

Synthesis, Spectroscopic Characterization and Antibacterial Properties of some Metal (II) Complexes of 2-(6-methoxybenzothiazol-2-ylimino)methyl)-4-nitrophenol.

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Research Article

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Keywords: antibacterial properties, geometry, benzothiazole, Schiff base.**ABSTRACT**

Mn(II), Co(II), Ni(II), Cu(II), Zn(II), and Pd(II) complexes of the Schiff base 2-(6-methoxybenzothiazol-2-ylimino)methyl)-4-nitrophenol (HL) derived from condensation of 2-hydroxy-5-nitrobenzaldehyde and 2-amino-6-methoxybenzothiazole have been synthesized and characterized by percentage metal, solubility, infrared and electronic spectra measurements. Their *in-vitro* anti-bacterial activities against *Bacillus subtilis*, *Staphylococcus aureus*, *Proteus mirabilis*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*, and *Escherichia coli* were also evaluated. The metal complexes formed as $[ML(NO_3)] \cdot bH_2O$, where M = Mn, Co, Cu, Zn; b = 0.5 - 4, with the exceptions of the Pd(II) and Ni(II) complexes that analyzed as $[Pd(L)_2] \cdot 4H_2O$ and $[Ni(L)Cl(H_2O)]$. The infrared spectra confirmed that coordination by the Schiff base is via the imine nitrogen and phenol oxygen atoms respectively, resulting in a NO chromophores around the metal ion. The electronic spectra measurement was corroborative of a 4-coordinate tetrahedral/square planar geometry for the complexes. The complexes mostly exhibit good *in-vitro* antibacterial activities against *S. aureus*, *E. coli* and *P. mirabilis* with inhibitory zones range of 16.0–27.0 mm, 13.0–15.0 mm and 17.0–20.0 mm respectively. Its noteworthy that the Pd(II) and Zn(II) complexes had broad spectrum anti-bacteria activity against the bacteria used with the exception of *B. subtilis* with inhibitory zones range of 13.0–22.0 and 13.0–27.0 mm respectively.

INTRODUCTION

Benzothiazole and its derivatives are one of the most important heterocyclic compounds that have received overwhelming response owing to its biological applications e.g. they have been reported as inhibitors of mycobacterium tuberculosis ATP phosphoribosyl tranferase [1], antidiabetic agents [2], antimicrobial agents [3,4,5], anti-inflammatory agents [6], analgesic agents [7], inhibitors of human cox-2 and exhibits hypoglycemic activity [8]. The combination of different heterocyclic compounds is a well-known approach for drug-like molecules build-up, which allows achieving compounds with new pharmacological profile, action strengthening or toxicity lowering abilities [9]. Schiff base formation affords the opportunity of such combinations, and has largely been reported as possessing wide range of biological applications [10]. Specifically, Schiff bases of benzothiazole derivatives have shown positive activity towards Parkinson's disease [11], and many are useful as antitumour/anticancer, anticandidous, antihistaminic, anti-inflammatory anti bacterial agents [12,13,14,15,16,17,18,19]. A thorough literature search shows that the Schiff base prepared from the condensation reaction of 2-amino-5,6-dimethylbenzothiazole and 2-hydroxy-5-nitrobenzaldehyde (2-(6-methoxybenzothiazol-2-ylimino)methyl)-4-nitrophenol) has not been reported.

Therefore, we are reporting the synthesis and characterization of metal (II) complexes of the Schiff base, of 2-(6-methoxybenzothiazol-2-ylimino)methyl)-4-nitrophenol for the first time as a continuation of the research activities of our group [20–25]. The aims of this study is to investigation the physicochemical properties of these metal complexes as well as their potency as *in-vitro* antibacterial agents

EXPERIMENTAL

Chemicals

Reagent grade 2-hydroxy-5-nitrobenzaldehyde, 2-amino-6-methoxybenzothiazole, manganese(II) nitrate hexahydrate, copper(II) nitrate hexahydrate, zinc(II) nitrate hexahydrate, palladium(II) chloride, nickel(II) chloride dihydrate and cobalt(II) nitrate hexahydrate were purchased from Aldrich and BDH chemicals, and were used as received. Solvents were purified by distillation.

Physical measurements

Percentage manganese, cobalt, nickel, copper, zinc and palladium in the complexes were determined titrimetrically [26]. Infrared spectra were measured in deuterated dimethyl sulfoxide solvent on a Shimadzu FTIR-8400 spectrophotometer while electronic spectra in chloroform were recorded on Unicam Helios γ spectrophotometer, and melting points (uncorrected) were done using a Stuart scientific melting point apparatus smp3.

Synthesis

Synthesis of 2-(6-methoxybenzothiazol-2-ylimino)methyl)-4-nitrophenol

The Schiff base was prepared by adding 5g (0.03 mole) of 2-amino-6-methoxybenzothiazole neat in bits to a stirring 5g (0.03 mole) of 2-hydroxy-5-nitrobenzaldehyde in 50 mL of hot methanol. The resulting orange-coloured homogeneous solution was then refluxed for 3 h after the addition of 4 drops of acetic acid. The products formed were filtered, washed with methanol and dried over silica gel.

Color (Yellow); IR (DMSO, cm^{-1}): ν_{OH} (3457s), $\nu_{\text{C=N}}$ (1608s 1558s) $\nu_{\text{C=C}}$ (1491s); UV (DMSO) λ_{max} (kK, ϵ): 40.32 (1×10^6), 34.97 (1×10^5), 28.25 (1×10^6); Formula mass: 329.33; melting point: 264°C.

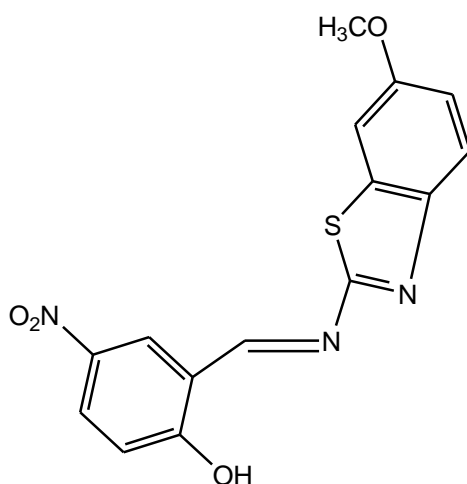


Figure 1: The structural formula of the Schiff base

Synthesis of metal complexes

To a stirring solution of 0.28 g (9.0×10^{-4} moles) of HL in 20 mL of methanol, and 0.16 – 0.30g (9.0×10^{-4} moles) of the metal (II) nitrates and chlorides (M= Cu, Mn, Co, Ni, Zn, Pd) in 20 mL of methanol were added dropwisely. The resulting homogeneous solutions pH was adjusted to 9, with triethylamine, and then refluxed for 3 h. The precipitates formed were filtered, washed with methanol and dried under vacuum.

[Mn(L)NO₃]0.5H₂O: Color (Yellow); IR (DMSO, cm^{-1}): ν_{OH} (3434s), $\nu_{\text{C=N}}$ (1600s) $\nu_{\text{C=C}}$ (1464 s); $\nu_{\text{M-N}}$ (637 m), $\nu_{\text{M-O}}$ (549 m); Vis/UV λ_{max} (kK, ϵ): 15.36 (100); 23.47 (100), 35.33 (1×10^4), 40.14(1×10^5); Formula mass 453.77; melting point: 318°C.

[Pd(L)₂]4H₂O: Color (Reddish brown); IR (DMSO, cm^{-1}): ν_{OH} (3445 s), $\nu_{\text{C=N}}$ (1601s) $\nu_{\text{C=C}}$ (1469 s); $\nu_{\text{M-N}}$ (672 m), $\nu_{\text{M-O}}$ (543 m); Vis/UV λ_{max} (kK, ϵ): 14.79(100), 22.37(200), 36.33 (1×10^4), 42.84(1×10^5); Formula mass 835.12; melting point: 318°C.

[Cu(L)NO₃]4H₂O: Color (Brown); IR (DMSO, cm^{-1}): ν_{OH} (3451s), $\nu_{\text{C=N}}$ (1600 s) $\nu_{\text{C=C}}$ (1461 s); $\nu_{\text{M-N}}$ (652 m), $\nu_{\text{M-O}}$ (501 m) UV λ_{max} (kK, ϵ): 23.81 (200), 30.30 (1×10^5), 38.46 (1×10^6); Formula mass 525.93; melting point: 350°C.

[Ni(L)Cl(H₂O)]: Color (Yellow); IR (DMSO, cm⁻¹): νOH (3440s), νC=N (1606 s 1548 s) νC=C (1469 s); νM-N(510 m), νM-O/M-Cl(370 m) Vis/UV λ_{max} (kK, ε): 23.73 (400); 35.30 (1 × 10⁵), 40.46 (1 × 10⁶) Formula mass 542.04; melting point 318°C.

[Co(L)NO₃]3H₂O: Color (Orange); IR (DMSO, cm⁻¹): νOH (3440 s), νC=N (1603 s) νC=C(1442 s); νM-N(657 m), νM-O(492 m) UV λ_{max} (kK, ε): 15.36(100), 23.47 (300); 27.47 (1 × 10⁵), 40.00 (1 × 10⁵), 40.00 (1 × 10⁵); Formula mass 503.34; melting point 320°C.

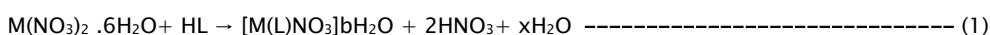
[Zn(L)NO₃]1.5H₂O : Color (Yellow); IR (DMSO, cm⁻¹): νOH (3399s), νC=N (1616 s) νC=C (1447 s); νM-N(501 m), νM-O(364 m); UV λ_{max} (kK, ε): 22.0 (100); 27.0(1000), 39.30 (1 × 10⁵), 42.0 (1 × 10⁶) Formula mass 482.33; melting point 342°C.

Antibacterial assay

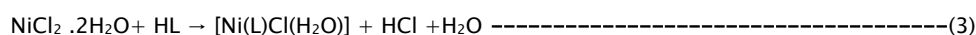
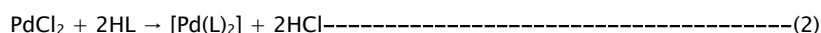
The assay was carried out on the metal-free Schiff base and their metal(II) complexes using Agar diffusion technique. The surface of the agar in a Petri dish was uniformly inoculated with 0.3 mL of 18 hours old bacteria culture of *Bacillus subtilis*, *Staphylococcus aureus*, *Proteus mirabilis*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*, and *Escherichia coli*. Using a sterile cork borer, 6 mm wells were bored into agar. Then 0.06 mL of 10 mg/mL concentration of each metal complex in DMSO was introduced into the wells and the plates were allowed to stand on the bench for 30 min before incubation at 37°C for 24 h. Inhibitory zones (in mm) were taken as a measure of antimicrobial activity. The experiments were conducted in duplicates and streptomycin was used as the reference drug [27].

RESULTS AND DISCUSSIONS

The equations for the formation of the complexes are given below;



(where M = Zn(II), Cu(II), Mn(II), Co(II), b = 0.5 - 4, x = 2 - 5.5)



The metal complexes formed as [ML(NO₃)₃].bH₂O, where M = Mn, Co, Cu, Zn; b = 0.5 - 4, with the exceptions of the Pd(II) and Ni(II) complexes that analyzed as [Pd(L)₂].4H₂O and [Ni(L)Cl(H₂O)]. Proposed structures for the ligand is shown in Figure 1. The formation of this ligand is confirmed by melting point, IR and electronic spectra measurements.

Electronic spectra

The Mn(II) complex showed two bands at 15.36 and 23.47 kK respectively, typical of a 4-coordinate tetrahedral geometry and were assigned as ⁶A₁ → ⁴E₁ and ⁶A₁ → ⁴T₂ transitions [20]. Similarly, the Co(II) complexes displayed two bands at 18.97 and 22.37 kK assigned to ⁴A₂ → ⁴T₁(F), (ν₂), and ⁴A₂ → ⁴T₁(P), (ν₃) transitions of a 4-coordinate tetrahedral geometry [9]. In addition, the Pd(II) complex exhibited two bands at 14.70 and 22.37 kK indicative of 4-coordinate square-planar geometry and were assigned to ¹A_{1g} → ¹B_{1g} and ¹A_{1g} → ¹E_{2g} transitions [23]. Literature confirmed that square-planar Ni(II) complexes absorb in the range 22.0–25.0 kK, while tetrahedral complexes Cu(II) complexes have single absorption band below 10.0 kK and square-planar Cu(II) complexes absorb between 10.0–20.0 kK [10]. Thus, the Ni(II) and Cu(II) complexes in this study displayed lone bands each at 23.73 and 23.81 kK typical of square planar geometry and were assigned to ¹A_{1g} → ¹A_{2g} and ²B_{1g} → ²E_{1g} transitions respectively [21]. The Zn(II) complex was expectedly diamagnetic with M → L CT band at 22.20 kK and since its CFSE is zero, the assumed geometry was tetrahedral [28, 29].

Normally, the molar extinction coefficient (ε) allows a distinction of tetrahedral geometry from octahedral geometry. Tetrahedral complexes are expected to have a higher molar extinction coefficient in the range 10²–10³ cm² mol⁻¹ and octahedral complexes should have ε between 1–50 cm² mol⁻¹. None of the complexes in this study had ε below 10² cm² mol⁻¹, this is indicative of a tetrahedral/square-planar geometry [20].

Infrared spectra

The strong bands at 1608 - 1558 cm⁻¹ in the metal free Schiff base was assigned to the νC=N of azomethine and thiazole stretching vibrations. These bands were mostly shifted to lower wave numbers in the range 1606 - 1548 cm⁻¹, with the exception of [Zn(L)NO₃] that experienced an increased shift to 1616 cm⁻¹. This indicated the coordination of the imine nitrogen to metal ions. Similarly, the band for νC=C at 1491 cm⁻¹ in the ligand was shifted to 1469–1462 cm⁻¹ in the metal complexes due to coordination [9–10]. The phenol band at 3457 cm⁻¹ in the spectrum of the Schiff base was absent in the spectra of the metal complexes due to the coordination of the enol oxygen atom [19]. In the spectra of all the metal complexes, a broad band at 3500 cm⁻¹ was observed

due to presence of coordinated /hydration water. The bands due to M–N and M–O/M–Cl were observed in the range 672–501 cm^{-1} and 549–364 cm^{-1} respectively. These bands were absent in the spectra of the Schiff base [29].

Antibacterial activities

The antibacterial activities are presented in Figure 2. The Schiff base was active against all the bacteria used *S. aureus*, *B. subtilis*, *K. oxytoca*, *E. coli* and *P. mirabilis* with the exception of *P. aeruginosa* with inhibitory zones range of 13.0–26.0mm. All the metal complexes were active against *P. mirabilis* with inhibitory zones range of 17.0–21.0mm. Similarly, all the complexes were active against *S. aureus* and *E. coli* with inhibitory zones range of 16.0–27.0 mm and 13.0–15.0 mm with the exceptions of Mn(II) and Cu(II) complexes. On the contrary, *P. aeruginosa* was sensitive to only Zn(II) and Pd(II) complexes with inhibitory zones of 13.0 mm and 15.0 mm respectively. In addition, *K. oxytoca* was sensitive to Zn(II), Mn(II) and Pd(II) complexes with inhibitory zones range of 13.0–15.0 mm while *B. subtilis* was sensitive only to the Ni(II) complex with inhibitory zone of 14.0 mm. Generally, the metal complexes were less active than the Schiff base. This is contrary to chelation theory (which states that chelation increases antimicrobial activity, because of partial sharing of its positive charge with donor groups of the ligand and possible π -electron delocalization which increases the lipophilic character) [9]. The lower activities of the metal complexes may be attributed to lower lipophilicity of the complexes, which decrease the penetration of the complexes through the lipid membrane [10]. Streptomycin activities (30.0–44.0 mm) against the various isolates relative to the metal complexes (13.0–27.0 mm) show that the activities of the latter are much lower, with the optimum activity being about 75 percent that of streptomycin in Zn(II) complex against *Staphylococcus aureus*. Its noteworthy that the Pd(II) and Zn(II) complexes had broad spectrum anti-bacteria activity against the bacteria used with the exception of *B. subtilis* with inhibitory zones range of 13.0–22.0 mm and 13.0–27.0 mm respectively, proving their potentials as broad-spectrum antibacterial agents [25,26,27,28,29].

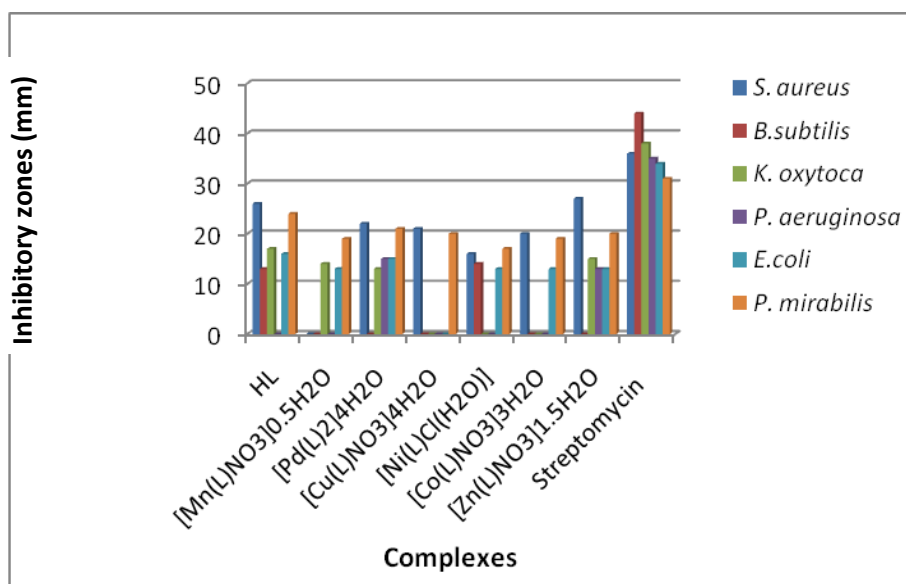


Figure 2: Histogram of the antibacterial activities of ligand and its metal complexes

CONCLUSION

Mn(II), Co(II), Ni(II), Cu(II), Zn(II), and Pd(II) complexes of the Schiff base 2-(6-methoxybenzothiazol-2-ylimino)methyl-4-nitrophenol coordinated to the metal ions via the imine nitrogen and phenol oxygen atoms respectively. The electronic spectra measurement was corroborative of a 4-coordinate tetrahedral/square planar geometry for the complexes. The complexes exhibited good *in-vitro* antibacterial activities against *S. aureus*, *E. coli* and *P. mirabilis*, while Pd(II) and Zn(II) complexes had broad spectrum anti-bacteria activity against all the bacteria used with the exception of *B. subtilis* proving their potentials as broad-spectrum antibacterial agents.

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