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Mixed Ligand Complexes of Schiff Base and Nicotinamide: Synthesis, Characterization and Antimicrobial Activities

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Abstract. A new Schiff base of HL has been synthesized from amoxicillin drug and 4-Chlorobenzophenone. Cr (III), Fe (III), Co (II), Ni (II), Cu (II), Cd (II) and Hg (II) mixed ligands complexes of Schiff base and Nicotinamide. Diagnosis of synthesis ligand and its complexes are done by ¹H NMR, ¹³CNMR and thermal analysis for HL ligand, FTIR, UV-visible, molar conductance, CHN analysis, magnetic susceptibility and atomic absorption. Octahedral geometries have been suggested for all complexes. All compounds under study were tested antimicrobial activity against four type of bacteria such as Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus Bacillis subtilis in nutrient agar.

Keywords. Amoxicillin, Nicotinamide, geometries, Antibacterial activities

1. Introduction

Complexes with Schiff bases as ligands attract a lot of researchers because of their wide application in analytical chemistry, dye industry, medicinal chemistry, food industry, catalysis, etc. and also for their simple methods to be synthesis. Many studies mention, varied complexes of these compound against different antibiotics and their biological activity [1-8]. In all these cases it was found that metal complexes showed increased antimicrobial activity than their parent Schiff bases. Amoxicillin "(2S,5R,6R)-6-[(2R)-2-amino-2-(4-hydroxyphenyl)acetyl]amino]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid” is a member of penicillin’s group which are a very important class of β-lactamic antibiotics used to treat bacterial infections caused by microorganisms and it's a chemicals produced by different types of microorganisms and other living systems that are capable of small concentrations inhibit the growth or killing of bacteria and other microorganisms.

Cu, Co and Zn complexes of amoxicillin and pyrrol-2-carbaldehyde as a Schiff base have been characterized by analytical techniques and tested for their in vitro antibacterial [11]. Five mixed ligand complexes synthesized from 4-dimethylaminobenzaldehyde and Amoxicillin (Schiff base) with nicotinamide was characterized. These complexes were tested against bacterial and comparison with the Schiff base [12]. Co and Ni complexes of ligand derived from salicylaldehyde & amoxicillin were synthesized [13]. Four complexes were prepared from amoxicillin and nicotinaldehyde with some metal ions such as; Co²⁺, Ni²⁺, Cu²⁺, and Zn²⁺[14]. Mixed ligand complexes was synthesized by the reaction of amoxicillin and boric acid (Schiff base) with its Co (II), Ni (II) and Cu (II) with 4-aminoantipyrine
In this research, theoretical, experimental and biological studies of ligand Schiff base complexes derived from Amoxicillin have been studied [16].

2. Experimental Section

2.1. Chemicals and Instrumentation
In general, chemical reagents and solvents have been in a analytical grade and employed without any additional distillation. By Stuart Melting Point Apparatus, melting points have measured, CHNS analysis was executed by using Euro EA 3000. The spectrums of IH NMR have been read by means of Brucker DRX system 500 (500 MHz) as well as 13 CNMR hetero nuclear correlation spectroscopy (COSY). The corresponding spectrums of UV-Vis have measured by Shimadzu UV (160A) Ultra Violet-Visible Spectrophotometer and FTIR spectrums have gotten from Shimadzu, FTIR (8400S) with KBr discs under 4000- 400 cm\(^{-1}\) range. The complexes and their metal substances have examined via Shimadzu AA (620G) atomic absorption spectrophotometer. Magnetic measured results have been measured via Bruker BM6 instrument at 298K based on Faraday’s method.

2.2. Synthesis of 6 - [2-{{(4-Chloro-phenyl)-phenyl-methylene}- amino}-3, 3-dimethyl-7-oxo-4-thia-1-aza-bicyclo[3.2.0]heptane-2-carboxylic acid (HL)
A solution of Amoxicillin .3H2O (1mmole, 0.419 gm) and (1mmole, 0.056) of KOH in 20 ml of methanol, were added to a solution of 4-chlorobenzophenone (1mmole, 0.216 gm) in methanol (10 mL). The mixture was refluxed about 4 hours with stirring. The outcome has been an orange solution allowable to be cooled and dried under room temperature. After that, it was re-crystallized to ethanol. The white colour solid mass made throughout refluxing has been cooled under room temperature, filtered and washed absolutely by methanol, washed by warm acetone and recrystallized from acetone for acquiring the purified sample, fig.1.

2.3. Synthesis of complexes (Mixed)
HL Solution of HL (0.564 gm, 1m mole) and Nicotinamide (0.366 gm, 3m m ole) in methanol (10mL) have been inserted to the stirring solution of metal chloride 1m mole in methanol 5 ml. The subsequent combination has been heated under reflux for one hour. At that point, the mixture has been precipitation, washed in ethanol and dehydrated under room temperature for one day.

3. Results and Discussion

3.1. Some Physical Properties of the Mixed Complexes [17, 18]
Generally, all complexes were found to be non-hygroscopic solids, and soluble in DMF and DMSO, but insoluble in water. The CHN analysis and the sensible features of the compounds are listed in table (1). The complexes can be symbolized as [M(L)(Nam)\(_3\)]Cl\(_2\) and [M(L)(Nam)\(_3\)]Cl where M\(^{3+}\) (Cr & Fe) and M\(^{2+}\) (Co, Ni, Cu, Cd & Hg) ions Schiff base ligand (HL) , and Nicotinamide (Nam).
Table 1: Some of the physical properties of the prepared compounds

| The molecular Formula | code | M. wt Calc. | Colour & Yield | M. p (Dec) | $\Delta m$ | Metal theory% (exp)% |
|-----------------------|------|-------------|----------------|-----------|-----------|---------------------|
| [Cr(L)(Nam)$_2$Cl]$_2$ | C1   | 1052.44     | Brown, 80      | 150       | 77.5      | 4.94(4.64)          |
| [Fe(L)(Nam)$_2$Cl]$_2$ | C2   | 1056.29     | Brown, 77      | 180       | 72.9      | 5.29(4.99)          |
| [Co(L)(Nam)$_2$Cl]    | C3   | 1023.88     | Blue-green, 85 | 258 Dec   | 33.4      | 5.76(6.78)          |
| [Ni(L)(Nam)$_2$Cl]    | C4   | 1023.66     | Green, 69      | 144       | 37.2      | 5.75(5.74)          |
| [Cu(L)(Nam)$_2$Cl]    | C5   | 1028.49     | Green, 74      | 150       | 31.8      | 6.18                |
| [Cd(L)(Nam)$_2$Cl]    | C6   | 1077.36     | White, 83      | 200       | 38.1      | 10.43(11.14)        |
| [Hg(L)(Nam)$_2$Cl]    | C7   | 1165.54     | Yellow, 88     | 270 Dec   | 33.8      | 17.21(16.89)        |

3.2. $^1$H NMR

$^1$H NMR spectrum of the ligand (HL) fig.1, the multiplied signals of the aromatic protons were appointed in range $\delta$ (6.68-6.72) ppm. The peaks were appeared as singlet at $\delta$ (1.41 & 3.26) ppm asecribed to (-CH$_3$ and -S-CH$_2$-) groups, 7.05 ppm and 8.43 ppm (m, N=CH proton and the resonance of the aromatic ring protons, 13 H); The signal characteristic of the proton of the (N=CH) group at some compounds was obscured by the multiple signals of aromatic protons and the hydroxyl proton with intermolecular hydrogen band resonates at the lower field. [19]. The formation of Schiff base is supported by the presence of a singlet signal at $\delta$ 8.43ppm (s, azomethine proton 1H), the protons on the $\beta$-lactum ring were appeared at $\delta$ (4.81-5.00) ppm. The signal noticed at $\delta$ (7.05) ppm represent to proton of the amide. NMR spectral data of HL was compared with the spectral data for the similar ligands reported in literatures [20].

![Figure 2. The $^1$H NMR spectrum for HL ligand](image)

3.3. $^{13}$CNMR

All the carbons atoms in the structure are established to be in their predictable regions [19]. The spectrum of the ligand HL shown in fig.3. Chemical shift of C6, C7 and C8 $\beta$-lactam at (139.24, 138. 25 and 137.26) ppm and 195.14 ppm for Schiff base group. Five resonance at (134.51, 133.10, 131.20, 130. 33 and 130. 25) ppm confirmed to carbon atoms of aromatic rings [21].
3.4. Thermal analysis of HL ligand

TG-DTA analysis of the ligand (HL) was carried out under Helium inert gas in the rete 20 mL / min performed from (20 – 900) °C. Thermal analysis has to be helpful in determining the (H2O) content in compounds and their thermal stability and decomposition mode under a controlled heating rate [22]. The TG/DTG curve of the HL Fig.3 and table (2) showed position three decomposition steps. The first step occurred at the temperature range 25-100 at peak temp 71.48 °C as is demonstrated by DTG curve mass loss 5.36 % (calcd. 5.48%), the weight loss of this step corresponds to the elimination of solvent molecules solvent (methanol). The second step occurred at the temperature range 150-290 at peak temp 199.83 °C mass loss 4.70 % (calcd. 4.75%), the slow decomposition in second step from 150-280°C with mass loss 60.20%, (calcd.62.05%), corresponds to removal of coordinated part of ligand (-H2O+4H2). The third step occurred at the temperature range 610-780 at peak temp 592.53°C mass loss 14.22 % (calcd. 14.62%), corresponds to (-H2O+Cl+3H2) stoichiometry of end product. [23]

Table 2. Thermal decomposition of HL

| Compounds        | Temp. range of Deco. °C | Peak Temp. of TG °C | % Wt. Loss (TG) |
|------------------|--------------------------|---------------------|-----------------|
| C29H26ClIN3O5S   | 25-100                   | 71.48               | 5.36 (5.48)     |
| -CH3OH           | 150-280                  | 199.83              | 4.70 (4.75)     |
| -H2O+4H2         | 610-780                  | 592.53              | 14.22 (14.62)   |
| -H2O+Cl+3H2      |                          |                     |                 |

3.5. FT-IR spectra

Table (3) shows the FT-IR spectra for HL and (Nam) ligands and its complexes. The spectrum of the ligand Nam exhibits a strong bands at (1699) cm⁻¹, was assigned of ν(CO) [24]. All data for coordinated
(Nam) characteristics through ring (N) were (C-N) str (py) for free ligand shifted lower frequencies in the spectra all complexes. Sharp band of HL ligand at (1666) cm\(^{-1}\) attributed to the stretching vibration of the \(\nu(\text{HC=N})\). On complexes this band was shifted to (1616-1651) cm\(^{-1}\) for all complexes, this indicating coordination the nitrogen atom with the metal ion [25]. In complexes the bands of asymmetric and symmetric for COO group stretching vibration for the group within the range (1597-1620) cm\(^{-1}\) and (1396-1377) cm\(^{-1}\) respectively, the large values more than 200 cm\(^{-1}\) from difference between the COO (asy. and sym.) shows the monodentate binding of the carboxylate group. The stretching vibration band at (1685) cm\(^{-1}\) is ascribed to \(\nu\) (C=O) for \(\beta\)-Lactam group, this band has been shifted to lower frequencies at (1674, 1670, 1670, 1674, 1674, 1666 and 1670) cm\(^{-1}\) for seven complexes respectively, indicated that the coordination is through the (Oxygen for \(\beta\)-lactam group) [26]. In the lower frequencies region a new weak bands observed at (478-451), (420-586) cm\(^{-1}\) and (513-528) cm\(^{-1}\) have been appointed, to coordinated the, \(\nu\) (M–O) and \(\nu\) (M–N) vibrations, respectively. Accordingly, one can deduce that the primary ligand (HL\(_2\)) binds the metal ion as tridentate monobasic type (NOO -) donors while the secondary ligand (Nam) binds the metal ion as mono dentate donors via (N) atom. [27, 28]

| compounds | O-H | (C-H) | (C=O)\(_{\text{lactam}}\) | (C=N)\(_{\text{Schiff}}\) | COO | \(\Delta\) | C-S | (M-N) \(_{\text{HL}}\&\ Nam\) | (M-O) \(_{\text{HL}}\&\ lactam\) |
|-----------|-----|------|----------------|----------------|-----|------|----|----------------|----------------|
| HL        | 3394 | 3031 | 1696         | 1739         | ----- | 574  |     |                  |                |
|           |      |      | 1666         | 1666         |      |      |     |                  |                |
|           |      |      | (2970,2927)  |              |      |      |     |                  |                |
| C1        | 3363 | 3062 | 1674         | 1616         | 218  | 555  | 486 | 455             |                |
|           |      | 2970 | 1655         | 1392         |      |      | 513 | 420             |                |
| C2        | 3302 | 3070 | 1670         | 1616         | 224  | 578  | 505 | 478             |                |
|           |      | 2962 | 1651         | 1392         |      |      | 532 | 447             |                |
| C3        | 3398 | 3194 | 1670         | 1600         | 208  | 586  | 559 | 424             |                |
|           |      | 2970 | 1620         | 1392         |      |      |     |                  |                |
| C4        | 3332 | 3110 | 1674         | 1612         | 224  | 590  | 501 | 474             |                |
|           |      |      | 1674         | 1612         |      |      |     |                  |                |
| C5        | 3305 | 3062 | 1674         | 1600         | 219  | 551  | 478 | 478             |                |
|           |      | 2974 | 1651         | 1381         |      |      | 528 | 443             |                |
| C6        | 3394 | 3074 | 1666         | 1597         | 201  | 574  | 505 | 474             |                |
|           |      | 2985 | 1624         | 1396         |      |      | 574 | 520             |                |
| C7        | 3387 | 3070 | 1670         | 1597         | 220  | 551  | 505 | 451             |                |

3.6. The Magnetic Measurements and Electronic Spectra

The electronic spectrum for ligand HL shows peaks at 214 nm and 255nm appointed to \(\pi \rightarrow \pi^*\) and n–
\(\pi^*\)transition respectively, Table (4) [28]. The magnetic moment shows the C1 complex is 3.81BM and the electronic spectrum show three bands at 783,632 and 275 nm, which due to the \(4\text{Ag}_{g} \rightarrow 3\text{T}_2\text{g},\) \(4\text{Ag}_{g} \rightarrow 3\text{T}_1\text{g}(F)\) and charge transfer (CT) transitions respectively. The C2 complex has 5.81BM, three bands at (823, 638 and 274) nm which due to the d-d transition; \(6\text{Ag}_{g} \rightarrow 4\text{T}_1\text{g}(4G),\) \(6\text{Ag}_{g} \rightarrow 4\text{E}_g\) and C.T Transitions [29, 30]. The C3 complex has 5.38 BM and display three peaks; the first peak at (282, 673 and 866) nm are ascribed to the charge transfer, \(4\text{T}_1\text{g} \rightarrow 3\text{T}_1\text{g}(p)\) and \(4\text{T}_1\text{g} \rightarrow 4\text{A}_g\) (F). The C4 complex has 2.37 B.M. and show one band appears at (279 nm) which due to the charge transfer transitions and showed absorption bands at (780 and 837) nm these transitions may be appointed to \(3\text{Ag}_{g}(F) \rightarrow 3\text{T}_1\text{g}(F)\) \(\psi_3\) and \(3\text{Ag}_{g}(F) \rightarrow 3\text{T}_1\text{g}(F)\) \(\psi_1\). The magnetic moment of the C5 is 1.85 B.M. and shows three bands at 829, 297 and 278 nm assignable to \(3\text{E}_g \rightarrow 3\text{T}_2\text{g}\) ligand field and charge transfer respectively [31]. C6 and C7 complexes are diamagnetic and their electronic spectra showed the absorption bands at (275 and
280) nm respectively ascribed to the charge transfer transitions. The lack of available d orbital precludes L to M charge transfer bands for these compounds which are compatible with these complexes having octahedral structures [32].

| Compounds | λ<sub>max</sub> | Е<sub>max</sub> | ν' | Number of µ<sub>eff</sub> |
|-----------|--------------|-------------|----|-----------------|
| HL        | 214          | 46728       | 1494| ----            |
|           | 255          | 39215       | 1953|                |
|           | 275          | 36363       | 1902| 3.81           |
| C1        | 632          | 15822       | 19 |                |
|           | 783          | 12771       | 7  |                |
|           | 274          | 36496       | 1949|                |
| C2        | 638          | 15673       | 9  | 5.81           |
|           | 823          | 12150       | 4  |                |
|           | 282          | 45460       | 2490|                |
| C3        | 673          | 14858       | 266 | 5.38           |
|           | 866          | 11547       | 210 |                |
|           | 279          | 35842       | 2207|                |
| C4        | 780          | 12820       | 5  | 2.37           |
|           | 837          | 11947       | 4  |                |
| C5        | 278          | 35971       | 1924|                |
|           | 297          | 33670       | 1924| 1.85           |
|           | 829          | 12062       | 39 |                |
| C6        | 275          | 36363       | 2055| Dia.           |
| C7        | 280          | 35714       | 2361| Dia.           |

### 4. Bacterial activities

The Schiff base ligand HL and its complexes under study are tested against different type of bacteria were given in, fig.5. The ligand HL and its complexes display different activity. Cr, Ni, Cu, Cd and Hg complexes show inactive against Staphylococcus aureus. The Cr, Cu and Cd complexes show inactive against Pseudomonas. The (Nam) did not show any activity toward all types of microorganism except Pseudomonas. All complexes shows good activity towards E-coli and Bacillus except Fe, Cd and Hg complexes shows very good antibacterial activity against four organisms. The increased of the metal (III and II) complexes can be elucidate on the Tweedy's theory [33-35].

![Figure 5. The biological activity of prepared compounds](image-url)
5. Conclusion
The new ligand HL derived from amoxicillin drug with 4-Chlorobenzophenone is prepared and diagnosed. The results show the linked of HL format with metal ions through the nitrogen of Schiff base group and oxygen β- lactam and carboxylate ion groups while Na coordinates through nitrogen atom of pyridin ring. Electronic spectra and magnetic susceptibility of prepared complexes show that the shape of complexes is octahedral geometry fig.6. The biological test of complexes has been shown various activities against different type of bacteria.

![Figure 6. Structure of complexes](image)

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