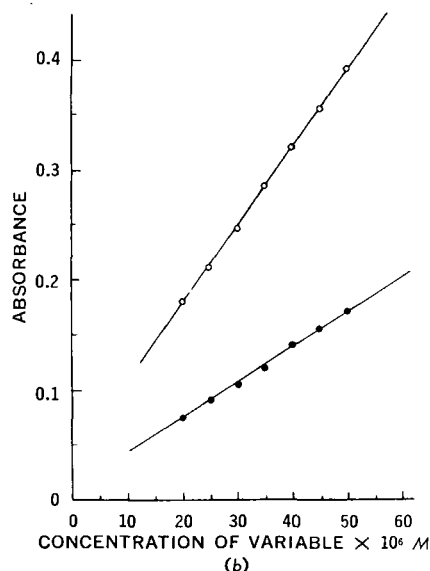
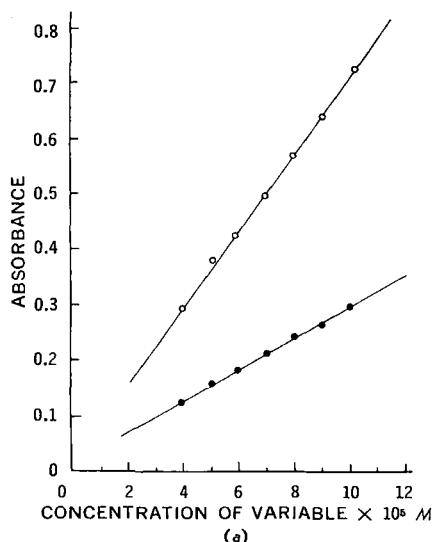


Figure 5—Application of the slope ratio method to: (a) vanadyl sulfate-iodochlorhydroxyquin system and (b) vanadyl sulfate-diiodohydroxyquin system. Key (a): O, [iodochlorhydroxyquin] is constant; and ●, [VO⁺²] is constant. Key (b): O, [diiodohydroxyquin] is constant; and ●, [VO⁺²] is constant.

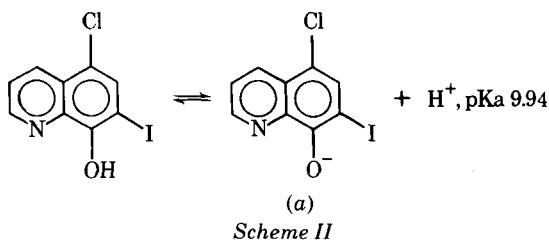
tion of the complex. As Fig. 6 indicates, the predominant complex is the 1:2.

Stability Constant of Vanadyl-Iodochlorhydroxyquin Complexes—The acid dissociation constant of the reagent was determined potentiometrically (Table II) and calculated from the equation:

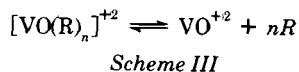
$$\text{pH} = \text{pK}_a + \log \frac{[\text{L}^-]}{[\text{HL}]} \quad (\text{Eq. 1})$$

The average value of pK_a was 9.94.

The reaction between vanadyl ion and iodochlorhydroxyquin was investigated in the pH range of 1.5–2.0. In this range, concentration of the quinolate ion (a) is negligible (Scheme II), meaning that the complexing species is probably the drug iodochlorhydroxyquin itself and not its anion. Similar results were obtained in the kinetic study of complexing Ni⁺² with 8-hydroxyquinoline and 8-hydroxyquinoline-5-sulfonate (25). Attempts to calculate the free ligand concentration [L⁻] and hence the ligand number *n*, using spectrophotometric methods, were unsuccessful. As a result, approximate methods were used to calculate the stability constant of the 1:2 vanadyl-iodochlorhydroxyquin complex (23).



Consider the equilibrium in Scheme III:



If the degree of dissociation is α and the concentration of the complex is C , then the instability constant is given by:

$$K' = (\alpha C)(n\alpha C)^n / C(1 - \alpha) \quad (\text{Eq. 2})$$

The value of n is 2, and C will be equal to the initial concentration of vanadyl sulfate. The value of α is given by:

$$\alpha = A_m - A_s / A_m \quad (\text{Eq. 3})$$

where A_m is the absorbance when all the vanadyl present is com-

plexed and A_s is the absorbance at the stoichiometric molar ratio of R to vanadyl ion in the complex (R = ligand).

The absorption spectra of solutions containing 1:1 to 1:30 vanadyl-ligand ratios were scanned. In Fig. 7a, absorbance is plotted against the number of moles of iodochlorhydroxyquin per mole of vanadyl sulfate for different vanadyl concentrations. From Fig. 7a, α was computed; and using Eq. 2, K' (instability constant) and K (stability constant) were computed. The average value of the stability constant for the complex was 1.1×10^9 , giving $\log \beta_2 = 9.04$. Other attempts to obtain an estimate of the overall stability constant β_2 were tried.

From Fig. 7a, the molar absorptivity of the complex ϵ_2 was computed. At the maximum absorbance the concentration of the complex was assumed to be that of the total metal (vanadyl sulfate). From the values of ϵ_2 and absorbances, the concentrations of the complex were computed; by knowing the total metal and ligand concentration, β_2 was calculated (Table III).

IR Spectra—IR spectra of the precipitated complexes were compared with those of the free ligands (Fig. 8). The stretching —OH band of diiodohydroxyquin appeared as a relatively broad band in the region of 3000 cm^{-1} , indicating strong hydrogen bonding. On complexation, the band was further broadened and red shifted. In the region of 1600 cm^{-1} , the band's characteristics of the C=N group were shifted on complexation. The same observation was made when iodochlorhydroxyquin was the ligand.

Diiodohydroxyquin-Vanadyl Complexes—To confirm these

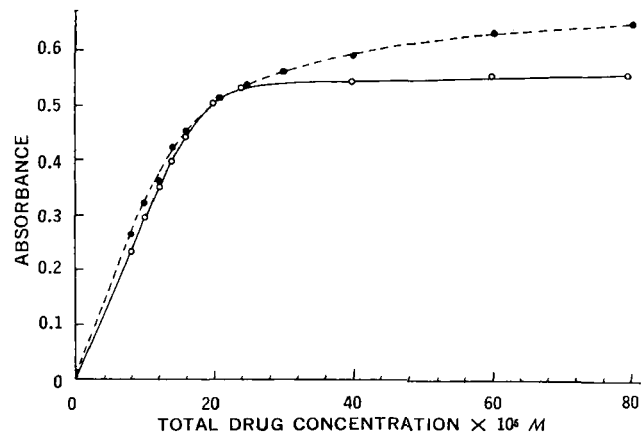


Figure 6—Absorbance as a function of drug concentration at a constant vanadyl-ion concentration. Key: O, using iodochlorhydroxyquin; and ●, using diiodohydroxyquin.

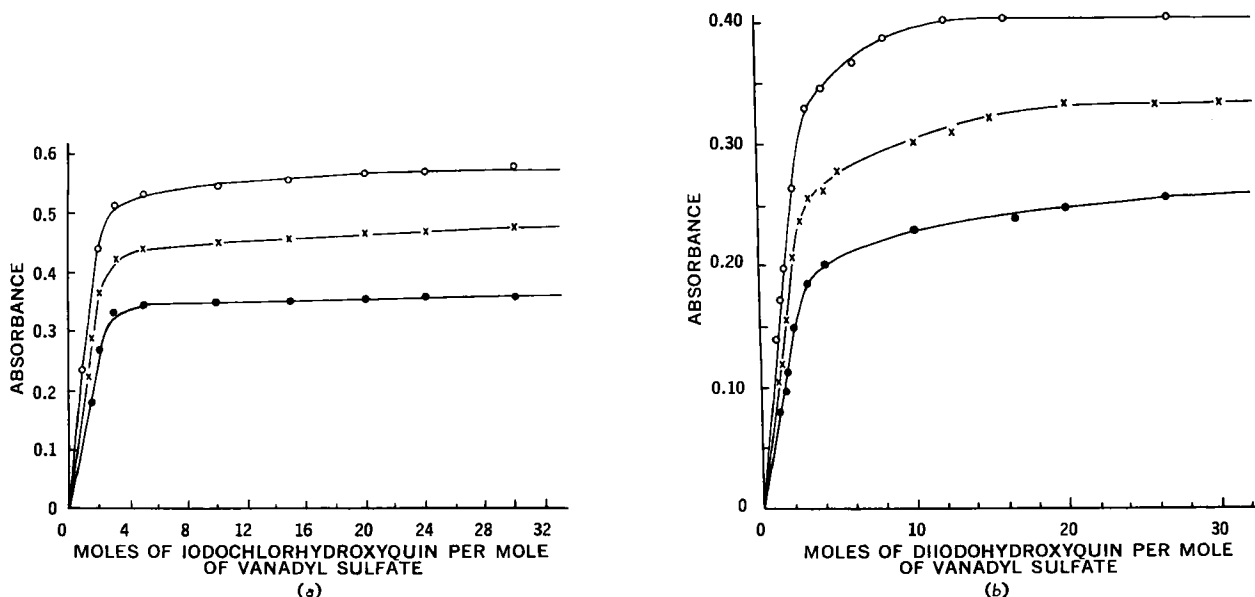


Figure 7—(a) Variation of absorbance of vanadyl sulfate-iodochlorhydroxyquin with iodochlorhydroxyquin concentration. Key: \circ , $[VO^{+2}] = 7.5 \times 10^{-5} M$; \times , $[VO^{+2}] = 6.25 \times 10^{-5} M$; and \bullet , $[VO^{+2}] = 5.0 \times 10^{-5} M$. (b) Variation of absorbance of vanadyl sulfate-diiodohydroxyquin with diiodohydroxyquin concentration. Key: \circ , $[VO^{+2}] = 5 \times 10^{-5} M$; \times , $[VO^{+2}] = 4 \times 10^{-5} M$; and \bullet , $[VO^{+2}] = 3 \times 10^{-5} M$.

results, the complexing properties of diiodohydroxyquin with vanadyl ion were investigated. The procedure used previously was adopted. The electronic structure of the two ligands is similar, yet differences in the electronegativities of iodine and chlorine may have some effects on the chelating properties of the drug.

The acid dissociation constant of diiodohydroxyquin was determined in ethanol, and an average value of 10.15 was obtained for pKa. This means that at pH 1.5-2.0 the concentration of the quinolate ion is negligible and that the ligand is the drug itself. Job's (22) method (Fig. 3) indicates that the 1:2 vanadyl-diiodohydroxyquin chelate is obtained. The complex obtained has an absorption band of its own with λ_{max} 414 nm. The slope ratio (Fig. 5b)

and mole ratio (Fig. 6) methods indicate also that the 1:2 complex predominates.

The instability constant (K') of the 1:2 vanadyl-diiodohydroxyquin complex was computed as before. Data needed for this calculation were taken from a plot (Fig. 7b) of absorbance versus the number of moles of diiodohydroxyquin per mole of vanadyl sulfate. The computed values of K' were used to calculate β_2 , and an average value of 1.8×10^9 was obtained ($\log \beta_2 = 9.25$) (Table III).

CONCLUSION

These results indicate that iodochlorhydroxyquin and diiodohy-

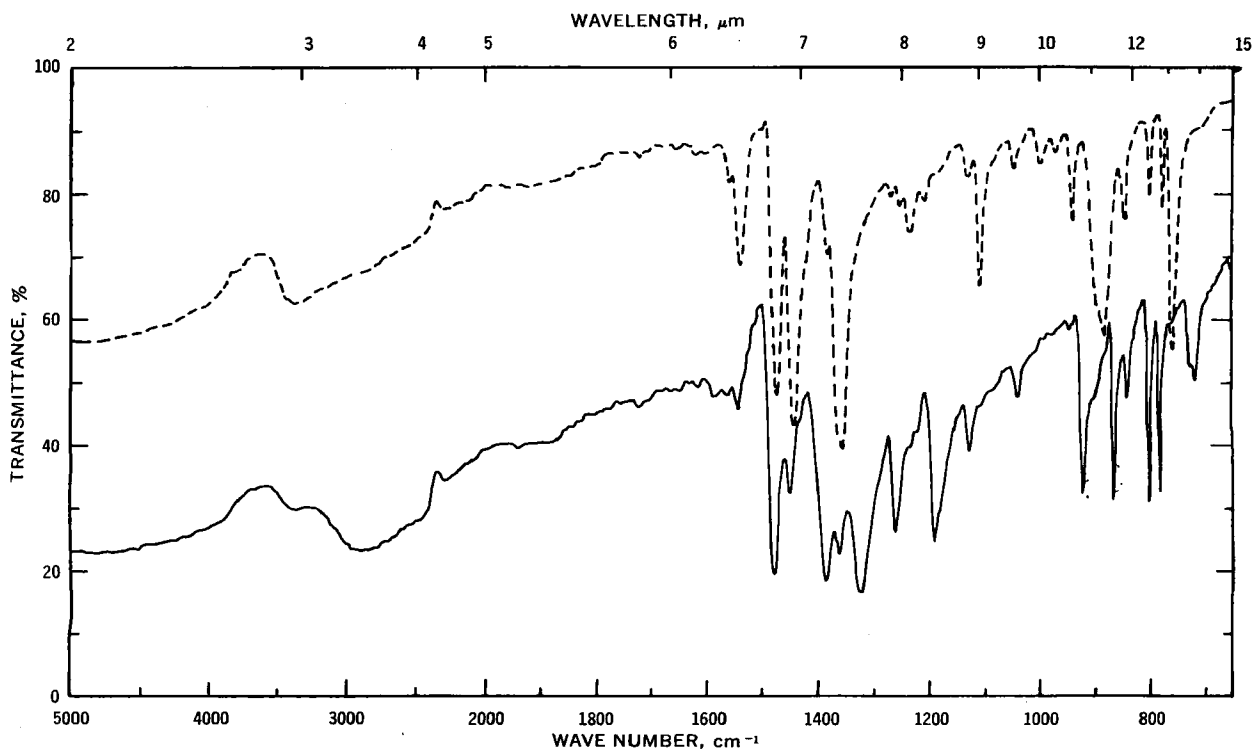


Figure 8—IR spectra of diiodohydroxyquin (—) and diiodohydroxyquin-vanadyl sulfate (---) complex.

Table I—Variation of Absorbance at 410 nm with Concentration of Iodochlorhydroxyquin and Vanadyl Sulfate

[VO ⁺²] = 1.0 × 10 ⁻³ M		Iodochlorhydroxyquin = 1.0 × 10 ⁻³ M	
[Iodochlorhydroxyquin] × 10 ⁵	Absorbance (410 nm)	[VO ⁺²] × 10 ⁵	Absorbance (410 nm)
4.0	0.125	4.0	0.295
5.0	0.155	5.0	0.385
6.0	0.185	6.0	0.425
7.0	0.215	7.0	0.495
8.0	0.245	8.0	0.570
9.0	0.265	9.0	0.640
10.0	0.295	10.0	0.735

Table II—Potentiometric Titration of Iodochlorhydroxyquin

Alcoholic Potassium Hydroxide, ml	pH	[HL], mg	[L ⁻], mg	log $\frac{[HL]}{[L^-]}$	pKa
0.5	9.046	27.335	3.255	0.9242	9.9702
1.0	9.380	24.080	6.510	0.5680	9.9480
1.5	9.580	20.830	9.760	0.3293	9.9093
2.0	9.780	17.570	13.020	0.1302	9.9102
2.5	9.930	14.315	16.275	-0.0557	9.9743
3.1	10.210	10.400	20.190	-0.2881	9.9219
3.5	10.310	7.805	22.785	-0.4651	9.9449
4.0	10.560	4.550	26.040	-0.7577	9.8023 ^a

^a Neglected.

Table III—Values Obtained for the Log β₂ at Different Metal and Ligand Concentrations

Total [VO ⁺²] × 10 ⁵ M	Total Iodochlorhydroxyquin Concentration × 10 ⁵ M	Log β ₂	Total [VO ⁺²] × 10 ⁵ M	Total Diiodohydroxyquin Concentration × 10 ⁵ M	Log β ₂
7.0	35	8.104	1.0	10	8.669
8.0	40	7.954	2.0	20	8.608
9.0	45	7.913	3.0	30	8.624

droxyquin form strong complexes with vanadyl ion, even in distinct acid media. The predominant complexes were the 1:2 complexes. The complex of diiodohydroxyquin is slightly more stable and has a band maximum at a longer wavelength than iodochlorhydroxyquin. These differences are attributed to the differences in electronegativities of chlorine and iodine. Since the com-

plexes were obtained in an acid medium (pH 1.5–2.0), the chelating agent is probably the drug itself and not its anion.

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