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Original Research Article

Chelation Trends and Antibacterial Activity of Some Mixed Ligand Chelates

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Abstract: Synthesis and investigation of new Co(II), Ni(II), Cu(II), Zn(II) and Cr(III) chelates of mixed ligands including aSchiff base [(E)-2-(4-(dimethylamino)benzylideneamino)phenol] (C15H16N2O) derived from the condensation of 4-dimethylaminobenzaldehyde with 2-aminophenol as main ligand (L1) and an amino acid; L-Histidine as co-ligand (L2) were studied. The obtained Schiff base and the mixed ligand chelates were subjected to several physiochemical techniques, in terms of CHN elemental analyses, molar conductivity, magnetic moment measurements, infrared, electronic and mass spectroscopies. The CHN analytical data showed the formation of the Schiff base compound and the mixed ligand chelates in 1:1:1[M:L1:L2] ratio. All the prepared mixed ligand chelates were non-electrolyte in nature. The infrared spectral data exhibited that the used ligands behaving as bidentate ligands towards the metal ions. The 1HNMR spectra of the ligands and their Zn(II) mixed ligand chelate exhibited the effect of the activated groups of the ligands by the metal ion. The electronic spectral results showed the existence of $\pi \rightarrow \pi^*$ (phenyl ring) and $n \rightarrow \pi^*$ (HC=N) and suggested the geometrical structures of the chelates. Meanwhile, the mass spectral data revealed the fragmentations of the Schiff base, Histidine and their Cu(II) mixed ligand chelate. The studies made on these chelates proposed a six coordinated octahedral geometry for all these chelates. The antibacterial activities of the Schiff base, Histidine, metal salts and mixed ligand chelates were screened. It is found that the mixed ligand chelates have the most biological activity in comparison to the free ligands and salts.

Keywords: Schiff base; 4-dimethylaminobenzaldehyde; 2-aminophenol; Histidine; mixed ligand chelates; Physiochemical techniques; Antibacterial activity.

INTRODUCTION

The complexes of Schiff base derived from the reaction of substituted aldehydes with aliphatic and aromatic amines represent a series of compounds containing nitrogen, oxygen and sulphur ligand donor atoms that has been widely reported [1]. Bioactive donor site of N, O and S atoms in organic ligand moieties were widely used in the development of different area such as metal based drugs, medicinal, industrial, analytical, agricultural, biological and clinical. The poly functional donor sites containing Schiff base moieties were extensively used in the field of analytical, biocidal, agrochemical, enzyme models, catalysis, food, chemical and dye industries [2]. The preparation and characterization of mixed ligand complexes derived from 2,6pyridinedicarboxaldehydebis (o-hydroxyphenylimine) or 2, 6-pyridinedicarbox- aldehydebis (p-hydroxy phenylimine) and 2-aminopyridne have been studied [3]. The mixed ligand chelates of Mn(II), Fe(III), Co(II) and Cu(II) ions derived from catechol and 2aminobenzothiazole have been prepared

characterized by using different physical tools. An octahedral geometry was proposed for Mn(II), Fe(III) and Co(II) mixed ligand chelates, whereas, Cu(II) mixed ligand chelate has a square planar geometry. The antibacterial activity of the ligands and their mixed ligand chelates were screened and no effects have been observed. This result suggests the absence of active components or due to the insensitivity of bacteria we assayed [4].

The present investigation aims to prepare and characterize some mixed ligand chelates containing Schiff base formed from [4-dimethyl aminobenzaldehyde with 2-aminophenol] and Histidine with Cr(III), Co(II), Ni(II), Cu(II) and Zn(II) metal ions. Also to study their antibacterial activity against some pathogenic bacteria.

MATERIAL AND METHODS

All chemicals and reagents used in this investigation are of pure grade (BDH or Aldrich), they include; 4-dimethyl amino benzaldehyde, 2-

aminophenol, Histidine, CoCl₂.6H₂O, NiCl₂.6H₂O, ZnCl₂, CrCl₃.6H₂O, DMSO, DMF, NH₄OH, CaCl₂, ethanol and double distilled water. The melting points of the prepared mixed ligand chelates were not corrected. The Schiff base and its mixed ligand chelates were subjected to elemental analysis using 2400CHN elemental analyzer. The molar conductivity of the chelates was measured in DMSO solvent using digital conductivity meter CMD-650, at chemistry department, Benghazi University, Benghazi, Libya. The infrared spectra of the Schiff base and its mixed ligand chelates were carried out applying KBr disc technique using IFS-25DPUS/IR spectrometer. The nuclear magnetic resonance spectra of the Schiff base, Histidine and Zn(II) mixed ligand chelate were recorded on Varian Gemini 200-200MHz spectrometer using TMS as internal standard and d6-DMSO as a solvent. The electronic spectra of the Schiff base, Histidine and their

mixed ligand chelates were measured in DMSO solvent by using Perkin-Elmer lambda-4 β spectro- photometer. The mass spectra were carried out by using Shimadzu QP-2010 Plus. Elemental analysis, infrared, nuclear magnetic resonance, electronic and mass spectroscopies were done at micro-analytical centre, Cairo University, Giza, Egypt.

Synthesis of the Schiff base(L1)

The Schiff base [(E)-2-(4-(dimethylamino) benzylideneamino)phenol],=L1 was prepared by dissolving 4-dimethylaminobenzaldehyde (0.01mole; 1.49g) and same mole of 2-aminophenol in 50mL of ethanol. The mixture was stirred magnetically at 60°C for two hours. Yellow crystals were formed then filtered, washed, dried and recrystallized from hot methanol to give pure yellow crystals in excellent yield of 92.98%. see Scheme (1).

Scheme (1): The Synthesis Route of Schiff base L1

Synthesis of mixed ligand chelates

A general procedure has been adopted for the preparation of chelates in basic media. Solutions of 0.01mole of the salts [CoCl₂.6H₂O; 2.38g, NiCl₂.6H₂O; 2.37g, CuCl₂.2H₂O; 1.71g, ZnCl₂; 1.36g and CrCl₃.6H₂O; 2.67g] in 25mL ethanol were added to the same volume of solutions of 0.01mole of Schiff base -(E)-2-(4- (dimethylamino) benzylideneamino) phenol (L¹) and Histidine (L²). Few drops of ammonium hydroxide(10%) until pH of the solutions becomes 8. The mixtures were refluxed for three hours then allowed to stand then cooled. The obtained chelates were filtered off, washed several times with hot ethanol and dried in desecrator over CaCl₂.

Antibacterial activity assay

All bacteria were obtained and identified from Benghazi medical center, Benghazi, Libya. The bacteria used were *Enterococcus faecalis, Escherichia coli, Pseudomonas aerginosa, staphylococcus aureus and Acinetobacter-SP*. The five species of bacteria were streaked on nutrient agar (OXID England) plates, so that the streaking covered the surface of the plates. The chelates were applied on the streaked nutrient agar plates as a powder (5, 10 and 15 mg), taking an area not more than 6 mm (Size of an antibiotic paper disc) and leaving enough distances between them. The plates were incubated at 37°C for 24 hrs. The inhibition zones were then measured in millimeters and recorded.

RESULTS AND DISCUSSIONS

The reaction between the Schiff base(L^1), Histidine(His) and metal ion is shown in Scheme (2):

In case of $M=Zn^{2+}$, Ni^{2+} , Co^{2+} and Cu^{2+} , X, $Y=H_2O$, when $M=Cr^{3+}$, $X=H_2O$, Y=OH n=2 in case of Ni and Cu chelates, 6 in case of Co, 8 in Zn chelate and 4 in Cr chelate. Chemical equation-1: Reaction between the ligands and metal ions

Scheme (2): The Synthesis Route of mixed ligand chelates

CHN elemental analyses and molar conductivity measurements

The CHN elemental analysis data (Table-1) exhibit the formation of the mixed ligand chelates in 1:1:1[M:L¹:L²] ratio. It was found that the theoretical values are in a good agreement with the found data. The purity of the Schiff base and mixed ligand chelates were

tested by melting points and CHN elemental analyses and mass spectra. The molar conductivity of the mixed ligand chelates were carried out in DMSO solvent and their values which summarized in (Table-1). The low values recorded for metal ion complexes indicate towards non electrolytic behavior [5].

Table 1: CHN elemental analyses, magnetic moments and conductivity of the mixed ligand chelates

Ligand/Chelates	M. Wt	colour	Yield (%)	C% Calc.	C% Exp.	%H Calc.	%H Exp.	%N Calc.	%N Exp.	μ BM	Λ*
Schiff base (L^1) $C_{15}H_{16}N_2O$	240.	Yellow	92.98	75.00	75.76	6.60	4.84	11.67	12.77	-	-
$[CoL^{1}L2(H_{2}O)_{2}].6H_{2}O$	560	Olive gray	77.78	45.00	44.65	4.10	3.45	12.50	11.16	3.22	4.20
$[NiL^{1}L^{2}(H_{2}O)_{2}].2H_{2}O$	487	Lamp black	53.33	51.74	50.77	4.72	3.45	14.37	13.39	4.66	1.53
$[CuL^{1}L^{2}(H_{2}O)_{2}].2H_{2}O$	528	Burnt umber	86.60	47.72	46.57	4.35	3.02	13.25	14.39	2.43	7.88
$[ZnL^{1}L^{2}(H_{2}O)_{2}].8H_{2}O$	602	yellow	68.73	41.86	41.02	3.82	3.65	11.63	13.07	0.00	0.20
$[CrL^{1}L^{2}(OH)(H_{2}O)].4H_{2}O$	517	pale Yellow	72.09	48.74	48.00	4.44	4.20	13.53	11.54	4.53	2.81

 $\Lambda^* = \sigma^{-1} \text{ cm}^2 \text{mol}^{-1}$, BM= Bohr Magneton

Infrared spectra

The infrared spectral data of the ligands and their mixed ligand chelates were listed in Table-2. The presence of water molecules outside the coordination sphere is indicated by a broad band in the region of 3409-3424 cm⁻¹ [6]. The infrared spectrum of the Histidine compound (L2) shows a band at 3017cm-1 corresponding to COO stretching, meanwhile, the NH₂ stretching displays a band at 3400 cm⁻¹, whereas, the first band is disappeared on chelate formation and the second band is overlapped with the bands of water molecules [7]. This means that both groups are

participated in complexation process. On chelate formation the HC=N stretching band (1588cm⁻¹) is shifted to higher frequency of range of 1595-1608 cm⁻¹ indicating the involvement of this group in complexation through Nitrogen atom [8]. New bands in the range of 614-615 and 460-470cm⁻¹ which are not present in the free ligands are assigned to υ (M-O) and υ (M-N) vibrations and the appearance of these bands supports the involvement of Oxygen and Nitrogen atoms of the azomethine(C=N-H) and -OH groups in chelation process [9].

Table 2: Infrared and electronic spectral data of Schiff base, Histidine and their mixed ligand chelates

Ligand / Chelate	υCO O	υОН	υNH ₂	υC=N-H	υM-O	υM-N	nm (cm ⁻¹)
Schiff Base (L ¹)	-	3337	-	1588	-	-	375 (26666)
Histidine (L ²)	3017	İ	3400	-	1	-	435 (22988)
$[Co(L^1L^2)(H_2O)_2].6H_2O$	-	3424	-	1602	615	470	310 (32258), 380 (26316)
$[Ni(L^1L^2)(H_2O)_2].2H_2O$	-	3409	-	1601	614	470	290 (34482), 355 (28169), 425 (23529)
$[CuL^{1}L^{2}(H_{2}O)_{2}].2H_{2}O$	-	3410	-	1595	415	465	425 (23529), 490 (20408)
$[Zn(L^1L^2)(H_2O)_2].8H_2O$	-	3410	-	1608	615	460	344 (42553), 385 (25974), 460 (21739)
$[Cr(L^1L^2)(OH)(H_2O)].4H_2O$	-	3409	-	1607	615	470	480 (20833)

Proton nuclear magnetic resonance spectra(1H-NMR)

In the present investigation, 1HNMR spectra of the ligands and their Zn(II) chelate Figure-1 exhibit signals at 7.50 and 8.70 ppm attributed to -OH and

HC=N groups, respectively. In the [ZnL¹L²] mixed ligand chelate, the change of the position of -NH₂ and HC=N groups indicates their participation in bonding with the metal ions, whereas, the hydroxyl group is disappeared on Chelation [10].

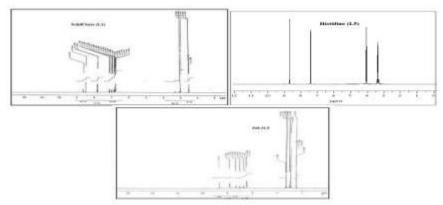


Fig-1: ¹HNMR of ligands, Schiff base and ZnL1L2chelate

Mass spectra

The mass spectral fragmentations of the Schiff base, Histidine and their Cu(II) mixed ligand chelate are given in Table-3, Figure-2. The peak at m/e⁺= 238 assigned to $C_{15}H_{14}N_2O+$. Whereas, another peak at m/e⁺=195 is corresponding to the formula of $C_{13}H_9NO^+$. The formula of $C_{12}H_7O^+$ is analogous to peak at m/e⁺= 167. Meanwhile, the last peak at m/e⁺= 64 is attributed to this formula $C_5H_4^+$. The base peak of Histidine at m/e⁺= 155 is attributed to the original molecular weight of the compound. The peak at m/e⁺=110 analogous to $C_6H_8NO^+$. The same

spectrum shows a peak at m/e $^+$ = 82 due to the formula of $C_5H_8N^+$. The structure of $C_3H_4N^+$ is assigned to the peak at m/e $^+$ = 54. Meanwhile, the peak of m/e $^+$ = 28 is attributing to this formula (CH $_2N^+$). The mass spectrum of the copper(II) mixed ligand chelate displays a molecular ion peak at m/e $^+$ = 395 which is corresponding to [Cu(C $_19H_16N_4O_2$)] $^+$. The peak at m/e $^+$ =237 is due to [Cu(C $_6H_4NO_2$)] $^+$. Whereas, the peak at m/e $^+$ =185 is analogues to [Cu(C $_6H_4NO_2$)] $^+$. The same mass spectrum displays a peak at m/e $^+$ =144 which can be to [Cu(C $_4H_3NO_2$)] $^+$ The final peak in the spectrum of the chelate appeared at m/e $^+$ =55 is due to C $_3H_3O^+$.

Table 3: Mass spectral fragmentation of Schiff base, Histidine and their Cu(II) mixed ligand chelate

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Compound	Fragmented ions	m/e ⁺ values
	$C_{15}H_{16}N_2O$	240
	$C_{15}H_{14}N_2O^+$	238
$C_{15}H_{16}N_2O$ (L1)	$C_{13}H_9NO^+$	195
	$\mathrm{C_{12}H_7O^+}$	167
	$\mathrm{C_5H_4}^+$	64
	$C_6H_9N_3O_2$	155
CHNO (12)	$\mathrm{C_6H_8NO}^{\scriptscriptstyle +}$	110
$C_6H_9N_3O_2$ (L2)	$\mathrm{C_5H_8N}^+$	82
	$\mathrm{C_3H_4N^+}$	54
	$\mathrm{CH_2N}^+$	28
	$[Cu(C_{15}H_{16}NO)(C_6H_9NO_2)(H_2O)]2H_2O$	528
	$[Cu(C_{19}H_{16}N_4O_2)]^+$	395
	$[Cu(C_9H_6N_2O_2)]^+$	237
[CuL1L2(H2O)2].2H2O	$\left[\mathrm{Cu}(\mathrm{C_6H_4NO_2})\right]^+$	185
	$[Cu(C_4H_3NO)]^+$	144
	C_3H_3O	55

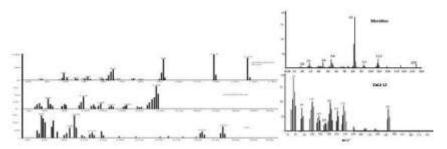


Fig-2: Mass spectra of Schiff base, Histidine and Cu(II) mixed ligand chelate

Electronic spectra and magnetic moment of the mixed ligand chelates

An electronic spectra for all the synthesized chelates were recorded in DMSO solvent. The electronic spectra of theL¹, L² and their mixed ligand chelates are shown in Figure-3. The electronic spectral data of Co(II), Ni(II), Cu(II), Zn(II) and Cr(III) mixed ligand chelates of with Schiff base (L1) and Histidine (L2) ligands are listed in Table-2. The electronic spectra of the Schiff base and Histidine display bands due to $\pi \rightarrow \pi^*$ (phenyl rings) and $\pi \rightarrow \pi^*$ (HC=N and -OH) transitions [11]. The Co(II) mixed ligand chelate exhibits two bands at 310nm (32258cm⁻¹) and 380 nm (26316 cm-1)assigned transfer, and ${}^{4}T_{1}g(F) \rightarrow {}^{4}T_{2}g(F)$ transitions, respectively. The strength of the obtained bands and the magnetic moment (4.98BM) confirm the existence of an octahedral geometry of the Co(II) chelate [12]. The electronic spectrum of Ni(II) chelate shows three bands; 290nm (34482cm-1), 355nm(28169cm⁻¹) and 425nm (23529cm⁻¹) corresponding to Ligand field, charge transferand $3A2g(F) \rightarrow 3T1g(F)$ transitions.

magnetic moment of the chelate (2.85BM) and the nature of the bands support the existence of an octahedral geometry of the Ni(II) chelate [13]. For Cu(II) chelate, the electronic spectrum shows two bands; 425nm (23529cm⁻¹) and 490nm (20408cm⁻¹) corresponding to charge transfer and ${}^{2}Eg \rightarrow {}^{2}T_{2}g$ transitions. Based on the magnetic moment (1.65BM) and the intensity of the bands, suggesting an octahedral structure for this chelate [14]. The electronic spectrum of Zn(II) chelate exhibits three bands; 344nm (42553cm⁻¹), 385nm (25924cm⁻¹) and (21739cm⁻¹) attributed to the existence of charge transfer, transitions. The intensity of the bands and the diamagnetic character of the chelate suggest the octahedral structure [15, 16]. The electronic spectrum of Cr(III) chelate of $[Cr(L^1L^2)(OH)(H_2O)]8H_2O$ exhibits one band at 480nm (20833cm⁻¹) due ${}^{4}A_{2}g(F) \rightarrow {}^{4}T_{2}g(F)$ transition. The magnetic moment of this chelate (4.53BM) and the nature of the mentioned band support the presence an octahedral geometry [15-171.

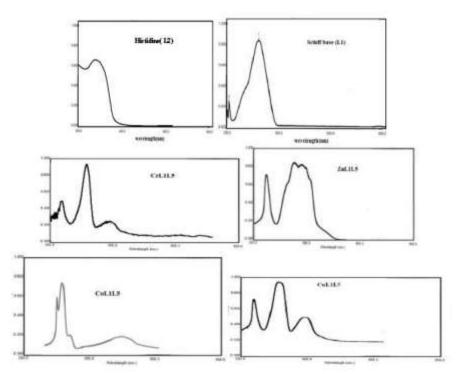


Fig-3: Electronic spectra of Schiff base, Histidine and their mixed ligand

Antibacterial activity

The Schiff base (L¹), Histidine (L²) and the mixed ligand chelates were screened for their antibacterial activity Table-3, figure 4. No effect was observed for both Schiff base and Histidine on the five types of tested bacteria. Whereas, the effect of all mixed ligand chelates was observed. Most mixed ligand chelates were resisted by *Enterococcus faecalis* and *Escherichia coli* except [NiL¹L²(H₂O)₂]2H₂O mixed ligand chelate had moderately effective, although the effectiveness of copper chelate of the formula

 $[CuL^1L^2(H_2O)_2].2H_2O$ was highly activate against Pseudomonas aerginosa screened in inhibition zone 38mm, on the contrary, $[CrL^1L^2(OH)(H_2O)]4H_2O$ mixed ligand chelate did not effect on the same organism. But, the other mixed ligand chelates of $[CoL^1L^2(H_2O)_2].6H_2O$ and $[ZnL^1L^2(H_2O)_2].8H_2O$ have moderate effect on staphylococcus aureus bacteria.

Most complexes have been observed that the metal complexes have a high activity than same metal salts. Coordination reduces the polarity of the metal ion mainly because of the partial sharing of its positive charge with the donor groups within the chelate ring system [18, 20] and can be explained on the basis of Tweedy's chelation theory [16, 21].

Table 3: Biological	activity of t	the mixed	mixed ligand	chelates	(mm)

	Inhibition Zone (mm)						
No.	Compound	Acinato	E. coli	E. feacals	P. aerginosa	S. aureus	
1	Schiff base (L ¹)	0	0	0	0	0	
2	Histidine (L ²)	0	0	0	0	0	
3	CoCl ₂ .6H ₂ O	18	34	11	16	25	
4	NiCl ₂ .6H ₂ O	8	29	9	13	28	
5	CuCl ₂ .2H ₂ O	7	15	5	10	15	
6	$ZnCl_2$	19	20	7	17	20	
7	CrCl ₃ .6H ₂ O	20	21	22	19	21	
8	$[Co(L^1L^2)(H_2O)_2].6H_2O$	8	0	0	10	14	
9	$[Ni(L^1L^2)(H_2O)_2].2H_2O$	30	17	11	11	22	
10	$[CuL^{1}L^{2})(H_{2}O)_{2}].2H_{2}O$	15	0	0	38	12	
11	$[Zn(L^{1}L^{2})(H_{2}O)_{2}].8H_{2}O$	23	5	5	18	15	
12	$[Cr(L^1L^2)(OH)(H_2O)].4H_2O$	26	7	16	10	9	

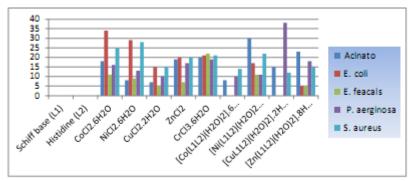


Fig-4: The (ZI) mm of compounds

CONCLUSION

The obtained Co(II), Cu(II), Zn(II), Ni(II) and Cr(III) mixed ligand were prepared and characterized by using several physiochemical tools, in terms CHN elemental analyses, molar conductivity, magnetic moments, infrared, proton nuclear magnetic resonance, electronic and mass spectroscopies. All the experimental data confirm the existence of an octahedral geometry for all chelates. All the complexes are non-electrolyte. Based on the reported data, it may be concluded that L-Histidine L^2 , coordinate to metal ions as bidentate ligand through (O) atom of (COO-) group and (N)atom of (NH₂) group in L-Histidin while L^2 coordination of the metal ion occur through the (O)atom of (OH) group and (N)atom of (C=N-H)group [18, 19].

The antibacterial activity results showed the presence of an effect for the ligands and chelates on the tested bacteria. Based on the physiochemical data. The geometrical structures as shown in the chemical [equation-1Scheme (2)] agreed with the obtained data cited in the text.

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