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Synthesis, Characterization and Antifungal Activity of Metal Chelates Based on 8-Hydroxyquinoline and Metronidazole

Swati Patel*[1], Yogesh Patel[2]

*[1]Department of Chemistry, Gujarat University, Ahmedabad, Gujarat, India.

[2]Government Science College, Limkheda, Dist. Dahod, (Gujarat), India.

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ABSTRACT

In the present study, 5-chloromethyl-8-hydroxyquinoline (CMQ) hydrochloride, a versatile derivative of 8-hydroxyquinoline is reacted with a potent antibiotic drug metronidazole in order to yield a novel ligand namely 5-((2-(2-methyl-5-nitro-1H-imidazol-1-yl) ethoxy) methyl)-8-hydroxyquinoline (NIHQ). The co-ordinate metal chelates of the ligand NIHQ were prepared using divalent metal ions viz., Cu^{2+}, Co^{2+}, Ni^{2+}, Mn^{2+}, Zn^{2+} and Cd^{2+} by simple method. The ligand NIHQ and its all metal chelates were duly characterized for elemental content, spectral features, M: L (metal: ligand) ratio, and magnetic moment. The results of electronic spectral studies and magnetic properties indicated octahedral geometry for all the metal chelates. Antifungal activity of all the samples was tested against plant pathogens such as Aspergillus niger, Botrydepladia thiobromine, Nigrospora Sp., and Fusarium oxyporium. The results showed promising antifungal activity of all the metal chelates.

Keywords: 8-hydroxyquinoline, metronidazole, metal chelates, spectral studies, magnetic properties, antifungal activity.

I. INTRODUCTION

In last few decades, 8-hydroxyquinoline (8-HQ) and its derivatives have grabbed an attention of researchers all over the globe due to its potent biological and medicinal applications as antimicrobial, antioxidant, , anti-inflammatory, antiviral, anti-tubercular, and anti-HIV agents[1-6]. 8-hydroxyquinolines (8HQs) are a family of lipophilic metal ion chelators which has a greater coordinating ability and good metal recognition properties [7-9]. The versatility of 8-HQs have attributed to their applications as OLEDs, fluoroscent chemo sensors and corrosion inhibitors [10-12]. Also, 8-HQs have proven efficacy as anticancer, anti-alzheimer, antineurodegenerative agents [13, 14].

5-chloromethyl-8-hydroxyquinoline (CMQ), one of the most versatile derivatives of 8-hydroxyquinoline, is easy to synthesize and stable in the form of its hydrochloride salt have been reported to possess anti-fungal as well as anti-cancer applications [15, 16].

Metronidazole has been used to treat wide range of infections since decades [17] and many metronidazole conjugates have been synthesized and examined so far for their antimicrobial, anticancer, anti-diabetic, anti-inflammatory, anti-HIV and anti-parasitic applications [18].

Hence, it was thought to club these two pharmacologically potent molecules viz., CMQ and metronidazole in a single molecular framework and synthesize various metal chelates by reacting the ligand with various divalent metal ions such as Cu^{2+}, Co^{2+}, Ni^{2+}, Mn^{2+},Zn^{2+} and Cd^{2+} in order to investigate their antifungal activity.

In the present work, the synthesis of ligand 5-((2-(2-methyl-5-nitro-1H-imidazol-1- yl)ethoxy) methyl)-8-hydroxyquinoline (NIHQ) is carried out by reaction between 5-chloromethyl-8-hydoxyquinoline hydrochloride and metronidazole followed by preparation of its metal chelates with Cu^{2+}, Co^{2+}, Ni^{2+}, Mn^{2+},Zn^{2+} and Cd^{2+} metal salts. Spectroscopic techniques such as FTIR, ^{1}H NMR and LC-MS were used for structural verification of the ligand. Electronic spectral studies and magnetic susceptibility measurements have been performed for determination of geometry of metal chelates. Ligand and metal chelates were further incorporated for analysing their antifungal activity.

II. METHODS AND MATERIAL

All the chemicals were purchased from local market and used directly without further purification.

Procedure for Synthesis of 5-((2-(2-Methyl-5-Nitro-1H-Imidazol-1-Yl) Ethoxy) Methyl)-8-Hydroxyquinoline (NIHQ):

5-chloromethyl-8-hydroxyquinoline (CMQ) was prepared by reported method [19, 20].

The simple nucleophilic substitution reaction between CMQ and metronidazole has facilitated formation of the ligand 5-((2-(2-methyl-5-nitro-1H-imidazol-1-yl) ethoxy) methyl)-8-hydroxyquinoline as described below.

To a mixture of metronidazole (0.15 mole) and 5-chloromethyl-8-hydroxyquinoline (0.15 mole) in ethyl acetate, 12.4 g sodium bicarbonate was added. The reaction mixture was refluxed on steam bath for 1.5 hour. The resultant mass was filtered and washed with ethyl acetate and finally with hot water and air-dried.

The yield of the product was 70%; melting point was 124-125^{0}C (uncorrected). It was directly used for metal chelate formation.

General Procedure for Synthesis of Metal Chelates of NIHQ:

The metal chelates of NIHQ with Cu^{2+}, Co^{2+}, Ni^{2+}, Mn^{2+},Zn^{2+} and Cd^{2+} metal salts were prepared in two steps.

Step-I Preparation of NIHQ Solution:

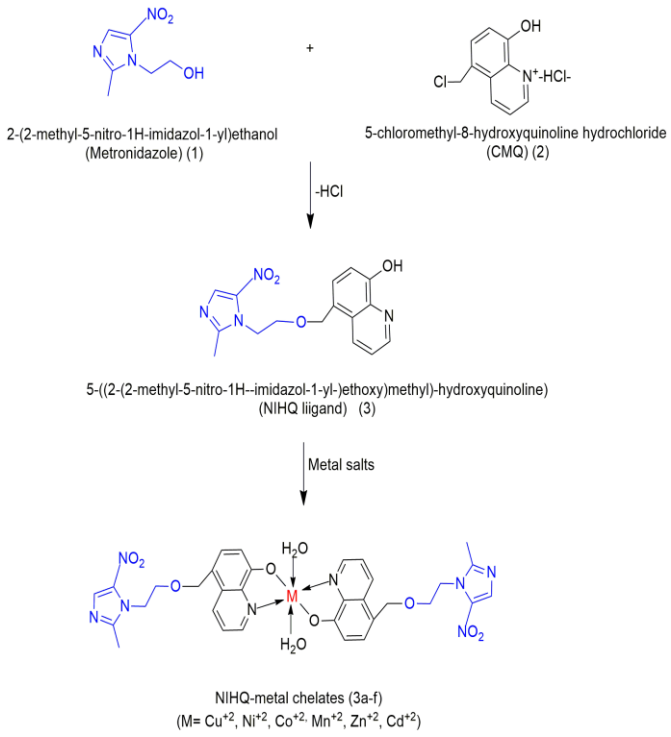
NIHQ (0.1 mole) was taken in 500 ml beaker and formic acid (85% v/v) was added up to slurry formation. To this slurry, water was added till complete dissolution of NIHQ and diluted to 100 ml.

Step-II Synthesis of NIHQ metal chelates:

In a solution of metal acetate (0.01mol) in acetone: water (50:50 v/v) mixture (40 ml), 20 ml of NIHQ solution (containing 0.02 M NIHQ) was added with vigorous stirring at room temperature. The appropriate pH was adjusted by adding sodium acetate for complete precipitation of metal chelate. The precipitates were digested on a boiling water bath. The precipitates of metal chelates were filtered off, washed by water and air-dried.

III. MEASUREMENTS

The elemental contents were determined by Thermo Finigen Flash1101 EA (Itally) and the metals of metal chelates were determined volumetrically by Vogel's method [21]. To a 100 mg chelate sample, each 1 ml of Conc. HCl, H_2SO_4 and $HClO_4$ were added and then 1 g of 0.1 N $NaClO_4$ was added. The mixture was evaporated to dryness and the resulting salt was dissolved in double distilled water and diluted to the mark. From this solution, the metal content was determined by titration with standard EDTA solution. Infrared spectra of the synthesized compounds were recorded on Nicolet 760 FT-IR spectrometer. NMR spectrum of NIHQ was recorded on 60 MHz NMR spectrophotometer. LC-MS of ligand was taken on LC-MSD-Trap-SL_01046. Magnetic susceptibility measurement of the synthesized chelates was carried out on Gouy Balance at room temperature. Mercury tetrathio cynato cobalatate (II) $Hg[Co(NCS)_4]$ was used as a calibrant. The electronic spectra of chelates in solid were recorded at room temperature. MgO was used as reference. A fungal activities of the samples were investigated by reported methods [22,23].



Reaction Scheme

Compound 3: yield= 70%; Anal. Cald for $C_{16}H_{16}N_4O_4$ (328.33) Cald.: %C, 58.53 ; %H, 4.91; %N, 17.06. Found: %C,58.5 ; %H,4.8 ; %N,17.0.

IR (KBR) (cm^{-1}): 3340 (-OH st.), 3020, 2875(C-H st.), 1595,1475(C-H bend.) 1625, 1585, 1460 and 750 (8-HQ characteristic bands),1550,1340 ($C-NO_2$), 1270 (C-N st.), 1120(C-O st.)

^1H NMR (δ ppm): 5.37 (s,1H,-OH), 7.10-8.92(m, 6H, Quinoline and aromatic), 4.35-3.80(t, 4H,$-CH_2-$), 4.82(s, 2H,$-CH_2-$), 2.55(s, 3H,$-CH_3$).

CMR signal(δ ppm): 72.5,146.8,125.7,111.8,153.5,137.5,150.1,122.3, 131.7, 126.9,70.8,42.2,138.6,132.3,151.8,13.1.

LC-MS: 329.37 m/z.

Compound 3a: yield=73%; Anal. Cald for $C_{32}H_{30}N_8O_8Cu^{+2}2H_2O$ (754.22) Cald.: %C,50.96; %H,4.54; %N,14.86; %Cu,8.43. Found: %C,50.9; %H,4.5; %N,14.8; %Cu,8.4.

IR (KBR) (cm^{-1}): 3020,2950,2875(C-H st.) 1595,1475 (C-H bend.), 1624, 1580, 1507 and 776 (8-HQ characteristic bands), 1550,1340($C-NO_2$),1239(C-N), 1128(C-O st.).

Compound 3b: yield=77%; Anal. Cald for $C_{32}H_{30}N_8O_8Ni^{+2}2H_2O$ (749.37) Cald.: %C,51.29; %H,4.57; %N,14.95; %Ni,7.83. Found: %C,51.2; %H,4.5; %N,14.9; %Cu,7.8.

IR (KBR) (cm^{-1}): 3025,2952,2872(C-H st.) 1598,1470 (C-H bend.), 1626, 1578, 1508 and 780 (8-HQ characteristic bands), 1550,1345($C-NO_2$),1240(C-N), 1125(C-O st.).

Compound 3c: yield=74%; Anal. Cald for $C_{32}H_{30}N_8O_8Co^{+2}2H_2O$ (749.60) Cald.: %C,51.27; %H,4.57; %N,14.95; %Co,7.86. Found: %C,51.2; %H,4.5; %N,14.9; %Cu,7.8.

IR (KBR) (cm^{-1}):
3020,2948,2877(C-H st.) 1598,1470 (C-H bend.), 1620, 1585, 1502 and 772 (8-HQ characteristic bands), 1540,1336($C-NO_2$),1239(C-N), 1120(C-O st.).

Compound 3d: yield=75%; Anal. Cald for $C_{32}H_{30}N_8O_8Mn^{+2}2H_2O$ (745.61) Cald.: %C,51.54;

%H,4.60; %N,15.06; %Mn,7.37. Found: %C,51.5; %H,4.5; %N,15.0; %Mn,7.3.

IR (KBR) (cm^{-1}): 3025,2953,2878(C-H st.) 1592,1476 (C-H bend.), 1625, 1575, 1500 and 780 (8-HQ characteristic bands), 1548,1343(C-NO_2),1238(C-N), 1126(C-O st.).

Compound 3e: yield=76%; Anal. Cald for $C_{32}H_{30}N_8O_8Zn^{+2}2H_2O$ (756.05) Cald.: %C,50.84; %H,4.53; %N,14.83; %Zn,8.65. Found: %C,50.8; %H,4.5; %N,14.8; %Zn,8.6.

IR (KBR) (cm^{-1}): 3020,2947,2870(C-H st.) 1598,1470 (C-H bend.), 1622, 1583, 1508 and 785 (8-HQ characteristic bands), 1555,1340(C-NO_2),1240(C-N), 1122(C-O st.).

Compound 3f: yield=72%; Anal. Cald for $C_{32}H_{30}N_8O_8Cd^{+2}2H_2O$ (803.08) Cald.: %C,47.85; %H,4.27; %N,13.96; %Cd,14.00. Found: %C,47.8; %H,4.2; %N,13.9; %Cu,13.9.

IR (KBR) (cm^{-1}): 3020,2945,2870(C-H st.) 1592,1472 (C-H bend.), 1620, 1585, 1505 and 779 (8-HQ characteristic bands), 1550,1335(C-NO_2),1240(C-N), 1121(C-O st.).

IV. RESULTS AND DISCUSSION

5-((2-(2-methyl-5-nitro-1H-imidazol-1-yl) ethoxy) methyl) – 8 - hydroxyquinoline (NIHQ) was prepared by condensation of 5-chloromethyl-8-hydroxyquinoline (CMQ) hydrochloride with metronidazole. The resulted NIHQ ligand was an amorphous yellow powder. The C, H, N contents of NIHQ are consistent with the structure predicted (Reaction Scheme). The results show that the metal: ligand (M:L) ratio for all divalent metal chelates is 1:2.

A. FT-IR Spectra

The successful formation of the ligand NIHQ and its metal chelates as per the reaction scheme can be inferred from the appearance of favourable bands on IR spectra. In the spectra of ligand NIHQ, typical C-H stretching and bending vibrations appeared at 3020,2875,1595 and 1475 cm^{-1}. The characteristic 8-HQ bands has been observed at 1625, 1585, 1460 and 750 cm^{-1}. A broad band at 1310 cm^{-1} is attributed to C-O-C linkage of the ligand refering to the succesful linkage of 5-CMQ and Metronidazole. While comparing the IR spectra of ligand NIHQ with those of the metal chelates considerable differences were spotted. The broad band at 3340 cm^{-1} which is attributed to hydroxy stretching of quinoline moiety is absent in IR spectrum of metal chelates suggesting that the phenolic oxygen is involved in chelate formation. Appearance of new bands at 1210 cm^{-1} could be assigned to C-O-M linkage. Also, a medium intensity bands present at ~675 cm^{-1} suggest the M-O linkage and linkage of N of pyridine ring to the central metal ion could be inferred from the sharp bands at ~560 cm^{-1} and ~730 cm^{-1} in the IR spectra of metal chelates[24,25]. In the investigated metal chelates, the bands at around 3000-3600 cm^{-1} (very broad), 1240 ,815 and 700 cm^{-1} are assigned for stretching, bending, rocking and wagging vibration of water molecule and also the presence of rocking vibration indicates the co-ordination of water molecule during chelate formation [26].

B. NMR Spectrum

^1H NMR and ^{13}C NMR data are already mentioned for the ligand NIHQ (compound 3) and are also in good accordance with the proposed structure of the ligand NIHQ.

C. LC–MS Mass Spectra

The recorded LC–MS spectrum and molecular ion peak for ligand (NIHQ) was used to confirm their molecular formula. Peak at 329.37 m/z values represent the molecular ion peak of ligand.

D. Diffuse Electronic Spectra And Magnetic Properties Data

The observed μ_{eff} values in the range 2.52-5.50 B.M are consistent with the above moiety. The value of magnetic moments and reflectance spectral data (table-

1) correlates with the octahedral geometry of all the chelates [27, 28].

Table-1 Diffuse electronic spectra and magnetic properties data

Metal chelates	Magnetic moment μ_{eff} (BM)	Electronic spectral data (cm^{-1})	Transition
NIHQ-Cu^{+2}.2H_2O	2.52	23454	Charge transfer
		15870	$^2B_{1g} \rightarrow {}^2A_{1g}$
NIHQ-Ni^{+2}.2H_2O	3.67	22586	$^3A_{1g} \rightarrow {}^3T_{1g}(P)$
		15370	$^3A_{1g} \rightarrow {}^3T_{1g}(F)$
NIHQ-Co^{+2}.2H_2O	4.60	22728	$^4T_{1g}(F) \rightarrow {}^4T_{2g}(F)$
		15263	$^4T_{1g}(F) \rightarrow {}^4T_{2g}$
		8938	$^4T_{1g}(F) \rightarrow {}^4T_{2g}(P)$
NIHQ-Mn^{+2}.2H_2O	5.50	23857	$^6A_{1g} \rightarrow {}^6A_{2g}\ {}^4E_g$
		18352	$^6A_{1g} \rightarrow {}^4T_{2g}$ (4G)
		16825	$^6A_{1g} \rightarrow {}^4T_{1g}(PG)$
NIHQ-Zn^{+2}.2H_2O	Diamagnetic	-	-
NIHQ-Cd^{+2}.2H_2O	Diamagnetic	-	-

E. Antifungal Activity of Ligand and Metal Chelates

The screening of antifungal activity of the NIHQ ligand and its all metal chelates reveals that the ligand is moderately toxic against fungi while all the chelates are more toxic than the ligand which can clearly be seen from the table-2. Especially, the copper chelate is found to possess the highest antifungal activity.

Table-2 Antifungal activity of ligand and metal chelates:

Ligand and its metal chelates	Zone of inhibition of fungus at 1000 ppm (%)			
	AN	BT	NS	FO
NIHQ	63	64	61	65
NIHQ-Cu^{+2}.2H_2O	78	80	75	79
NIHQ-Ni^{+2}.2H_2O	76	76	72	74
NIHQ-Co^{+2}.2H_2O	75	77	74	78
NIHQ-Mn^{+2}.2H_2O	72	74	75	76
NIHQ-Zn^{+2}.2H_2O	72	75	77	77
NIHQ-Cd^{+2}.2H_2O	73	72	71	75

V. CONCLUSION

A novel ligand is sythezised by condensation of CMQ and metronidazole and its metal chelates were prepared in good yield and duly characterized. In the metal chelates, the ligand coordinates with one central metal atom at four coordination sites, with two water molecules. Structure proposed for the ligand and its metal chelateses are consistent with the results from elemental and spectral analysis. The octahedral geometry of the chelates was confirmed by electronic spectra and magnetic susceptibility measurements. The data provide good evidence of
chelate formation. All the chelates exhibited good antifungal activity.

Conflict of Interest:
None declared

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