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Fluorescence Characteristics of 8-O-Phosphoryl, -Sulfuryl, and -Carboxymethyl Derivatives of 8-Hydroxyquinoline and Its Application to Micro-determination of Cadmium and Zinc

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Fluorescence characteristics of 8-quinolyl derivatives substituted with anionic dissociative group at the hydroxyl in 8-hydroxyquinoline were examined in relation to that of 8-methoxyquinoline. A remarkable effect of Cd(II) and Zn(II) on the increase in fluorescence intensity of both 8-quinolyl dihydrogen phosphate (8-QP) and 8-hydroxyquinoline carboxymethylate (8-HQCM) was observed. Fluorometric examinations of these compounds in the presence of Cd(II) or Zn(II) suggested a possible formation of the fluorescent chelate; 8-QP-Cd(II) (1: 1) and 8-HQCM-Zn(II) (1: 1).

Fluorometric determination of Cd(II) and Zn(II) was successfully achieved by measuring the increase in fluorescence intensity of 8-QP or 8-HQCM in the presence of these ions.

A number of reports on the photoluminescence of 8-hydroxyquinoline (8-HQ)[2] and its derivatives have appeared since Popovych, *et al.* (1959) reported the solvent effect on fluorescence characteristics of 8-HQ.[3] The formation of fluorescent chelates with 8-HQ and its derivatives has been applied to the fluorometric determination of metal ions.

This paper deals with the fluorescence characteristics of 8-HQ derivatives substituted with anionic dissociative group at the hydroxyl in 8-HQ and the fluorometric determination of Cd(II) and Zn(II) with them.

Experimental

Instruments——All fluorescence measurements were made with an Aminco-Bowman Spectrofluorometer using 10 mm square quartz cells. Slit width was set according to the description in Slit Arrangement No. 3.

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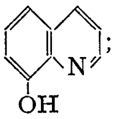
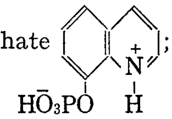
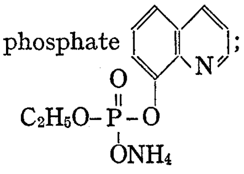
[2] The following abbreviations will be used: 8-HQ, 8-hydroxyquinoline



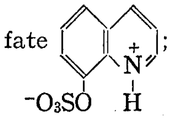
; 8-QP, 8-quinolyl dihydrogen phosphate



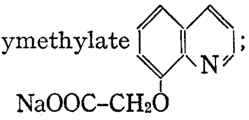
; 8-QEP, 8-quinolyl ethylhydrogen phosphate



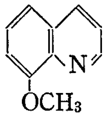
; 8-QS, 8-quinolyl sulfate



; 8-HQCM, 8-hydroxyquinoline carboxymethylate



; 8-MQ, 8-methoxyquinoline



[3] O. Popovych and L.B. Rogers, *Spectrochim. Acta*, **15**, 584 (1959); R.E. Balland and J.W. Edwards, *J. Chem. Soc.*, **1964**, 4868; S. Schulman and Q. Fernand, *Tetrahedron*, **24**, 1777 (1968); M. Goldman and E.L. Wehry, *Anal. Chem.*, **42**, 1178 (1970); S. Schulman, *ibid.*, **43**, 285 (1971); M.P. Bratzel, J.J. Aaron, J.D. Wineforrdner, S.G. Schulman and H. Gershon, *ibid.*, **44**, 1240 (1972).

For the measurement of fluorescence intensity, quinine sulfate solution (1 μg/ml) was used as an internal standard. The pH was measured with a Hitachi-Horiba pH meter F-7 using a combined electrode for the test tube use (#6028-10 T).

Reagents——The metal chlorides used were of commercial analytical grade. Quinine sulfate (Japanese Pharmacopeia) was purified by recrystallization from water and dried over P_2O_5 at 60° for 3 hr. Methanol of special reagent grade was purified by the procedure of Lund, *et al.*[4] Deionized and redistilled water was used in the preparation of all the reagents and standard solutions. 8-Quinolyl dihydrogen phosphate (8-QP, free ester, mp 218—222° (decomp.), UV $\lambda_{\max}^{0.1\text{N HCl}}$ 243 nm (ε 40900)) was synthesized by the method of Murakami, *et al.*[5] and 8-quinolyl ethylhydrogen phosphate (8-QEP, NH_4 salt, mp 145—148°, UV $\lambda_{\max}^{0.1\text{N HCl}}$ 243 nm (ε 32100)) was synthesized by the condensation of 8-QP with ethanol in the presence of dicyclohexylcarbodiimide.[6] 8-Hydroxyquinoline carboxymethylate (8-HQCM, Na salt, mp 238.5°, UV $\lambda_{\max}^{0.1\text{N HCl}}$ 249 nm (ε 39300)) and 8-methoxyquinoline (8-MQ, bp_6 145°) were prepared by the methods of Nagel[7] and of Bedall,[8] respectively. 8-Quinolyl sulfate (8-QS, free ester, mp 158—162° (decomp.), UV $\lambda_{\max}^{0.1\text{N HCl}}$ 240.8 nm (ε 52500)) was synthesized by sulfating 8-hydroxyquinoline with chlorosulfonic acid and N,N-dimethylaniline.[9]

The solutions of metal chlorides (0.1 M) were prepared by dissolving each metal chloride in water, and they were diluted appropriately before use. Tris-HCl buffer was prepared by the method of Gomori using Trizuma base (Sigma Co. Lot 82C-5330),[10] and sodium acetate–HCl buffer was prepared by the method of Walpole.[11] Michaelis' phosphate buffer (pH 7.10) was used for spectral measurement. Quinine sulfate solution (1 μg/ml) was prepared by dissolving 100 mg of purified quinine sulfate in 0.1 N H_2SO_4 to 1000 ml, then diluting 10 ml of this solution to 1000 ml with 0.1 N H_2SO_4.

A 5×10^{-4} M solution of 8-MQ, 8-QP, 8-QEP, 8-QS, and 8-HQCM was prepared by dissolving each of them in water and was used for all fluorescence measurements. The pHs of the solution of 8-QP, 8-QEP, 8-QS, and 8-HQCM were 3.83, 6.62, 3.91, and 6.76, respectively.

Stability of 8-Quinolyl Derivatives——Stability of 8-QP, 8-QEP, 8-QS, and 8-HQCM was examined by spectrometry with their 5×10^{-4} M solutions at pH 3.1, 6.9, and 10.1. Each solution of the compounds was adjusted to the desired pH with 0.1 N HCl or 0.1 N NaOH, and allowed to stand at 5° or 20°. An aliquot taken from these solutions was diluted 20 fold with 0.1 N HCl, and its absorbancy at 252 nm, which is the UV $\lambda_{\max}^{0.1\text{N HCl}}$ of 8-hydroxyquinoline, was measured. The solutions of 8-QS, 8-QEP, and 8-HQCM were stable at 20° for at least 120 hr over all the pHs. Under the same conditions, the solution of 8-QP was stable at pH 10.1 but it was unstable in both acidic and neutral media as indicated by the following data: 10% hydrolysis, 120 hr, 20° (pH 3.1); 22% hydrolysis, 120 hr, 20° (pH 6.9); <5% hydrolysis, 1 week, 5° (pH 3.83).

Result and Discussion

Excitation and Fluorescence Spectra of 8-Quinolyl Derivatives

Figure 1 (a—e) shows the uncorrected excitation and fluorescence spectra of five 8-quinolyl derivatives in acidic, neutral, and alkaline media. The fluorescence intensity of each spectrum is represented by the relative value to that of quinine sulfate solution (1 μg/ml) at 450 nm with excitation at 366 nm. The excitation spectrum of each compound was recorded at its fluorescence maximum wavelength, and the fluorescence spectrum of each compound was recorded at its excitation maximum wavelength. As can be seen from Fig. 1 (b—e), the fluorescence spectra of 8-QEP, 8-QS, and 8-HQCM were similar in their hypsochromic shift in 1 N NaOH with that of 8-MQ. In alkaline medium, the molecular species of 8-MQ in its excited state is a neutral type (Chart 1). The similarity among their fluorescence properties suggests that all of 8-QEP, 8-QS, and 8-HQCM in 1 N NaOH would exist as their neutral molecular species. As can be seen in Fig. 1a, the fluorescence spectrum of 8-QP in 1 N NaOH gave a rather broad peak with a maximum wavelength of 468 nm, whereas the maximum fluores-

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[4] H. Lund and J. Bejerrum, *Ber.*, **64**, 210 (1931).
[5] Y. Murakami, J. Sunamoto, H. Sadamori, H. Kondo and M. Takagi, *Bull. Chem. Soc. Japan*, **43**, 2518 (1970).
[6] K. Nagasawa and H. Yoshidome, *Chem. Pharm. Bull.*, (Tokyo), **20**, 1840 (1972); *idem*, *ibid.*, **21**, 2438 (1973).
[7] O. Nagel, *Monatsh. Chem.*, **18**, 32 (1897).
[8] C. Bedall and O. Fischer, *Ber.*, **14**, 2570 (1881).
[9] K. Nagasawa and H. Yoshidome, *J. Org. Chem.*, in press (1974).
[10] G. Gomori, *Proc. Soc. Exptl. Biol. Med.*, **62**, 33 (1946).
[11] G.S. Walpole, *J. Chem. Soc.*, **105**, 2521 (1914).

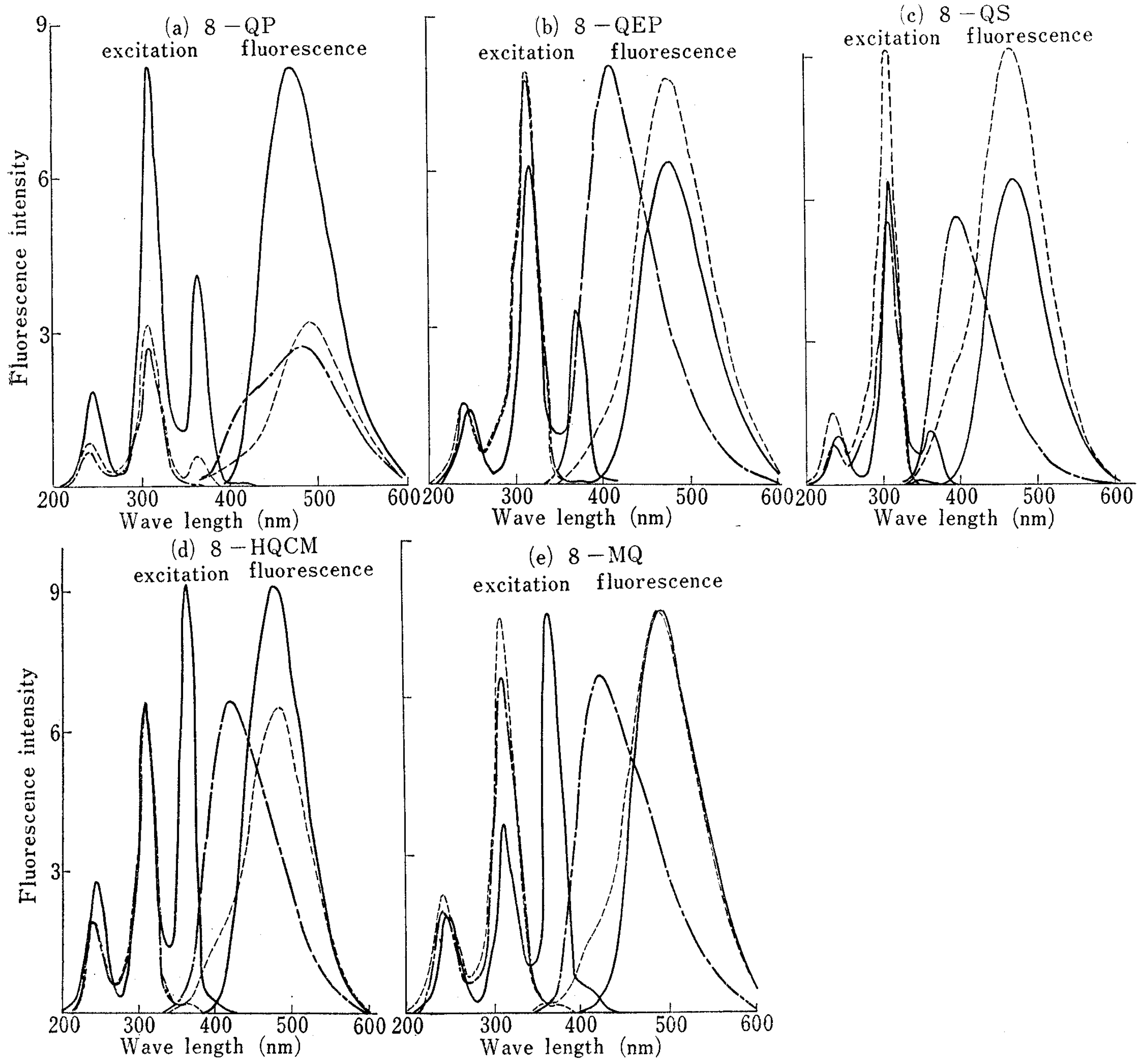

Fig. 1(a—e). Excitation and Fluorescence Spectra of 8-Quinolyl Derivatives

Final concentration of each 8-quinolyl derivative was 5×10^{-5}M. All spectra were recorded with the meter scale set at 30% intensity with reference to the fluorescence intensity of quinine sulfate solution (1 μg/ml), except those of 8-QEP in phosphate buffer which were recorded with one third the sensitivity.

———: 1N HCl; - - - - -: phosphate buffer (pH 7.10); —·—·—: 1N NaOH

cence wavelength of 8-QP shifted completely to 426 nm in 5N NaOH. These results suggest that 8-QP is still partly protonated in 1N NaOH. The appearance of a new peak with a maximum wavelength of 366 nm in the excitation spectra of all the 8-quinolyl derivatives in acidic media suggests the formation of a non-dissociative molecular species of these derivatives in 1N HCl.

The mechanism of fluorescence seems to be somewhat different between the compounds with an anionic group which may form a complex molecular species, as in the case of 8-HQ, and 8-MQ which can form only a cationic or a neutral type as its molecular species.

Influence of Metal Ions on the Fluorescence of 8-Quinolyl Derivatives

As can be seen in Table I, the intensity of fluorescence of 8-QP in Tris-HCl buffer was markedly increased by the addition of both Cd(II) and Zn(II), and that of 8-HQCM was also increased by these ions in both Tris-HCl and sodium acetate–HCl buffers. On the other hand, no marked effect was observed in the case of 8-QEP and 8-QS. The excitation and fluores-

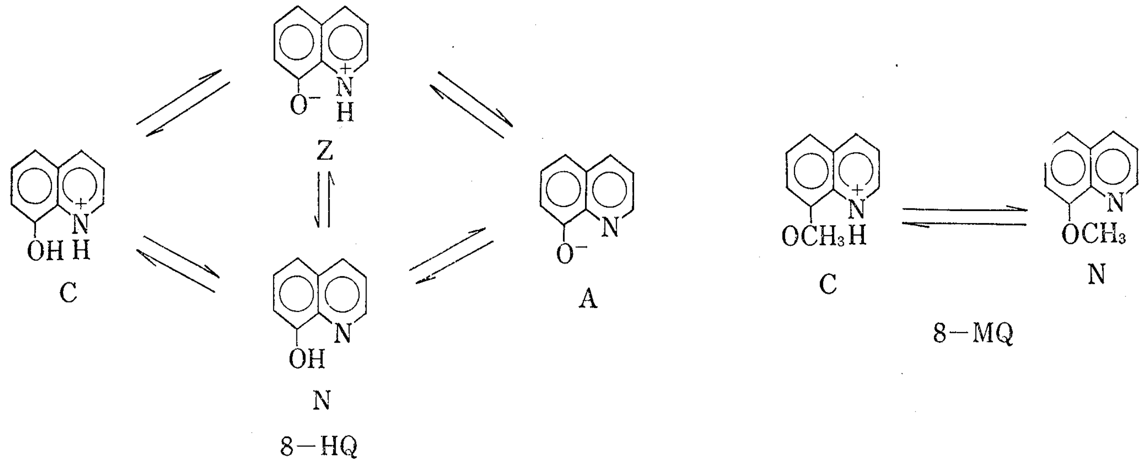

Chart 1. Prototropic Equilibria of 8-HQ and 8-MQ

C: cation molecule, A: anion molecule,
Z: zwitterion molecule, N: neutral molecule

Table I. Effect of Metal Ions on Fluorescence Intensity of 8-Quinolyl Derivatives

8-Quinolyl derivative	Buffer[a]	Fluorescence[b] λmax (nm)	Increase in fluorescence intensity (%)[c]							
			Cd^{2+}	Zn^{2+}	Hg^{2+}	Mg^{2+}	Ca^{2+}	Sr^{2+}	Pb^{2+}	Al^{3+}
8-QP	A	488	325.0	175.0	0	0	0	2.0	−22.0	−15.0
	B	500	1.1	1.1	0	−3.4	0	2.3	−2.3	4.5
8-QEP	A	416	1.9	0	0	0.8	0	0	0.5	−0.8
	B	416	0.9	0	0	4.3	1.4	0	1.2	0.9
8-QS	A	480	1.3	1.9	0.6	3.2	0	1.3	−0.6	−0.6
	B	480	0.6	2.4	1.2	1.2	0	4.2	1.2	−1.8
8-HQCM	A	480	340.0	354.8	0.7	0	7.4	0.7	0.7	−0.7
	B	480	238.3	306.3	11.7	0	0.8	0	3.9	0

Fluorescence intensity at the indicated fluorescence λ maxima was measured on the solution consisted of 5×10^{-4}M 8-quinolyl derivative (0.50 ml), 10^{-4}M metal chloride (1.00 ml), the indicated buffer solution (1.50 ml), methanol (5.00 ml), and water (2.00 ml). The meter scale of the instrument was set at 30% intensity with reference to the fluorescence intensity of quinine sulfate solution (1 μg/ml) at 450 nm with 366 nm excitation.

a) A: 5×10^{-4}M Tris-HCl buffer (final pH 8.58), B: 0.25M sodium acetate–HCl buffer (final pH 5.60)

b) Figures are the fluorescence wavelength maxima of each 8-quinolyl derivative without metal chloride in the indicated medium.

c) Increase in fluorescence intensity (%) was expressed by the ratio of increase in fluorescence intensity to fluorescence intensity of the blank.

cence wavelength maxima of 8-QP with Cd(II) were 316 and 440 nm, and those of the blank were 316 and 488 nm, respectively, indicating a significant hypsochromic shift of the fluorescence wavelength maximum of 8-QP with Cd(II) (Fig. 2). The excitation and fluorescence spectra of 8-HQCM with or without Zn(II) gave the same wavelength maxima, 316 and 416 nm, respectively (Fig. 3).

Fluorescence Characteristics of 8-QP in the Presence of Cd(II)

The pH-fluorescence intensity of 8-QP with Cd(II) or Zn(II) in Tris-HCl buffer indicated that the fluorescence intensity of 8-QP with Cd(II) was larger than that with Zn(II) at pH> 7.60, and the difference between them was the largest at pH 8.75—9.30 (Fig. 4). The result in Table II shows that the alcohols, especially methanol, are highly effective in enhancing the fluorescence intensity. The medium containing 70—80% (v/v) methanol was found to be the most suitable (Fig. 5). Fig. 6 shows, for 0.05 μmole Cd(II), that constant readings are obtained between 0.15 and 0.3 μmole of 8-QP. The fluorescence intensity was stabilized 30 min after the mixture was prepared and did not change for at least 2 hr (Fig. 7).

Assuming that the increase in fluorescence intensity of 8-QP with Cd(II) is due to the formation of a fluorescent chelate, the flurometric molar ratio study was applied to the 8-QP-Cd(II) system.[12] The result shown in Fig. 8 suggests the formation of a fluorescent

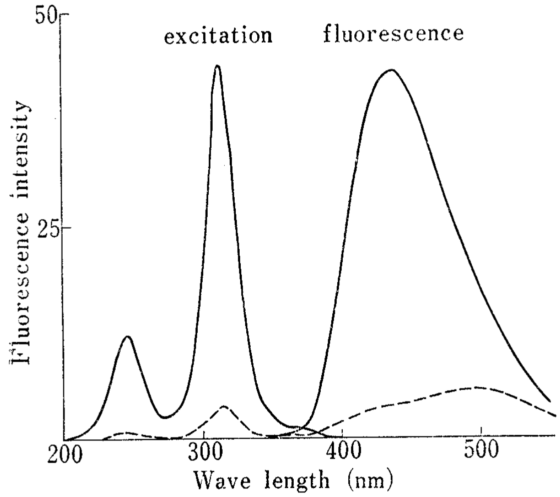

Fig. 2. Excitation and Fluorescence Spectra of 8-QP in the Presence of Cd(II)

Excitation and fluorescence spectra were measured on the solution consisted of 5×10^{-4} M 8-QP (0.50 ml), 10^{-4} M $CdCl_2$ (0.50 ml), 5×10^{-2} M Tris-HCl buffer (pH 9.16, 1.50 ml), and methanol (7.50 ml). The fluorescence spectra were recorded under excitation at 316 nm and the excitation spectra were recorded at 440 nm. Final pH of the solution was 8.82.
——: with Cd(II); ----: without Cd(II)

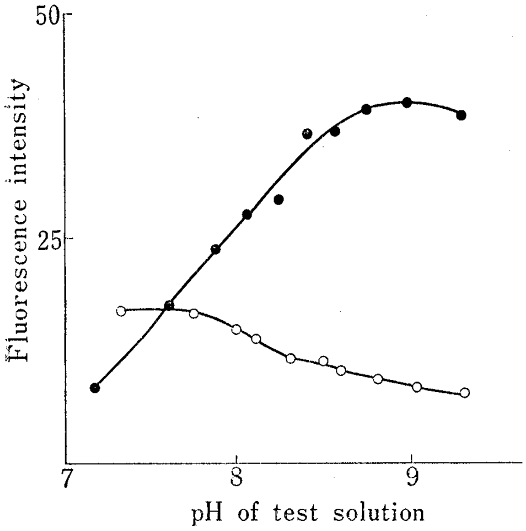

Fig. 4. Effect of pH on Fluorescence Intensity of 8-QP in the Presence of Cd(II) or Zn(II)

Fluorescence intensity at 440 nm with 316 nm excitation was measured on the solutions having the indicated final pHs which consisted of 5×10^{-4} M 8-QP (0.50 ml), 10^{-4} M $CdCl_2$ or $ZnCl_2$ (0.50 ml), 5×10^{-2} M Tris-HCl buffer (pH 7.15—9.33, 1.50 ml), and methanol (7.50 ml).
—●—: 0.05 μmol Cd(II); —○—: 0.05 μmol Zn(II)

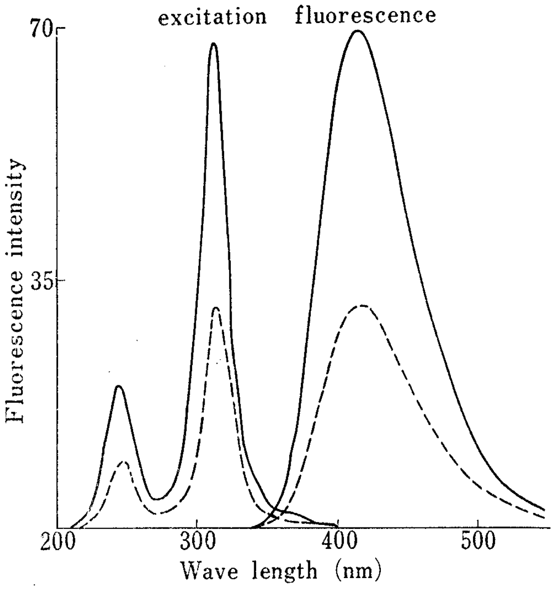

Fig. 3. Excitation and Fluorescence Spectra of 8-HQCM in the Presence of Zn(II)

Excitation and fluorescence spectra were measured on the solution consisted of 5×10^{-4} M 8-HQCM (0.50 ml), 10^{-4} M $ZnCl_2$ (0.50 ml), 0.25 M sodium acetate–HCl buffer (pH 3.70, 1.50 ml), and methanol (7.50 ml). The fluorescence spectra were recorded under excitation at 316 nm and the excitation spectra were recorded at 416 nm. Final pH of the solution was 5.35.
——: with Zn(II); ----: without Zn(II)

Table II. Effect of Organic Solvent on Fluorescence Intensity of 8-QP in the Presence of Cd(II)

Solvent	Increase in fluorescence intensity	Final pH
Methanol	100	8.82
Ethanol	73.1	8.85
n-Propanol	80.1	8.80
Isopropanol	72.1	8.90
Methyl cellosolve	54.7	8.92
Tetrahydrofuran	44.8	8.61
Acetonitrile	41.3	8.73
Dioxane	34.8	8.88
Dimethylformamide	10.5	9.73
Dimethylsulfoxide	4.0	10.55

Fluorescence intensity at 440 nm with 316 nm excitation was measured on the solution consisted of 5×10^{-4} M 8-QP (0.50 ml), 10^{-4} M $CdCl_2$ (0.50 ml), 5×10^{-2} M Tris-HCl buffer (pH 9.16, 1.50 ml), and the indicated organic solvent (7.50 ml). The increase in fluorescence intensity was expressed by the value relative to that of methanol.

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[12] D.C. Freeman, Jr. and C.E. White, *J. Amer. Chem. Soc.*, **78**, 2678 (1956).

chelate consisting of 8-QP and Cd(II) in 1:1 ratio, although its linkage would be somewhat loose since the curve obtained was not very sharp, and any change in its absorption spectra with or without Cd(II) was not observed.

Fluorometric Determination of Cd(II) with 8-QP

On the basis of the results described above, the following procedure was constituted for the fluorometric determination of Cd(II) with 8-QP: A series of standard $CdCl_2$ solution

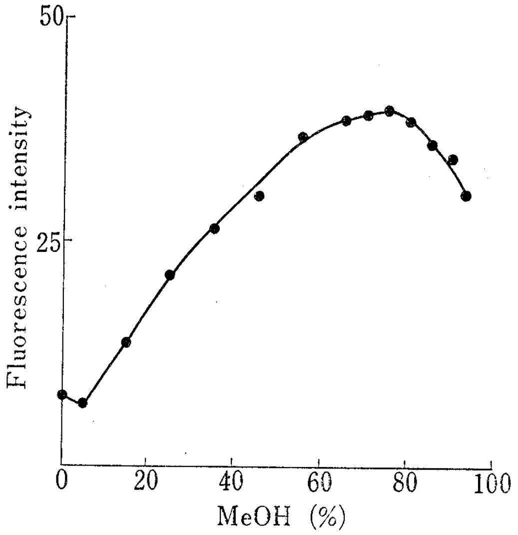

Fig. 5. Effect of Methanol Concentration on Fluorescence Intensity of 8-QP in the Presence of Cd(II)

Fluorescence intensity at 440 nm with 316 nm excitation was measured on the solutions having the indicated final methanol concentration which consisted of 2.5×10^{-3}M 8-QP (0.10 ml), 5×10^{-4}M $CdCl_2$ (0.10 ml), 0.1M Tris-HCl buffer (pH 9.02, 0.50 ml), and aqueous methanol varying in concentration (9.30 ml).

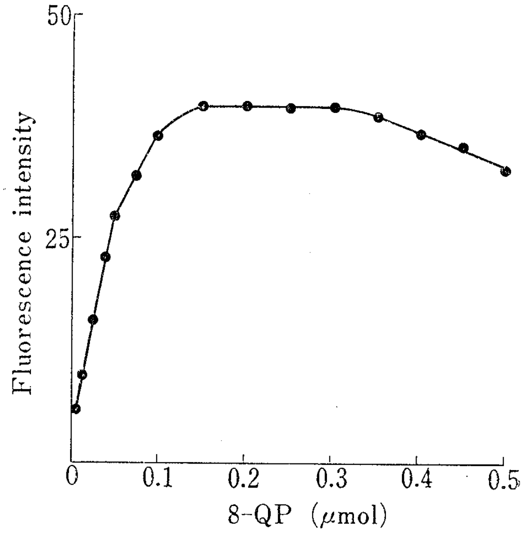

Fig. 6. Effect of 8-QP Concentration on Fluorescence Intensity in the Presence of Cd(II)

Fluorescence intensity at 440 nm with 316 nm excitation was measured on the solutions (10 ml) having the indicated amount of 8-QP which consisted of 10^{-4}M $CdCl_2$ (0.50 ml), 5×10^{-2}M Tris-HCl buffer (pH 9.16, 1.50 ml), and methanol (7.0 ml).

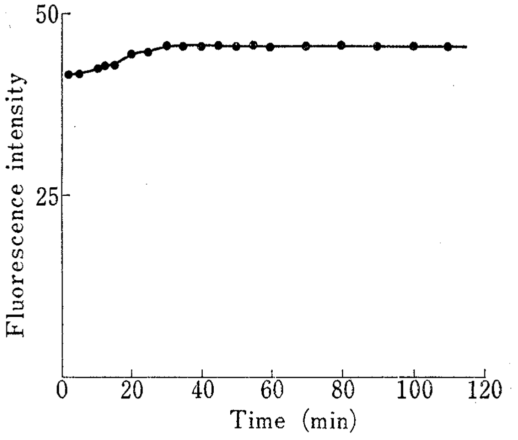

Fig. 7. Stability of Fluorescence Intensity of 8-QP in the Presence of Cd(II)

Fluorescence intensity at 440 nm with 316 nm excitation was measured on the solution consisted of 5×10^{-4}M 8-QP (0.50 ml), 10^{-4}M $CdCl_2$ (0.50 ml), 5×10^{-2}M Tris-HCl buffer (pH 9.16, 1.50 ml), and methanol (7.50 ml) at indicated intervals. Final pH of the solution was 8.82.

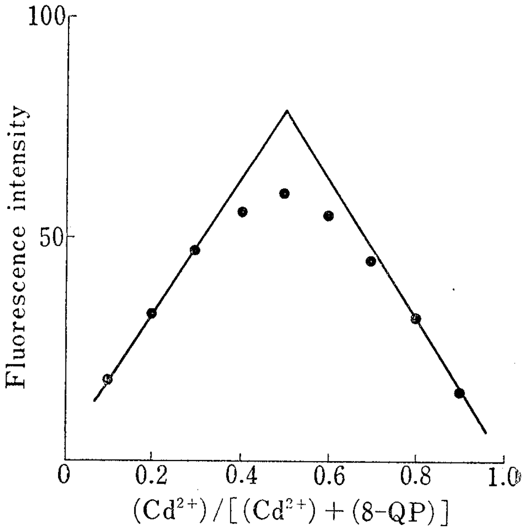

Fig. 8. Fluorometric Mole Ratio Determination for 8-QP and Cd(II) System

The solutions (10 ml) with a final concentration of $[Cd^{2+}]+[8\text{-}QP]=3\times10^{-5}$M were prepared from 1.5×10^{-4}M 8-QP (0.10—0.90 ml), 1.5×10^{-4}M $CdCl_2$ (0.90—0.10 ml), 5×10^{-2}M Tris-HCl buffer (pH 9.16, 1.50 ml), and methanol (7.50 ml). Fluorescence intensity was measured at 440 nm with 316 nm excitation.

(0.50 ml) are mixed with 5×10^{-4} M 8-QP (0.50 ml), 5×10^{-2} M Tris-HCl buffer (pH 9.16, 1.50 ml), and methanol (7.50 ml). After standing at room temperature for at least 30 min, the fluorescence intensity of these mixtures is measured at 440 nm using an excitation wavelength of 316 nm. The intensity is also measured on the blank prepared by replacing the $CdCl_2$ solution with water. The meter scale of the instrument is set at 30% intensity with reference to the fluorescence intensity of quinine sulfate solution (1 μg/ml) at 440 nm with 316 nm excitation.

The result obtained by this analytical procedure showed a good linearity between the increase in fluorescence intensity and the concentration of Cd(II) from 0 to 0.1 μmole. The standard deviation based on the five determinations on Cd(II) (0.05 μmole) was 0.62%. The effect of metal ions on the determination of Cd(II) (0.05 μmole) was examined. As shown in Table III, the colored ions such as Ni(II), Cu(II), and Co(II) interfered seriously at a level of 50 times the molar amount. Therefore, it is necessary for the determination of Cd(II) with 8-QP to remove these interfering ions before the operation.

Fluorescence Characteristics of 8-HQCM in the Presence of Zn(II)

The pH-fluorescence intensity of 8-HQCM with Zn(II) or Cd(II) in sodium acetate–HCl buffer indicated that the maximum fluorescence intensities of 8-HQCM with Zn(II) and Cd(II) were attained at pH 5.2—5.75 and pH>7.60, respectively, and the difference in these intensities was the largest at pH 5.0—5.4 (Fig. 9). Table IV shows that alcohols, especially methanol, are highly effective in enhancing the fluorescence intensity as in the case of 8-QP with Cd(II). A medium containing 75—85% (v/v) methanol was found to be the most suitable (Fig. 10).

TABLE III. Influence of Metal Ion on the Determination of Cd(II) with 8-QP

Metal ion added	Amount of metal ion (μmole)	Recovery of Cd(II) (%)
None	—	100
Ca^{2+}	2.5	71.5
	0.3	78.7
Hg^{2+}	2.5	85.9
	0.3	91.9
Mg^{2+}	2.5	48.5
	0.3	55.7
Al^{3+}	2.5	48.2
	0.3	78.7
Zn^{2+}	2.5	229.0
	0.3	159.0
Ni^{2+}	2.5	4.1
Cu^{2+}	2.5	2.7
Co^{2+}	2.5	1.1

The amount of Cd(II) taken was 0.05 μmole. Metal ions were added as choride. The procedure was identical to that used in the determination of Cd(II) with 8-QP described in the text, except addition of the metal chlorides indicated.

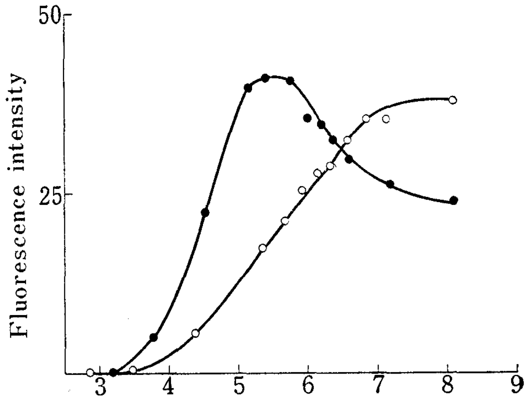

Fig. 9. Effect of pH on Fluorescence Intensity of 8-HQCM in the Presence of Cd(II) or Zn(II)

Fluorescence intensity at 416 nm with 316 nm excitation was measured on the solutions having the indicated final pHs which consisted of 5×10^{-4} M 8-HQCM (0.50 ml), 10^{-4} M $CdCl_2$ or $ZnCl_2$ (0.50 ml), 0.25 M sodium acetate–HCl buffer (pH 3.05—5.50, 1.50 ml), and methanol (7.50 ml).
—●—: 0.05 μmol Zn(II); —○—: 0.05 μmol Cd(II)

For 0.05 μmole Zn(II), the effect of varying the concentration (0.01 to 0.5 μmole) of 8-HQCM on its fluorescence intensity was examined in a mixture consisting of 10^{-4} M $ZnCl_2$ (0.50 ml), 8-HQCM solution of varying concentrations (1.00 ml), 0.25 M sodium acetate–HCl buffer (pH 3.70, 1.50 ml), and methanol (7.00 ml). The fluorescence intensity increased linearly with the concentration of 8-HQCM until 0.1 μmole. At higher concentrations between 0.1 and 0.5 μmole, the intensity continued to increase with 8-HQCM concentration although its increment slowed down. The fluorescence intensity was stabilized 15 min after the mixture was prepared and did not change for at least 60 min (Fig. 11).

The fluorometric molar ratio study was applied to the 8-HQCM-Zn(II) system as in the case of 8-QP-Cd(II). The result shown in Fig. 12 suggests the formation of a fluorescent chelate consisting of 8-HQCM and Zn(II) in 1:1 ratio. However, the linkage of the chelate, if formed, would be rather loose since the curve obtained was broad, and any change in the

Table IV. Effect of Organic Solvent on Fluorescence Intensity of 8-HQCM in the Presence of Zn(II)

Sovlvent	Increase in fluorescence intensity	Final pH
Methanol	100	5.35
Ethanol	65.4	5.61
n-Propanol	48.1	5.42
Isopropanol	42.1	5.62
Acetonitrile	36.2	5.76
Methyl cellosolve	33.9	5.84
Dimethylformamide	7.8	7.26
Dimethylsulfoxide	3.9	7.60
Tetrahydrofuran	0	6.08
Dioxane	0	6.42

Fluorescence intensity at 416 nm with 316 nm excitation was measured on the solution consisted of 5×10^{-4}M 8-HQCM (0.50 ml), 10^{-4}M $ZnCl_2$ (0.50 ml), 0.25M sodium acetate-HCl buffer (pH 3.70, 1.50 ml), and the indicated organic solvent (7.50 ml). The increase in fluorescence intensity was expressed by the value relative to that of methanol.

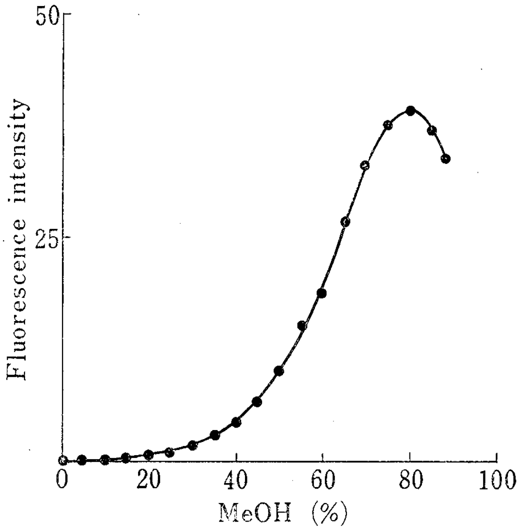

Fig. 10. Effect of Methanol Concentration on Fluorescence Intensity of 8-HQCM in the Presence of Zn(II)

Fluorescence intensity at 416 nm with 316 nm excitation was measured on the solutions having the indicated final methanol concentration which consisted of 2.5×10^{-3}M 8-HQCM (0.10 ml), 5×10^{-4}M $ZnCl_2$ (0.10 ml), 0.6M sodium acetate-HCl buffer (pH 3.70, 1.00 ml), and aqueous methanol varying in concentration (8.80 ml).

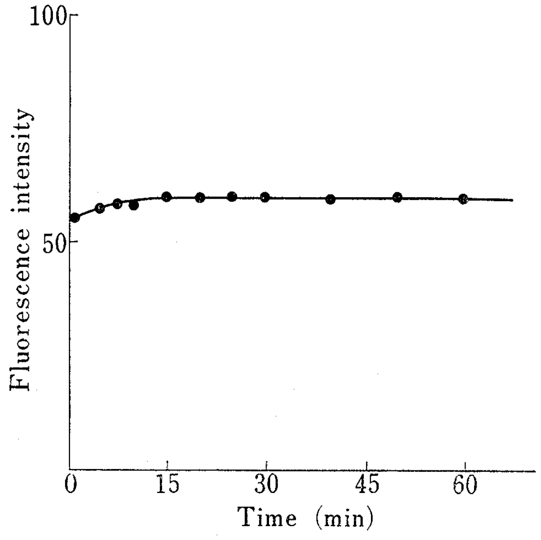

Fig. 11. Stability of Fluorescence Intensity of 8-HQCM in the Presence of Zn(II)

Fluorescence intensity at 416 nm with 316 nm excitation was measured at indicated intervals on the solution consisted of 5×10^{-4}M 8-HQCM (0.50 ml), 10^{-4}M $ZnCl_2$ (0.50 ml), 0.25M sodium acetate-HCl buffer (pH 3.70, 1.50 ml), and methanol (7.50 ml). Final pH of the solution was 5.35.

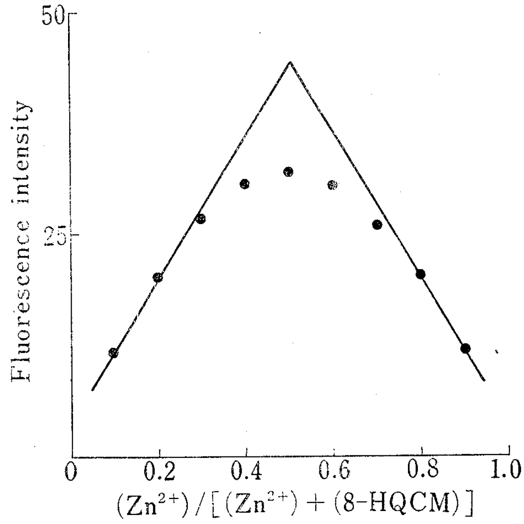

Fig. 12. Fluorometric Mole Ratio Determination for 8-HQCM and Zn(II) System

The solutions (10 ml) with a final concentration of $[Zn^{2+}]+[8\text{-HQCM}]=3\times10^{-5}$M were prepared from 1.5×10^{-4}M 8-HQCM (0.10—0.90 ml), 1.5×10^{-4}M $ZnCl_2$ (0.90—0.10 ml), 0.25M sodium acetate-HCl buffer (pH 3.70, 1.50 ml), and methanol (7.50 ml). Fluorescence intensity was measured at 416 nm with 316 nm excitation.

wavelength maxima of both the fluorescence and absorption spectra of 8-HQCM with or without Zn(II) was not observed.

Fluorometric Determination of Zn(II) with 8-HQCM

From the results described above, the following procedure was constituted for the fluorometric determination of Zn(II) with 8-HQCM: A series of standard $ZnCl_2$ solution (0.50 ml) are mixed with 5×10^{-4}M 8-HQCM (0.50 ml), 0.25M sodium acetate–HCl buffer (pH 3.70, 1.50 ml), and methanol (7.50 ml) in this order. After standing at room temperature for at least 15 min, the fluorescence intensity of the mixtures is measured at 416 nm with 316 nm excitation. The intensity is also measured on a blank prepared with water in place of $ZnCl_2$ solution. The meter scale of the instrument is set at 15% intensity with reference to the fluorescence intensity of quinine sulfate solution (1 µg/ml) at 440 nm with 316 nm excitation.

Practice of this procedure gave a good linearity between the increase in fluorescense intensity and the concentration of Zn(II) from 0 to 0.1 µmole. The standard deviations based on each five determinations on 0.025 and 0.05 µmole of Zn(II) were 0.88 and 0.81%, respectively. Examination on the effect of foreign ions on the determination of Zn(II) (0.025 µmole) showed, as indicated in Table V, no marked effect on the fluorescence intensity of 8-HQCM with Zn(II) except Cd(II) which enhanced it considerably at a level 5 times the molar amount.

TABLE V. Influence of Foreign Ion on the Determination Zn(II) with 8-HQCM

Ion	Recovery of Zn(II) (%)	Ion	Recovery of Zn(II) (%)
None	100	Sr^{2+}	101.4
Hg^{2+}	98.0	Cd^{2+}	303.4
Mn^{2+}	97.7	Cu^{2+}	78.1
Pb^{2+}	99.4	F^-	98.7
Ni^{2+}	86.0	Br^-	103.1
Co^{2+}	94.9	I^-	97.3
Cr^{3+}	97.2	SO_4^{2-}	100.0
Fe^{3+}	96.3	NO_3^-	95.3
Mg^{2+}	99.4	HPO_4^{2-}	98.9
Ca^{2+}	107.7	SCN^-	98.9
Al^{3+}	98.9		

The amount of Zn(II) taken was 0.025 µmole. Metal ions were added as chloride, and anions were added as sodium salt except ammonium rhodanate. The procedure was identical to that used in the determination of Zn(II) with 8-HQCM described in the text, except addition of the metal chlorides (0.125 µmole) or anions (0.25 µmole) indicated.

Although there have been many reports on spectrofluorometric study of the complexation of Cd(II) or Zn(II) ion with free and substituted 8-hydroxyquinolines,13) only a few fluorometric methods for the determination of these metal ions have been reported. The fluorescence of Cd(II)-8-hydroxyquinoline-5-sulfonic acid complex permits the determination of 1 ppb of Cd(II) with an accuracy of ±0.1 ppb.14) The fluorescence of a stabilized dispersion of Zn(II)-8-hydroxyquinoline complex has been used for the determination of Zn(II) in the concentration range of 0.4 to 12 ppm.15) Studies on three fluorescing Zn(II)-2-methyl-8-hydroxyquinoline complexes have not resulted in an analytical application.16) The present

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13) H.M. Stevens, *Anal. Chim. Acta*, **20**, 389 (1959); J.A. Bishop, *ibid.*, **29**, 172 (1963); D.C. Bhatnagar and L.S. Forster, *Spectrochim. Acta*, **21**, 1803 (1965).

14) D.E. Ryan, A.E. Pitts and R.M. Cassidy, *Anal. Chim. Acta*, **34**, 491 (1966).

15) L.L. Merritt, Jr., *Anal. Chem.*, **16**, 758 (1944).

16) D.A. Carter and W.E. Ohnesorge, *Anal. Chem.*, **36**, 327 (1964).

methods using 8-QP and 8-HQCM permit the determination of Cd(II) and Zn(II) at a 2.25 and 1.25 ppm lower concentration limit, respectively. The method for determination of Cd(II) is evidently inferior to that using 8-hydroxyquinoline-5-sulfonic acid, both in the sensitivity and metal ion interferences. However, the sensitivity of the method for Zn(II) determination was comparable to that using 2-methyl-8-hydroxyquinoline together with its limited metal ion interferences.