Synthesis, Characterization and Antibacterial Evaluation of Mixed Ligand Complexes of 2-phenyl-2-(o-tolylamino) Acetonitrile and Histidine ligand with Co, Cu, Zn divalent Metal Ion Complexes

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Abstract

Anew mixed compound complexes derived from 2-phenyl-2-(o-tolylamino) Acetonitrile as primary ligand (L1) and histidine (L2) as secondary ligand have been prepared and characterized by conventional techniques, elemental microanalysis (C.H.N), Fourier transform infrared, ultra violet-visible spectra, , flame atomic absorption, molar conductivity, magnetic susceptibility measurement and ^{1}H-NMR spectra. From IR data which appear chelating behavior of the amino acid ligand (L2) toward transition metal ions is via carboxylate oxygen, amino nitrogen and imidazol nitrogen as tridentate ligand while second ligand (L1) chelating through N-nitrile and N-aniline, according to all above technics the octahedral shapes were expected for these complexes as proposed. The molar conductivity confirms the non-electrolytic nature of both cobalt(II),copper(II) and Zinc((II) complexes. Accordingly the suggested formula of cobalt, copper and zinc complexes could be [CoClL1L2].H2O, [CuClL1L2] and [ZnClL1L2] respectively.

Keywords: *mixed ligand, histidine.*

1. Introduction

The consistency of amino acids with various elements very important subject of argumentation since a long time ago, and the thinking of getting the linkage modes was difficult to know especially with Histidine which have bigger side chain [1], related to various kinds of giver atoms existing in amino acid structure. For this reason, it became very important to learn its effective side and binding similarity to transition metals together theoretic and practical terms. In between all amino acids existing in natural surroundings, Histidine(His) as a ligand usually present in many forms of metalloenzymes for this reason it is the main amino acids remains in various reactions of enzyme [2] this in turn comes back to its stereo chemical site of the managing atom in this compound which is essential surrounds the imidazole side series containing double N_2 atoms able of contributing in metal-ligand organization spheroid thus it was gifted to accepted several metal-bound formulae in proteins. Therefore, it is necessary to recognize the coordination modes of the (His) to recognize the reaction mechanism of metalloenzymes [3]. In order for more expanded concept about relations between metal ions with big molecules containing amino acids, understanding about arrangement and the active metal ions direction to amino acids big molecules are necessary.

2. Experimental

Reagents and Chemicals

All the substances and diluters were employ for the production were of chemical position and were gained commercially from British Drug House (BDH) Company using an exclusion of cobalt chloride hexahydrate, copped chloride dehydrate and zinc chloride salts were gained from Fluka Company. The acetonitrile ligand which was prepared, production and characterized under to this published work [4, 5].

2.1 Synthesis of mixed ligand complexes

A mixture of (His) (L2) ligand which was prepared by dissolving it in equimolar sodium hydroxide. The α-amino nitrile 2-phenyl-2-(o-tolylamino) acetonitrile (L1) can organized through melting it in C_2H_5OH. The solutions of metal ions Co^{+2}, Cu^{+2} and Zn^{+2} were preparing in an equal amount of HCl acid. To make metal complex, both melted ligands L1 and L2 were combine by 3 ml of 0.1 mol/L solutions of metal ion in 1:1:1 molar percentage at natural heat and the *p*H >7 with overall size fit for ten ml(adding distilled water). The important note is that the mix beginning with adding of L2 to metal ion solution after that addition of L1. The *p*H of mixture was adjusted by added of sodium hydroxide solution. The reactions color of solution was variation and refluxed for four hours, the color of precipitation was placid by filtration and wash away a number of times with cold ethanol-water 50٪ combination then become dry in the oven at 50 ˚C for one day[6,7].

3. Result and Discussion.

3.1 Conductivity measurement

The physical properties of the three synthesizes complexes are tabulated in tables [1], and they in a good agreement with the suggested formula of cobalt, copper and zinc complexes. The molar conductance of 0.001 M solution of the complexes in absolute ethanol was measured at 25˚C.The conductivity values indicating the non-electrolytic behavior of the complexes.

Table [1]: Physical data of the mixed ligand complexes

Compound	Yield percentage	Color	Melting point	Molar conductivity $\Lambda m\ \Omega^{-1}\ cm^2\ mol^{-1}$
$C_{15}H_{14}N_2$	79%	yellow	202-206	---------
$C_6H_9N_3O_2$	---------	white	122-124	---------
[CoClL1L2].H2O	77%	brown	100-104	12.10
[CuClL1L2]	81%	black	203-205	8.71
[ZnClL1L2]	70%	Off-white	188-192	13.25

3.2 Microanalysis:

The α-aminonitrile compounds was very benefit because of their flexibility as Original materials in the production of several compounds. Ligands(L1 and L2), side by side with complexes, were recognized by C.H.N (Table 2), FT-IR (Table 3), UV-Vis (Table 4), ^{4}H-NMR (Table 5) .

Table [2]: Elemental micro analysis of the ligand (L_1 and L_2) and their prepared complex

Comp.	FORMULA M.Wt $g.mol^{-1}$	Micro elemental analysis calc. (Found)					
		C%	H%	N%	M%	Cl %	other
L_1	$C_{15}H_{14}N_2$ (222,0)	65.27 (65.00)	3.42 (3.27)	7.11 (7.08)	---------	-------	

L_2	$C_6H_9N_3O_2$ (155.16)	46.45 (46.40)	5.85 (5.81)	27.08 (27.00)	---------	20.68 (20.55)	**O_2%**
C_1	[CoClL1L2].H2O (489.09)	45.52 (43.90)	3.62 (4.32)	4.96 (4.81)	11.37 (10.96)	12.90 (12.48)	
C_2	[CuClL1L2] (475.7)	38.48 (37.16)	4.99 (3.96)	4.74 (4.03)	7.78 (7.10)	17.15 (16.78)	
C_3	[ZnClL1L2] (477.54)	49.76 (48.05)	4.43 (4.12)	4.17 (3.18)	18. 75 (17.39)	11.69 (10.00)	

3.3 Infrared Spectral Data

The coordination with α-amino nitrile (L1) was expected through amine (-NH-) and nitrile (-C≡N) groups which appeared at the region (3300-3500) cm-1 and (2242-2264) cm-1 respectively [8], both vibrations were shifted to higher energy field, which represented the success of bonding with the metal ion as primary ligand in addition the appearances of other related groups like out of plain deformation. All the mentioned data represented as evidence for the coordination bonding between the reactants. Table [3] shows the related data in addition to some other characteristic bands. In general amino acid -NH3+ group appear in the range (3130-3030) cm-1[9]. from infrared band of histidine (L2) which was appeared near 3072 cm-1 but similarities with anther vibrations like νNH (imidazole group) and νCH2 group of heterocyclic near 3016 cm-1[10]. In theory νNH3+ became die out and disappeared on coordination that it eventually converted to amino group (-NH2) in the proper basic medium that normally accurse as two bands in the region 3300-3500 cm-1. However, in metal complexes some band appears in the same region near 3300 cm-1, which should be stand up from other vibrations looking in same place belong to amino group (-NH-) of α-amino nitrile (L1) as one band. Therefore, the comparison of shifting of histidine amino group was discard and out off our account ,but carboxylate group shifting was accounted, that the νasyCOO- and νsyCOO- absorptions appear at 1634 and 1414 cm-1 respectively which are shifted to lower or higher frequencies[10,11]

Table[3]: Infrared frequencies of L1and L2 ligand and their complexes with Co(II) , Cu(II) and Zn(II) ions

Comp	υ (C≡N)	υ (N-H) amine	υ(C=N) imidazole	υ(COO)asy	υ(COO)sy	υM-O	υM-N
L_1	2188w	3368m	--------	-----	-----	-----	-----
L2	-----	-------	1568m	1634m	1414m	----	----
C_1 (Co)	2242m	3350m	1539w	1625w	1385w	443w	549s
C_2 (Cu)	2210m	3326w	1542m	1590s	1367w	448m	550s

C_3 (Zn)	2264s	3353w	1547m	1620vs	1385s	455m	559w

3.4 Ultraviolet-visible spectroscopy

The histidine, α-amino nitrile and the complexes show $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ bands in the region (344-230) nm overall. The spectrum of the cobalt complex was presented three transfers detected at (947, 711 and 532) nm in that order, these bands possible related to the shifts $^4T_1g(F) \rightarrow {}^4T_2g(F)$, $^4T_1g(F) \rightarrow {}^4A_2g$ and $^4T_1g(F) \rightarrow {}^4T_1g(P)$ individually. The magnetic moment value (4.13) B.M was in agreement with roles of octahedral arrangement [12]. The electronic band of copper compound, was shown interest group at (656) nm belong to $^2Eg \rightarrow {}^2T_2g$ transferal showing octahedral geometry[13] so that this complex presented magnetic moment 1.80 B.M. because of existence of alone unpaired electron, which gives probability of an octahedral structure[14]. The Zinc(II) complex was diamagnetic. In accordance with Empirical formulae, an octahedral shapes were suggested for these complexes [15].

Table [4]: Electronic spectral data of L1and L2 ligand with their complexes in absolute ethanol

Comp.	Assignments	Wavelength λ (nm)	Geometry Suggested	Meff (B.M)
L_1	$n \rightarrow \pi^*$ $\pi \rightarrow \pi^*$	344 285	---------	----
L_2	$n \rightarrow \pi^*$ $\pi \rightarrow \pi^*$	270 230	---------	----
C_1 Co(II)	$^4T_1g(F) \rightarrow {}^4T_2g(F)$ $^4T_1g(F) \rightarrow {}^4A_2\,g(F)$ $^4T_1g(F) \rightarrow {}^4T_1g(P)$	947 711 532	Octahedral	4.13
C_2 Cu(II)	$^2Eg \rightarrow {}^2T_2g$	656	Octahedral	1.80
C_3 Zn(II)	C.T	374	Octahedral	Diamagnetic

3.5 ^{1}H-NMR Spectra

The initial ligand (L_1) was described by ^{1}H-NMR spectroscopic techniques, besides the complexes from C_1 to C_3 in the present of dimethyl sulfoxide (**d6**) as a solvent as found in the Tables (5). The ^{1}H-NMR range for the first ligand (L_1) was appeared many signal; the First appeared at (2.23) ppm this be present to (-C$\underline{H}_3$) protons, additional occurred at (2.63) ppm this peak may be referred to the solvents peak , also another one located at δ(4.32) ppm back to the (-N-$\underline{H}$) proton but a quarter looked at δ(5.92) ppm that was agreed with (-C$\underline{H}$C≡N). The final peak which related to the aromatic protons seemed at (6.77- 7.98) ppm [16], however in (his) (L_2) was displayed peaks acted at (7.58–8.83) ppm be located to the aromatic protons[17]. In all the spectrum of complexes there were some similarity between them and the ligands [L_1 and L_2], the lone disagreement were presented at (-N-$\underline{H}$) of the ligand was removed From these complexes and the motion of (-C$\underline{H}$-C≡N) was moved in complexes provided a hint to the formation of complexes

Table [5] :^{1}H-NMR (L_1 and L_2) and their metal ion complexes

Comp.	Formula	Groups	Chemical Shifts δ(ppm)
L_1	$C_{15}H_{14}N_2$ (222,0)	-C<u>H</u>$_3$ (-N<u>H</u>) (-C<u>H</u>-C≡N) (Ar-<u>H</u>)	2.23 4.32 5.92 6.77-7.98
L_2	$C_6H_9N_3O_2$ (155.16)	(Ar-<u>H</u>)	7.58–8.83
C_1 Co(II)	[CoClL1L2].H2O (489.09)	-C<u>H</u>$_3$ (-N<u>H</u>) (-C<u>H</u>-C≡N) (Ar-<u>H</u>)	2.10 4.42 5.81 6.73-7.87
C_2 Cu(II)	[CuClL1L2] (475.7)	-C<u>H</u>$_3$ (-N<u>H</u>) (-C<u>H</u>-C≡N) (Ar-<u>H</u>)	2.31 4.53 5.61 6.92-8.43
C_3 Zn(II)	[ZnClL1L2] (477.54)	-C<u>H</u>$_3$ (-N<u>H</u>) (-C<u>H</u>-C≡N) (Ar-<u>H</u>)	1.86 4.13 5.99 6.23-8.33

4. Antibacterial Evaluation

Many Microorganisms may be responsible for different types of diseases to human and animals. Finding of chemotherapeutic agents gave very essential character in the way of regulatory and stopping such diseases especially those of mixed ligand complexes. The antibacterial activities of both (L_1 and L_2) and ternary complexes beside *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* are pointed in Table [6]. were separated and recognized, to be worked in two *in vitro* techniques at minimal inhibitor concentration (MIC), the first was disk-diffusion method and the second was well diffusion method. Tetracycline anti biotic was used for comparison and dimethyl sulfoxide was used as solvent for all the tested mixtures. The antibacterial results were not promising. In all results, founded, the complexes ion gave an active better than amino nitrile (L_1) because of chelation [18], this was responsible for reducing of the polarization of the metal atom, essentially for the reason that of fractional division of its favorable charge with giver set of the ligand and present of probable π-electron delocalization on the aromatic rings, for all this reason the lipophilic character was increased, favoring its infusion into the bacterial wall, leads to the loss of living organisms[19,20].

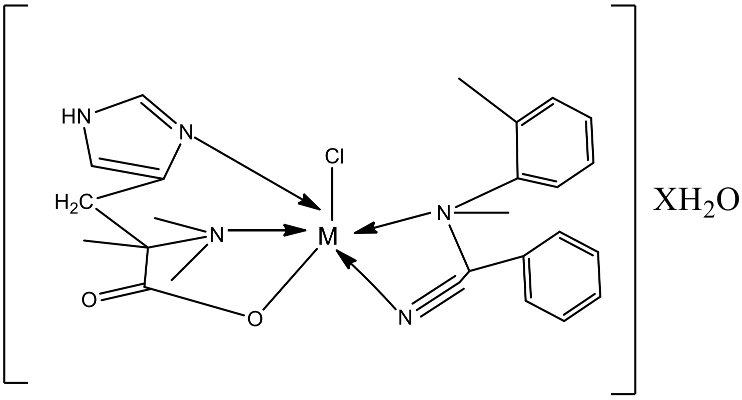
Table [6]: Antibacterial activity of tetracycline as standard and mixed ligand Co(II) ,Cu(II) and Zn(II) complexes in DMSO

Compound	Inhibition zone (mm) *Escherichia coli*	Inhibition zone (mm) *Pseudomonas aeruginosa*	Inhibition zone (mm) *Staphylococcus aureus*	Inhibition zone (mm)*Bacillus subtilis*
Tetracycline	----	----	14.4	----
L_1	12	----	15	10
L_2	------	18	26	16

C_1	33	20	29	28
C_2	54	30	30	26
C_3	20	22	16	33

Conclusion:

According to the suggestions acquired from overhead readings propose an octahedral structure for Co, Cu and Zn complexes with tridentate histidine ligand and the other acetonitrile as abidentate ligand. The molar conductivity of complexes has non- electrolytic shape of all complexes. Through our result we founded that complexes was consider to be further activation than L1and L2 against all the microorganisms that was using, pointing to the organization of the ligands to Co(II), Cu(II) and Zn has been improved its antimicrobial activity, while the second L2 was offered very good inhibitor factor in the direction of all bacterial further than L1 but fewer than complexes this compatible with that management (L1and L2) with metal ion enhanced thier antimicrobial Activity. The suggested structure for the prepared complexes could be in octahedral geometry and general formula be [MClL1L2] H2O, M: Co or [MClL1L2] M: Cu, Zn for copper and Zn complexes respectively as shown in figures below.



M: Co, X: 1, [MClL1L2] XH$_2$O
M: Cu, Zn, X: 0, [MClL1L2]

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