

Synthesis and characterization of novel Schiff base complexes of Cu (II), Cd (II), Ni (II), and Co (II) ions with N'-(4-nitrobenzylidene) isonicotinohydrazide

ABSTRACT

The Schiff base complexes of Cu (II), Cd (II), Ni (II), and Co (II) ions were synthesized utilizing Schiff base ligand. The Schiff base ligand was produced through the reaction of isoniazid with equimolar quantities of *p*-nitrobenzaldehyde yielding final product of N'-(4-nitrobenzylidene)isonicotinohydrazide (L). The ligand and its metal complexes were assessed by TLC, melting point determination, conductivity measurement, FTIR spectroscopy, magnetic moment analysis, and UV-Vis spectroscopy. The FTIR spectra revealed that the Schiff base ligand established coordination bonds with the metal ion by utilizing the nitrogen (N) atom of the azomethine group and the oxygen (O) atom of the carbonyl group. All synthesized compounds exhibited cytotoxicity against brine shrimp nauplii, thereby confirming their biological activity. [Ni(L)₂] and [Cd(L)₂] complexes displayed greater cytotoxic or anticancer efficacy. In addition, the antibacterial efficacy of both ligand and its complexes was investigated against several harmful bacteria. In comparison with the ligand (L), the antibacterial activity of the complexes of [Cu(L)₂], [Cd(L)₂], and [Ni(L)₂] is higher, whereas that of [Co(L)₂] is lower.

Keywords: Novel Schiff base, Metal complexes, Characterization, Antibacterial activity, Cytotoxic efficacy

1. INTRODUCTION

A hydrazine or hydrazide reacts with a ketone or aldehyde producing a hydrazone containing a C=N-NH group [1]. The wide range of pharmacological activity profiles that hydrazones possess, along with their structural flexibility and ligating behavior, have attracted a lot of attention and led to their extensive application in the field of chemistry [2]. The ability of Schiff bases to readily create stable complexes with most of the transition metal ions in the periodic table, that is why they play such an essential role in inorganic chemistry [3]. They also exhibited luminous characteristics [4]. Schiff base metal complexes are important in industrial, agricultural, analytical and synthetic chemistry [5,6]. They are precursors of possible anti-diabetic medications and have been utilized extensively as intermediates for the synthesis of numerous medically significant heterocyclic chemical species [7]. These compounds have found widerange of application in building blocks for several biologically significant heterocyclic compounds, including those with anti-diabetic, anti-mycotic [8], anti-malarial, anti-proliferative [9], anticonvulsant [10], genotoxic [11], and anti-depressant properties [12]. A variety of synthetic chemical processes, physiologically active compounds, pigments, plant growth regulators, and more could be catalyzed by Schiff bases [13]. The emergence of bioinorganic chemistry has sparked a growing fascination with Schiff base complexes, as it has been acknowledged that many of these complexes can function as prototypes for biologically significant entities [14]. There has been a lot of interest in metal complexes of S-, N-, and O-chelating ligands due to their intriguing physio-chemical features, strong biological activity, and their functions as models

for active sites of metalloenzymes [15]. Nitrogencontaining heterocyclic compounds have the capability to serve as probes for DNA structure, as well as chemotherapeuticagents and diagnostic medicines [16].

Tuberculosis (TB), a long-standing affliction of humans, has begun a global pandemic environment mostly caused by Mycobacterium tuberculosis [17]. Isoniazid is one of the most effective hydrazides that has been prepared available for the treatment of TB [18]. The isoniazid and ketoprofen Schiff base can be utilized for the treatment of tuberculosis, as well as for alleviating fever and exhaustion associated with the disease. Schiff bases possess significant medical and pharmaceutical significance owing to their diverse array of biological activities, including anti-inflammatory, antibacterial, antioxidant, and anti-tubercular properties. Schiff basewith azomethine activity in heterocyclic systems are recognized for their cytotoxic, antibacterial, anticancer, and antifungal properties [19]. As long list of carcinogenic and ecological pollutant impacts, some research has shown that organic Cd complexes can specifically induce cell death in cancer cells. These complexes are also interesting due to the many bioactivities they exhibit, including DNA binding, anticancer, and antibacterial effects [20]. Thus, the aim of the present study is to synthesize, characterize, and biological screening of novel Schiff base complexes of Co (II), Cu (II), Cd (II) and Ni (II) ions with (4-nitrobenzaldehyde)isonicotinohydrazide.

2. METHODOLOGY

2.1 Instrumentation

The ligand and complexes were subjected to microanalysis using a Leco CHNS elemental analyzer (C, H, N) [21]. Using electrochemical melting equipment, the melting points of several complexes and ligands were ascertained. A Perkin-Elmer Lambda spectrophotometer is used to record the UV-Vis spectra [22]. A Bruker Tensor 27 FTIR Spectrometer (KBr pellets, Nujol mulls, 4000-400 cm^{-1}) was used to record the infrared (IR) spectra [23].

2.2 Materials and Reagents

Isonicotinic hydrazide acid, p-nitro benzaldehyde, and $\text{Co}(\text{CH}_3\text{COO})_2 \cdot 4\text{H}_2\text{O}$, $\text{Cu}(\text{CH}_3\text{COO})_2 \cdot \text{H}_2\text{O}$, $\text{Ni}(\text{CH}_3\text{COO})_2 \cdot 4\text{H}_2\text{O}$ and $\text{Cd}(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$, methanol, absolute ethanol, chloroform and DMSO were used in this work supplied by Sigma-Aldrich.

2.3 Synthesis

2.3.1 Synthesis of SB ligand [N'-(4-nitrobenzylidene)isonicotinohydrazide]

The ligand (L) was prepared through a condensation reaction involving 10 mmol of 4-nitrobenzaldehyde (1.51g) and 10 mmol of isoniazid (1.37g) in a round-bottomed flask. The resulting solutions were combined and refluxed for a duration of 4 hours. After cooling, a yellow-colored product was obtained. We subsequently washed this product with acetone and diethyl ether, and then dried it in a vacuum desiccator using anhydrous CaCl_2 . We evaluated the progress and purity of the ligand using TLC with various solvents. The product showed solubility in methanol, ethanol, chloroform, DMSO, and other solvents, yielding 91% and displaying a yellow color.

Molecular formula: $\text{C}_{13}\text{H}_{10}\text{N}_4\text{O}_3$; appearance: yellow crystalline solid; yield: 89%; melting point: 287-289°C; FTIR (KBr pellet): $\nu(\text{-OH})$ -3409 cm^{-1} , $\nu(>\text{C}=\text{O})$ -1685 cm^{-1} , $\nu(>\text{C}=\text{N})$ -1562 cm^{-1} ; UV-Vis. (DMSO): λ_{max} at: 268nm and 336nm; Ana Cal: C-57.78, H-3.73, N-20.73, Found: C-57.71, H-3.75, N-20.80.

2.3.2 Synthesis of Ni (II), Cd (II), Cu (II), and Co (II) metal complexes

The complexes that were synthesized using the general formula $[M(L)_2]$, where M stood for the metal ions Cu (II), Cd (II), Ni (II), and Co (II) and L for the Schiff base ligand. In the complex formation process, a 15-mL ethanolic solution of metal acetate salts [Cu (II), Co (II), Cd (II), Ni (II)] was placed in a two-necked round-bottom flask and stirred magnetically. An ethanolic solution (20 mL) of the prepared Schiff base ligand was added dropwise with continuous stirring on a magnetic stirrer for 2–3 hours. After cooling, A colored solid product was obtained, washed it with hot ethanol and diethyl ether, and dried in a vacuum desiccator using anhydrous $CaCl_2$. The reaction's progress was monitored by TLC, using n-hexane and chloroform as solvents. The complexes exhibited solubility in DMSO. Fig. 1 illustrates the chemical structure of the target ligand (L) and the common structure of metal complexes.

[Co(L)₂]: appearance: dark brown; yield: 80%; melting point: >300°C; molecular formula: $CoC_{26}H_{18}N_8O_6$; conductivity: $8 \text{ Sm}^2\text{mol}^{-1}$; μ_{eff} B.M.: 3.94; FTIR (KBr pellet): $\nu(-OH)$ -3429 cm^{-1} , $\nu(C-O)$ -1339 cm^{-1} , $\nu(>C=N)$ -1594 cm^{-1} , $\nu(Co-O)$ -604 cm^{-1} , $\nu(Co-N)$ -470 cm^{-1} ; UV-Vis. (DMSO): λ_{max} at: 274nm, 344nm and 408nm; Ana Cal: C-52.27, H-3.04, N-18.76, Co-9.86; Found: C-52.30, H-3.00, N-18.81, Co-9.82.

[Ni(L)₂]: appearance: yellow; yield: 58%; melting point: >300°C; molecular formula: $NiC_{26}H_{18}N_8O_6$; conductivity: $7 \text{ Sm}^2\text{mol}^{-1}$; μ_{eff} B.M.: 2.90; FTIR (KBr pellet): $\nu(-OH)$ -3396 cm^{-1} , $\nu(C-O)$ -1345 cm^{-1} , $\nu(>C=N)$ -1592 cm^{-1} , $\nu(Ni-O)$ -599 cm^{-1} , $\nu(Ni-N)$ -477 cm^{-1} ; UV-Vis. (DMSO): λ_{max} at: 273nm, 344nm and 412nm; Ana Cal: C-52.29, H-3.05, N-18.76, Ni-9.83; Found: C-52.32, H-3.01, N-18.80, Ni-9.79.

[Cu(L)₂]: appearance: green; yield: 65%; melting point: >300°C; molecular formula: $CuC_{26}H_{18}N_8O_6$; conductivity: $5 \text{ Sm}^2\text{mol}^{-1}$; μ_{eff} B.M.: 1.84; FTIR (KBr pellet): $\nu(-OH)$ -3428 cm^{-1} , $\nu(C-O)$ -1340 cm^{-1} , $\nu(>C=N)$ -1591 cm^{-1} , $\nu(Cu-O)$ -597 cm^{-1} , $\nu(Cu-N)$ -471 cm^{-1} ; UV-Vis. (DMSO): λ_{max} at: 261nm, 342nm and 411nm; Ana Cal: C-51.87, H-3.01, N-18.61, Cu-10.56; Found: C-52.92, H-3.04, N-18.56, Cu-10.52.

[Cd(L)₂]: appearance: dark yellow; yield: 78%; melting point: >300°C; molecular formula: $CdC_{26}H_{18}N_8O_6$; conductivity: $6 \text{ Sm}^2\text{mol}^{-1}$; μ_{eff} B.M.: diamagnetic; FTIR (KBr pellet): $\nu(-OH)$ -3424 cm^{-1} , $\nu(C-O)$ -1338 cm^{-1} , $\nu(>C=N)$ -1587 cm^{-1} , $\nu(Cd-O)$ -602 cm^{-1} , $\nu(Cd-N)$ -473 cm^{-1} ; UV-Vis. (DMSO): λ_{max} at: 263nm, 294nm and 421nm; Ana Cal: C-47.98, H-2.79, N-17.22, Cd-17.27; Found: C-48.02, H-2.74, N-17.25, Cd-17.24.

Fig. 1: - Synthesis pathway of Schiff base ligand $C_{13}H_{10}O_3N_4$ (L) and it's metal complexes

[Here, M=Cu (II), Cd (II), Ni (II) and Co (II)]

3. RESULTS AND DISCUSSION

3.1 Investigation of Elemental and Molar Conductivity

An elemental analysis was conducted using a Leco CHNS elemental analyzer to determine the percentage of carbon (C), hydrogen (H), and nitrogen (N) atoms in the Schiff base ligand $C_{13}H_{10}N_4O_3$ (L) and its metal complexes with Ni (II), Cd (II), Co (II), and Cu (II) are represented in Table 1 [24]. Complexometric titration with EDTA was used to calculate the metal percentage in accordance with the method described in the literature [25]. The molar conductance of metal complexes was determined at a concentration of $10^{-3} \text{ mol dm}^{-3}$. The molar conductivity values of the complexes fall within the range of 5 to 8 $\text{ohm}^{-1} \text{cm}^2 \text{mol}^{-1}$ (Table 1). The lower values indicate that the metal complexes are not electrolytic in nature [26].

Table 1: Chemical composition and physical properties of L and its metal complexes

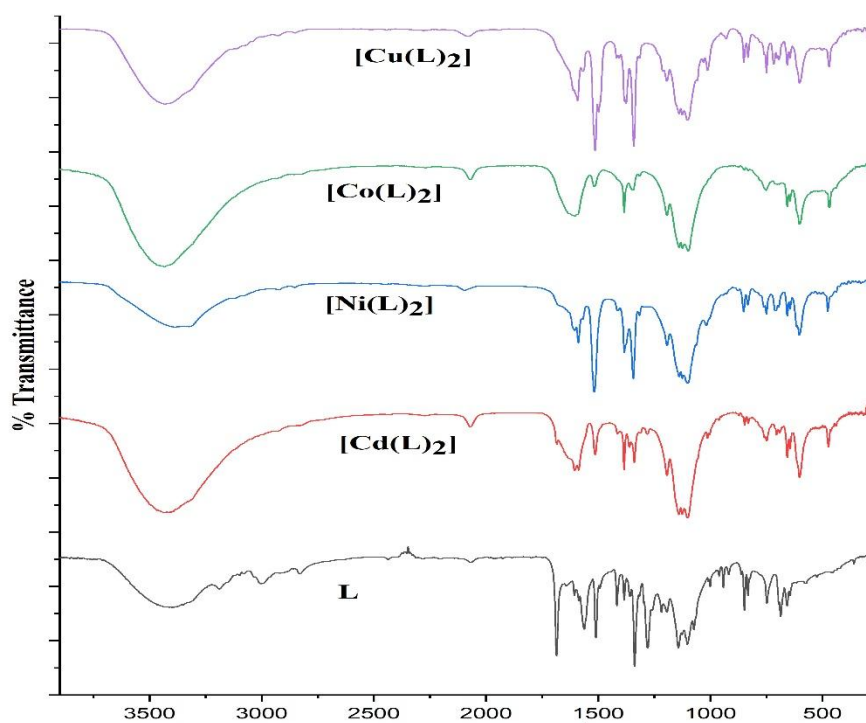
Compound (M.W.)	Empirical formula	Melting Point($^{\circ}\text{C}$)	Color	Λ^a	(%) Found (Cal.)			
					C	H	N	M
Ligand (L) (239.3)	$C_{13}H_{10}N_4O_3$	287-289	yellow	----	57.71 (57.78)	3.75 (3.73)	20.80 (20.73)	----
$[\text{Ni}(\text{L})_2]$ (535.3)	$\text{NiC}_{26}\text{H}_{18}\text{N}_8\text{O}_6$	>300	yellow	7	52.32 (52.29)	4.44 (4.48)	15.74 (15.69)	11.01 (10.97)
$[\text{Cd}(\text{L})_2]$ (650.9)	$\text{CdC}_{26}\text{H}_{18}\text{N}_8\text{O}_6$	>300	dark yellow	6	48.02 (47.98)	2.74 (2.79)	17.22 (17.25)	17.24 (17.27)
$[\text{Co}(\text{L})_2]$ (535.3)	$\text{CoC}_{26}\text{H}_{18}\text{N}_8\text{O}_6$	>300	dark brown	8	52.30 (52.27)	3.00 (3.04)	18.81 (18.76)	9.82 (9.86)
$[\text{Cu}(\text{L})_2]$ (540.1)	$\text{CuC}_{26}\text{H}_{18}\text{N}_8\text{O}_6$	>300	green	5	52.92 (51.87)	3.04 (3.01)	18.56 (18.61)	10.52 (10.56)

3.2 FTIR Spectra

The infrared spectra of L exhibit three distinct bands at 3409 cm^{-1} , 1685 cm^{-1} and 1562 cm^{-1} . These bands can be identified as the stretching vibrations $\nu(\text{-OH})$, $\nu(\text{C=O})$ and azomethine groups $\nu(>\text{C=N})$ respectively [27]. In the complexes, the bands associated with the $\nu(\text{C=O})$ vibration vanished [28], while a new C–O band emerged. Additionally, this band saw a downward change in frequency, suggesting the participation of the carbonyl group in the metal bonding. The absorption band associated with the imine group, $\nu(\text{C=N})$, exhibited a shift to 1592 cm^{-1} , 1594 cm^{-1} , 1587 cm^{-1} and 1591 cm^{-1} for the $[\text{Ni}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complexes, respectively. This is consistent with the nitrogen atom of the $\nu(\text{C=N})$ participating in bonding with the metal (II) ions [29]. The presence of nitrogen and oxygen in coordination was confirmed by the emergence of additional bands at specific wavenumbers for each metal complex. Specifically, the bands at 477 cm^{-1} and 599 cm^{-1} were observed for the $[\text{Ni}(\text{L})_2]$, 470 cm^{-1} and 604 cm^{-1} for the $[\text{Co}(\text{L})_2]$, 473 cm^{-1} and 602 cm^{-1} for the $[\text{Cd}(\text{L})_2]$, and 471 cm^{-1} and 597 cm^{-1} for the $[\text{Cu}(\text{L})_2]$. These bands were attributed to the $\nu(\text{M-N})$ and $\nu(\text{M-O})$ modes [30]. $[\text{Ni}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ FTIR spectra all showed a broad peak at $3350\text{--}3450 \text{ cm}^{-1}$. There was probably water in the KBr particle, which caused this [31]. Fig. 2 displays the spectra and Table 2 shows the data for the ligand (L), $[\text{Ni}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complexes.

Table 2: The FTIR spectral data (in cm^{-1}) for ligand(L) and its metal complexes

Compounds	$\nu(-\text{OH})$	$\nu(>\text{C}=\text{N})$	$\nu(\text{C}=\text{O})$	$\nu(\text{C}-\text{O})$	$\nu(\text{M}-\text{O})$	$\nu(\text{M}-\text{N})$
L	3409	1562	1685	----	----	----
$[\text{Ni}(\text{L})_2]$	3396	1592	----	1345	599	477
$[\text{Co}(\text{L})_2]$	3429	1594	----	1339	604	470
$[\text{Cd}(\text{L})_2]$	3424	1587	----	1338	602	473
$[\text{Cu}(\text{L})_2]$	3428	1591	----	1340	597	471

**Fig. 2: FTIR spectra of L & its metal complexes**

3.3 UV-Vis spectra

The ligand (L) exhibited two absorption bands, a 285 nm and a 345 nm one, resulting from the $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ transitions, respectively. The availability of non-bonding and π electrons in ligand L is indicated by these absorption bands. The $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ bands are shifted to longer wavelengths for $[\text{Ni}(\text{L})_2]$, $[\text{Cu}(\text{L})_2]$ and $[\text{Co}(\text{L})_2]$ complexes, whereas they are shifted to shorter wavelengths for $[\text{Cd}(\text{L})_2]$ complexes. The LCMT (Ligand to metal charge transfer transition) caused a shift towards longer wavelengths in one of these bands after complex formation. $[\text{Ni}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complexes exhibited d-d transitions in their UV-Vis spectra at around 412 nm, 408 nm, 411 nm, and 421 nm, respectively [32]. The magnetic moment values of the $[\text{Ni}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complexes are 2.90, 3.94

and 1.84 BM, respectively. The magnetic moment value and UV-Vis spectral data suggest that the $[\text{Ni}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complexes [Fig-4(c-1,2)] probably are tetrahedral structure.

Fig. 3 presents the spectra and Fig. 4 (a, b, c & d) represent the proposed structure while Table 3 provides the values for the maximum wavelength (λ_{max}) and magnetic moment for the L, $[\text{Ni}(\text{L})_2]$, $[\text{Cd}(\text{L})_2]$, $[\text{Co}(\text{L})_2]$ and $[\text{Cu}(\text{L})_2]$ complex.

Table3: Electronic spectral data and magnetic moments for L and its metal complexes

Symbol of Compounds	Compound	λ_{max} nm	μ_{eff} B.M	Assignment
L	$\text{C}_{13}\text{H}_{10}\text{N}_4\text{O}_3$	268 336	---	$\pi \rightarrow \pi^*$ $n \rightarrow \pi^*$
$[\text{Ni}(\text{L})_2]$	$[\text{NiC}_{26}\text{H}_{18}\text{N}_8\text{O}_6]$	273 344 412	2.90	$\pi \rightarrow \pi^*$ $n \rightarrow \pi^*$ C.T (L \rightarrow M)
$[\text{Co}(\text{L})_2]$	$[\text{CoC}_{26}\text{H}_{18}\text{N}_8\text{O}_6]$	274 344 408	3.94	$\pi \rightarrow \pi^*$ $n \rightarrow \pi^*$ C.T (L \rightarrow M)
$[\text{Cu}(\text{L})_2]$	$[\text{CuC}_{26}\text{H}_{18}\text{N}_8\text{O}_6]$	261 342 411	1.84	$\pi \rightarrow \pi^*$ $n \rightarrow \pi^*$ C.T (L \rightarrow M)
$[\text{Cd}(\text{L})_2]$	$[\text{CdC}_{26}\text{H}_{18}\text{N}_8\text{O}_6]$	263 294 421	diamagnetic	$\pi \rightarrow \pi^*$ $n \rightarrow \pi^*$ C.T (L \rightarrow M)

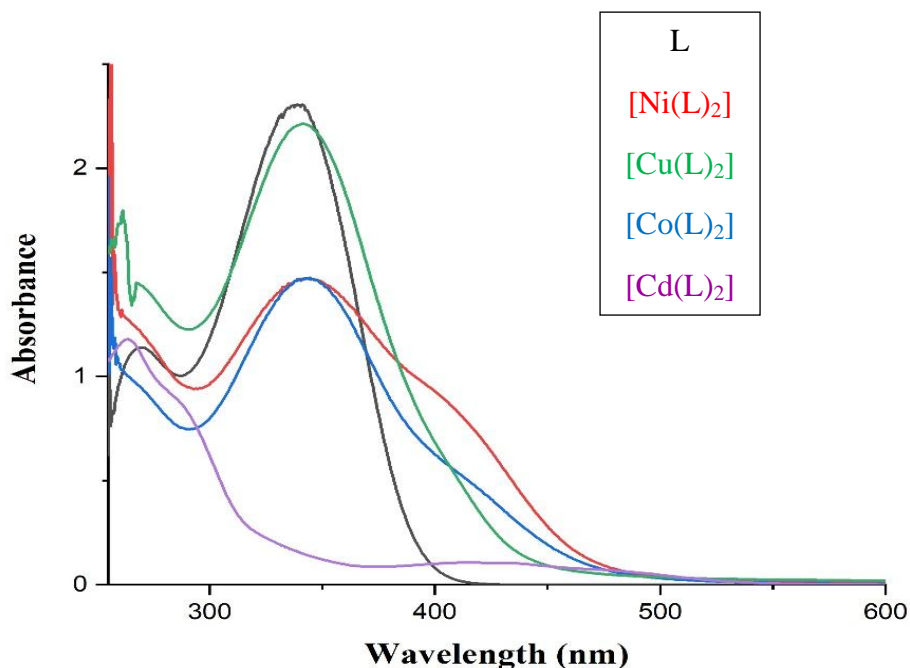


Fig. 3: Electronic spectra of ligand(L) & its metal complexes

UNDER PEER REVIEW

Fig. 4(a): Proposed Structure of [Ni(L)₂]complex (Tetrahedral)

Fig. 4(b): Proposed Structure of [Cu(L)₂]complex (Tetrahedral)

Fig. 4(c): Proposed Structure of [Co(L)₂]complex (Tetrahedral)

Fig. 4(d): Proposed Structure of [Cd(L)₂]complex (Tetrahedral)

3.4 Assessment of cytotoxicity

All the synthesized compounds exhibited significant larvicidal action against brine shrimp. The level of mortality was directly correlated with the content of the extract [33]. While the highest number of deaths occurred at a dose of 100 µg/mL, the lowest mortality rate was seen at concentrations of 6.25 µg/mL which is shown in Table 4. It was noted that at greater concentrations of treatment chemicals, the shrimps began to die after 8 hours, and all the shrimps had died within 24 hours [34]. The LC₅₀ values obtained from the brine shrimp lethality bioassay, as shown in Table 4 (Fig. 5), were 12.92, 16.67, 30.52, 11.46, and 5.46 µg/mL for ligand(L), [Cu(L)₂], [Co(L)₂], [Ni(L)₂], and [Cd(L)₂], respectively. When compared to the positive control vincristine sulphate, with a lethal concentration 50% of 12.09 µg/mL, the complexes [Ni(L)₂] and [Cd(L)₂] shown significant larvicidal action against brine shrimp. These complexes have the potential to be further studied [35].

Table 4: The data from the lethality bioassay of brine shrimp

Sample	Conc. of Sample (µg/mL)	Log conc. (µg/mL)	Number of nauplii used	Number of nauplii survived (After 24h)	Percentage of mortality	Probit value	LC ₅₀ Value (µg/mL)
Standard (vincristine sulphate)	6.25	0.7959	30	23	23	4.26	12.09
	12.5	1.0969	30	14	57	5.18	
	25	1.3979	30	10	67	5.44	
	50	1.6990	30	5	83	5.95	
	100	2.0000	30	0	100	8.95	
L	6.25	0.7959	14	12	14.29	3.92	12.92 ± 1.49
	12.5	1.0969	14	6	57.14	5.18	
	25	1.3979	14	4	71.43	5.55	
	50	1.6990	14	1	92.86	6.41	
	100	2.0000	14	1	92.86	6.41	
[Cu(L) ₂]	6.25	0.7959	14	12	14.28	3.92	16.67 ± 2.66
	12.5	1.0969	14	7	50	5.00	
	25	1.3979	14	5	64.28	5.36	
	50	1.6990	14	4	71.43	5.55	
	100	2.0000	14	1	92.85	6.41	
[Co(L) ₂]	6.25	0.7959	13	12	7.69	3.52	30.52 ± 2.87
	12.5	1.0969	13	11	15.39	3.96	
	25	1.3979	13	9	30.77	4.48	
	50	1.6990	13	2	84.62	5.99	
	100	2.0000	13	1	92.31	6.41	
[Cd(L) ₂]	6.25	0.7959	14	6	57.14	5.18	5.46 ± 1.106
	12.5	1.0969	14	5	64.28	5.36	
	25	1.3979	14	3	78.57	5.77	
	50	1.6990	14	1	92.85	6.41	
	100	2.0000	14	0	100	8.95	
[Ni(L) ₂]	6.25	0.7959	14	8	42.86	4.82	11.46 ± 1.84
	12.5	1.0969	14	7	50	5.00	
	25	1.3979	14	6	57.14	5.18	
	50	1.6990	14	4	71.43	5.55	
	100	2.0000	14	2	85.71	6.04	

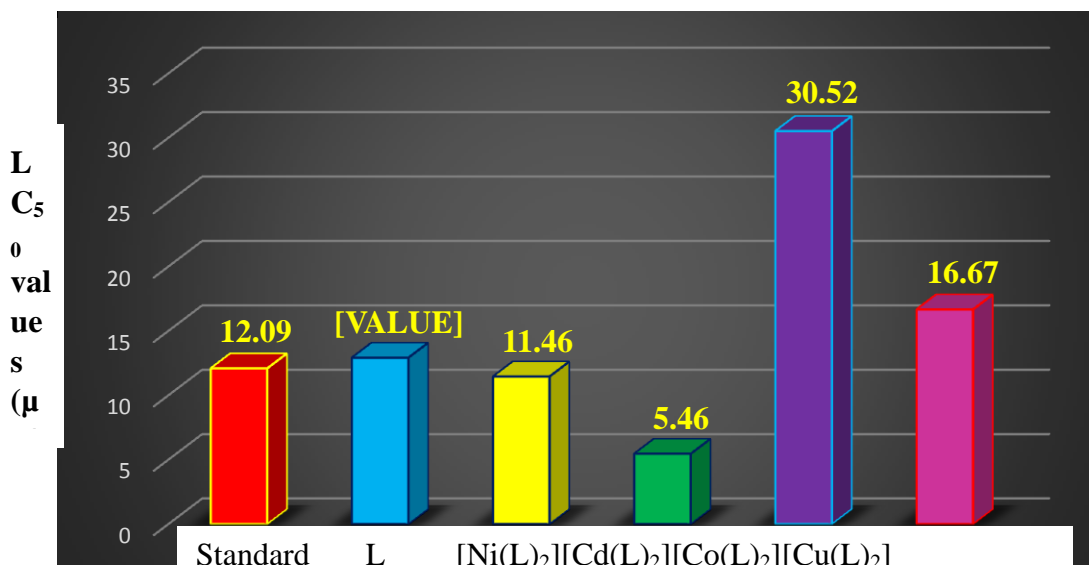


Fig. 5: LC₅₀ values of sample and standard

3.5 Antibacterial evaluation

The ligand and complex antibacterial activity are assessed by means of the paper disc diffusion method with respect to *P. aeruginosa*, *B. cereus*, *E. coli*, and *S. aureus* [36]. The diameter of the inhibitory zone was determined in millimeters and the results were compared with the standard antibacterial medication Kanamycin-30 shown in Table 5 and represented in Fig. 6 [37, 38]. It had been found that the ligands and the metal complexes showed a significant impact on killing every kind of pathogenic bacteria [39]. There was an increase in bacteriostatic activity compared to the parent ligand in all the complexes [40]. The antibacterial efficiency of the [Cd(L)₂] and [Ni(L)₂] complexes was higher than that of the unbound L against nearly all the bacteria tested. The [Cu(L)₂] complex was more active than L against *E. coli* and *S. aureus* bacteria, while it was moderately active or nearly as active against *P. aeruginosa*, *B. cereus*. The L was more active than the [Co(L)₂]. The enhanced activity of the metal complexes is explained by chelation hypothesis.[41].The inclusion of nitrogen (N) and oxygen (O) donor groups in the ligand and its metal complexes suppressed the synthesis of enzymes. Chelation decreases the polarity of the central ion by partially sharing its positive charge with the donor groups.

Table 5: Antibacterial activities (zone of inhibition in mm) of samples against pathogens

Samples	<i>P. aeruginosa</i>	<i>E. coli</i>	<i>B. cereus</i>	<i>S. aureus</i>
Kanamycin-30	27	24	25	26
L	12	11	13	12
[Cu(L) ₂]	12	14	13	13
[Cd(L) ₂]	15	16	15	17
[Co(L) ₂]	5	6	8	7
[Ni(L) ₂]	13	12	14	14

The electron delocalization within the chelating ring also enhances the lipophilic properties of the central atom, promoting its ability to pass through the lipid layer of the membrane [42]. The increased activity of the complex may also be attributed to the intrinsic characteristics of the metal ion in causing the precipitation or denaturation of proteins. Given that enzymes are composed of proteins, it is reasonable to anticipate that the heavy metal would deactivate these catalysts.

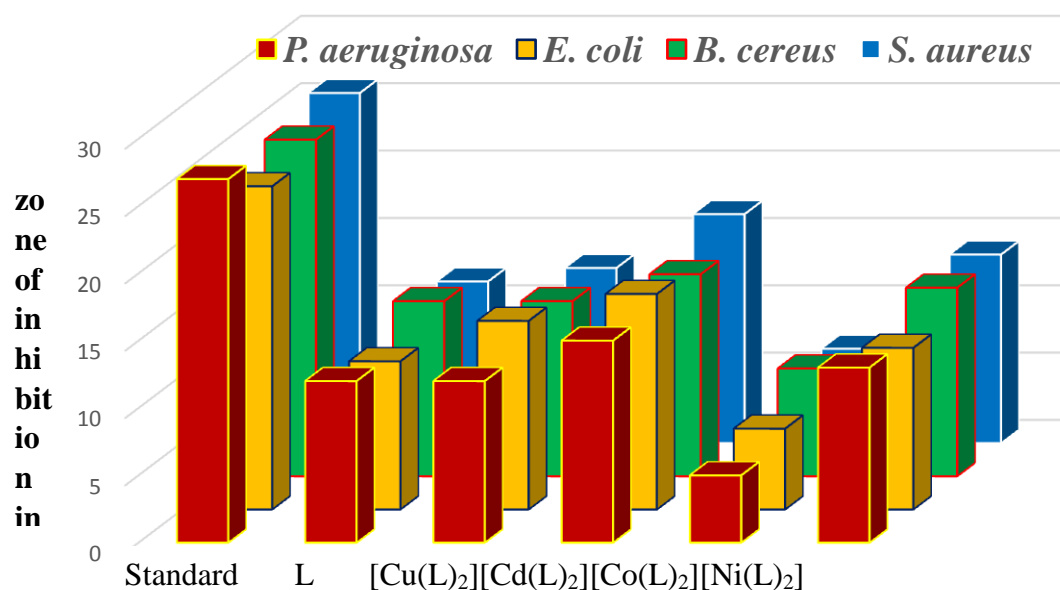


Fig. 6: Zone of inhibition of samples and standard against *P. aeruginosa*, *E. coli*, *S. aureus*, and *B. cereus*

4. CONCLUSION

The N'-(4-nitrobenzylidene) isonicotinothiazide (L) was used to build metal complexes with Cu (II), Cd (II), Ni (II), and Co (II) ions. The ligand and its metal complexes underwent analysis using TLC, melting point determination, conductivity measurement, FTIR spectroscopy, magnetic moment measurement, mass spectrometry, and UV-Vis. spectroscopy. The tetrahedral structure of the [Cu(L)₂], [Cd(L)₂], [Co(L)₂], and [Ni(L)₂] complexes is supported by the magnetic moment value and UV spectral data. The L and its metal complexes exhibited biological activity and showed cytotoxicity towards brine shrimp nauplii. The complexes [Cd(L)₂] and [Ni(L)₂] exhibited greater cytotoxic activity. Also, the antibacterial activity of L and its complexes was assessed against several microorganisms. The complexes [Cu(L)₂], [Cd(L)₂], and [Ni(L)₂] showed higher antibacterial activity compared to the ligand (L).

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