Indian Journal of Chemistry
Vol. 64, August 2025, pp. 761-767
DOI: 10.56042/ijc.v64i8.11412

Synthesis, spectral and biological evaluation of *in vitro* antimicrobial activity and molecular docking studies of Schiff base ligand 1,2-cyclohexadiene/*o*-vanillin and its transition metal complexes

J Priya*, S Manimalathi & D Madheswari
Government Arts College for Women, Salem 636 008, Tamil Nadu, India
E-mail priyaasstprofessor@gmail.com, smmalathislm@gmail.com

Received 27 May 2024; accepted (revised) 28 July 2025

Exploring application of cost-effective transition metal-based drugs with high pharmaceutical properties has led to the progression of Schiff-base centred medicinal chemistry. Following this line, herein a series of new class of transition metal complexes Ni(II), Cd(II), Mn (II) and Zn(II) have been synthesized using a Schiff base ligand derived from 2 moles of *o*-vanillin and 1 mole of 1,2-cyclohexanediamine. The elemental analysis, conductivity, ^{1}H NMR, UV-Vis, IR spectra and TGA studies have been used to characterize the as-synthesized ligand together with associated metal complexes. Additionally, testing the antimicrobial properties against Gram-positive (*S. aureus*) and Gram-negative (*E. coli*) bacteria establishes a higher inhibiting efficiency and high-points the role of the compounds in stronger antibiotic use action. As a result, the current study acknowledges the potential of the new Schiff-base derived complexes as prescription medication.

Keywords: Schiff base ligand, *o*-Vanillin, 1,2-Cyclohexanediamine, Metal complexes, Molecular docking, Microbial activity

The chemistry of coordination compounds has always a contest to the inorganic chemists as it has more diversions presently. Many factors namely structural flexibility, chemical properties as well as ease of synthesis and tunable co-ordination features, Schiff base ligands and their complexes represent research-centric materials. In addition, coordination complexes of Schiff bases have been studied extensively because of their unique magnetic characteristics, innovative structural features, and physiological significance[1-3].

The Schiff bases are the condensation products of the carbonyl and amino compounds. Because they can easily coordinate with metal ions through the nitrogen atom of azomethine, the C=N bond that results from this process is crucial for enhancing biological function[4,5]. As a result, it was revealed that a number of compounds containing azomethine have exceptional diuretic, antibacterial, antifungal, and anticancer properties. Schiff bases are widely used in the food, dye, and agrochemical industries as well as in heterogeneous catalysis, fungicidal, and biological sectors[6,7]. Due to the reactivity of this type of multidentate core, Schiff base ligand and transition metal complexes with different donor atoms in the ligands have been investigated extensively in this context to create metal complexes.

The aromatic moieties of Schiff base have an important role in supporting the capacity of metal complexes to bond between base pairs of biomolecules. Schiff base coordinates with a metal ion, resulting in a metal complex that may have biological and pharmacological properties. With the aim of synthesizing salicylaldehyde derivative of Schiff base, we have successfully prepared *o*-vanillin and 1,2-cyclohexanediamine comprising ligand and characterized it using spectral studies. The transition metal complexes of various Schiff bases containing 3,5-dichlorosalicylaldehyde and trans 1,2-diaminocyclohexane as core unit and their anticancer properties were reported earlier from our laboratory[8,9]. We have examined potent anticancer properties of histidine derived Schiff base metal complex earlier[10].

In this manner, significant endeavors are being made to displace expensive medication with reasonable choices, and various transition metal complexes have been combined and their cytotoxic properties were examined. This has given inspiration to develop prodrugs, which respond at explicit destinations along the DNA strand, as receptive models for protein-nucleic communications[11]. Mostly, the extent of DNA interaction with metal complexes is governed by the structure, planarity, coordinating

atoms, nature of the ligands and hydrophobicity. Herein, we have performed the *in vitro* molecular docking studies of synthesized metal complexes and evaluated their suitability as a drug. In this work, mononuclear NO donor Schiff base ligand and their structural and biological evaluation has been chosen for the up-to-date. The antimicrobial studies suggested the metal complexes show significantly enhanced antibacterial and antifungal activities.

Experimental Section

Having purchased from commercially available sources at Loba Chem. Ltd., Merck, all of the reagents and solvents were utilized without additional purification. Hexane/ethyl acetate was used as the solvent solution for the mobile phase in TLC, which was carried out on Merck pre-coated aluminum sheets of 60 F-254 silica gel plates. UV and visible light as well as iodine spray were also used. The Buchi Melting Point B-545, an electrothermal device with uncorrected capillary tubes, was used to measure melting points. Proton chemical shifts (δ) are measured using a Bruker 400 MHz or 100 MHz high resolution NMR spectrometer, respectively, and the solvent (DMSO-d_6 at 2.51 ppm) is used as a reference for the ^{1}H and ^{13}C NMR spectra. Using KBr disks, FT-IR spectra were recorded on a Bruker FT-IR27 spectrometer. Agilent 6520 Q-TOF spectrometer. Organic solvents utilized for experiments were of reagent grade. The antibiotic was purchased from Egyptian markets and used as standard for antifungal activities.

Synthesis of Ligand

o-Vanillin (2 mmol) and 1,2-Cyclohexanediamine (1 mmol) were added successivelyto ethanol (50 mL). The resulting solution was stirred for 30 minutes in an ice bath. The brown yellow coloured Schiff base obtained was filtered and recrystallized from ethanol.

Synthesis of Metal Complexes

Ni(II), Cd(II), Mn(II) and Zn(II) complexes of the Schiff base ligand were prepared by combining the ligand with equimolar amounts of $M(AcO)_2.\ 4H_2O$ where M= Mn(II), Ni(II), Cd (II) Zn(II), respectively in 30 mL of ethanol for 1 h under constant stirring. The contents were transformed to a petri dish to allow the solvent to evaporate slowly. The crystalline complexes formed were filtered and recrystallized from ethanol (Scheme 1).

As-synthesized metal complexes namely Mn, Ni, Cd and Zn were designated further as A, B, C and D respectively. The ligand is referred as L.

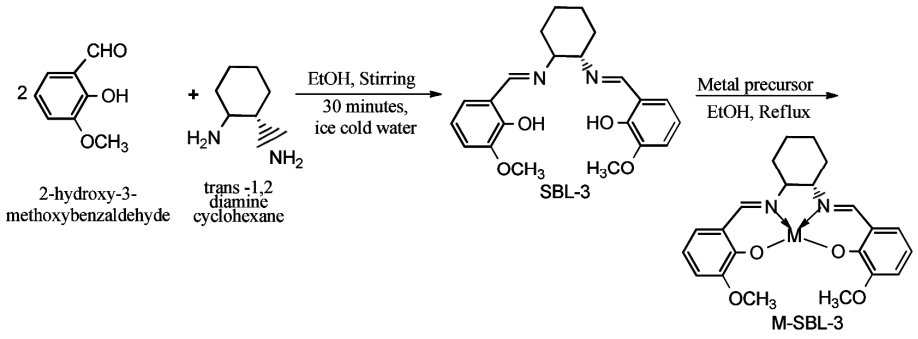
Results and Discussion

UV spectra

The ligand and metal complexes' UV-Vis spectra, which were captured in a DMSO solution at room temperature, are displayed in Fig. 1. Three bands were observed by the ligand at 418, 294, 261 and 214 nm, which were ascribed to the electronic transitions π → π*, π → π* and n → π*, respectively. The n→ π* transition of the azomethine group is also attributed to the bands in the 418-294 nm region. In addition to electronic changes of the metal d orbitals (d-d electronic transition), metal complexes (A-D) display similar electronic transition as seen in the ligand[11,12] (Fig. 1).

FT-IR spectra

The infrared spectrum of ligand showed certain characteristic stretching bands at 1630 and 1126 cm^{-1} assigned to υ (C=N) and υ (C–O) υ (C=N) bands. However, in the spectra of (A-D) metal complexes these bands were got shifted to a lower wave number compared to the ligand, signifying that the coordination took place *via* the nitrogen atom of the ligand. The FT-IR spectra of L and (A-D) are represented in Fig. 2. In addition, the stretching of



Scheme 1 — Schiff base ligand and metal complexes

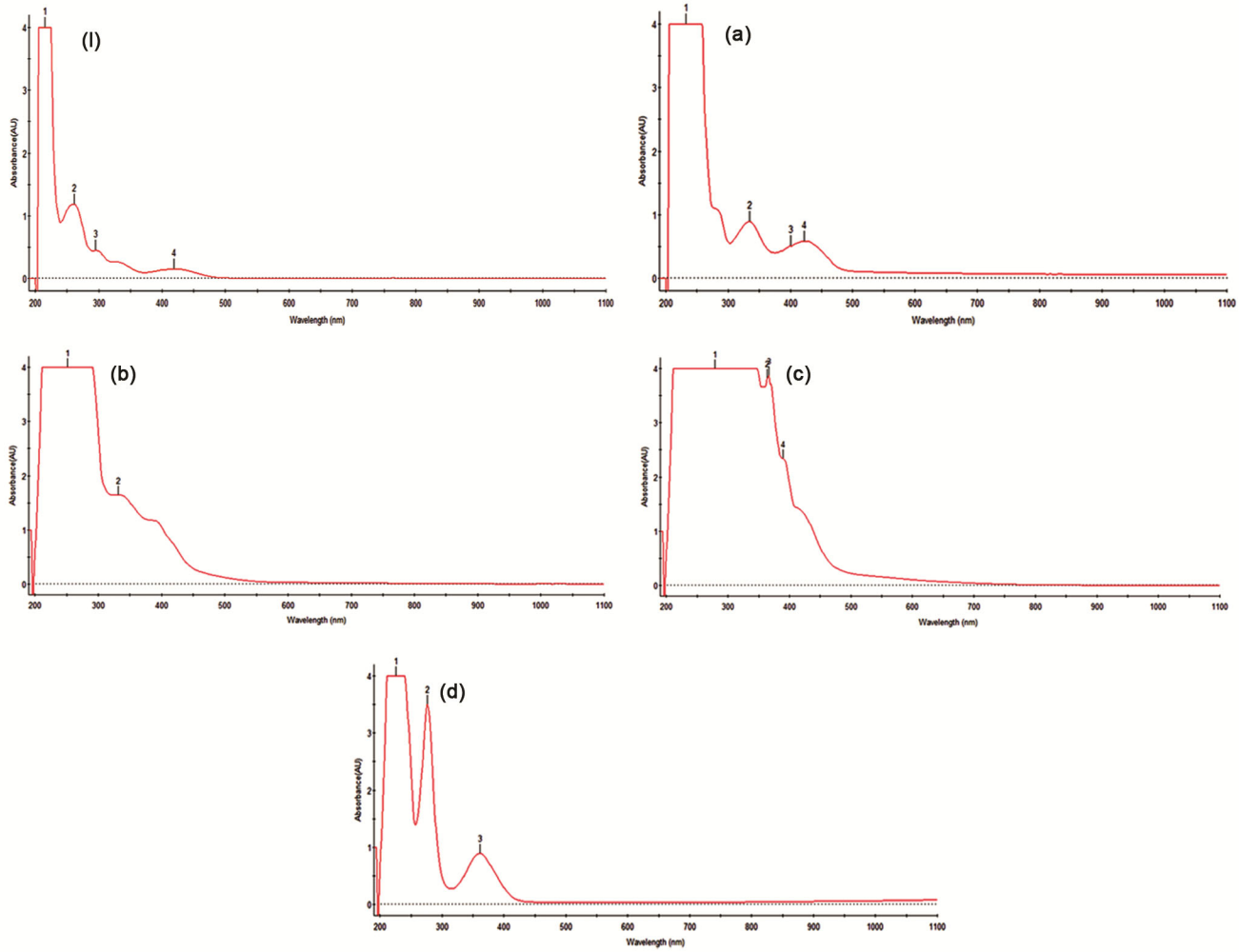


Fig. 1 — Electronic spectra of ligand and metal complexes

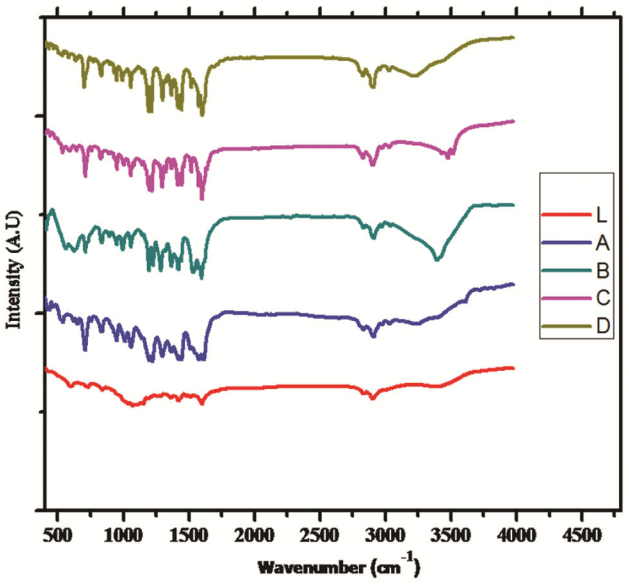


Fig. 2 — FTIR spectra of ligand and metal complexes

metal to oxygen and metal to nitrogen bands of the complexes appeared in the lower[13-15] wave number region appearing in the range of 550-490 and 491-450 cm^{-1} also suggesting the complexation through nitrogen and oxygen atoms of the ligands.

The ^{1}H NMR spectra of the complexes, in reference with that of the ligand, present significant changes due to the coordination process[14]. The -NH proton signal of Schiff base ligand (7.80 ppm) disappears due to complexation with Mn, Ni, Cd and Zn (Fig. 3). The aromatic protons and the methyl protons do not seem to show any changes as a result of coordination.

ESI Mass spectra

The fragmentation of the target molecule is seen in the mass spectrum as an ordered collection of peaks that represent each of the different fragments. The intensity indicates the level of stability the Schiff base

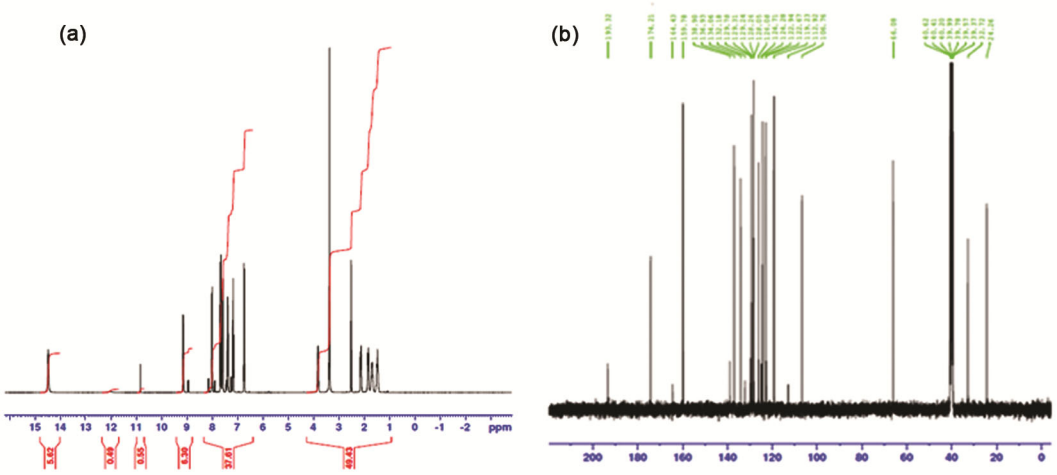


Fig. 3 — (a) ^{1}H NMR and (b) ^{13}C NMR spectra of ligand

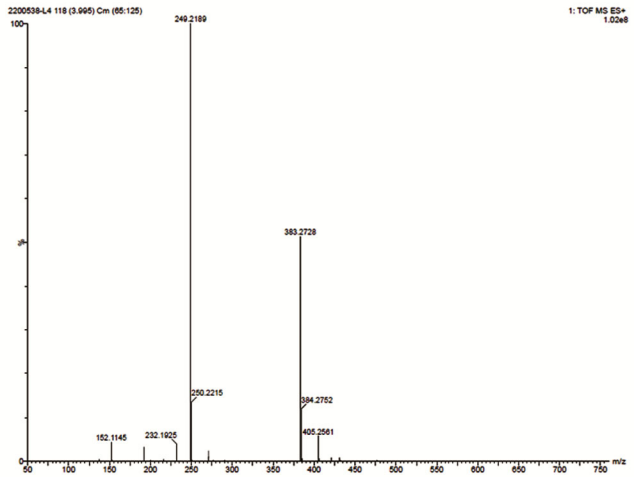


Fig. 4 — The mass spectra of the ligand

fragments and metal complexes are. Fig. 4 displays the ESI-MS spectra for L and metal complexes. For the ligand (L), a mass peak is seen in the m/z 383 region. It suggests the creation of the M+1 peak. The 1:1 ligand peak is shown by the fragment peak at 249 m/z. The spectra were shown in the ESI-MS (Fig. 4). Molecular ion peaks and the mass spectra of ligands and their metal complexes were used to confirm the postulated formula mass. Elemental analysis and molar conductance of the Schiff base ligand and their complexes were shown in Table 1.

Thermogravimetric Analysis

The thermogravimetric analysis (TGA) curves of the metal complexes, recorded over a temperature range of 20°C to 700°C, are shown in Fig. 5. The TGA graphs reveal that thermal decomposition occurs in three distinct and sequential steps[16]. The initial step involves the removal of coordinated water molecules in the temperature range of 100–120°C, resulting in a 5% weight loss. In the second step, as the temperature rises above 500°C, the ligand moiety undergoes decomposition, with a mass loss of 25.05%, leaving behind a metal oxide (MO) residue. This phase represents the major breakdown of the complex. The TGA data also provide information about stability ranges, weight loss percentages, and residue percentages. After complete decomposition, the complexes exhibit approximately 80% weight loss. The final weight loss corresponds well with the calculated values for the conversion of the complexes into their respective metal oxides, confirming the thermal stability and decomposition pathway of the complexes.

Biological screening studies

Molecular Docking Studies

Docking with BSA protein molecule

Using the Autodock Vina program, metal complexes (A–D) had been docked with the BSA protein (PDB ID: 3V03) molecule. The protein molecule consists of two chains, A and B. Target amino acids were used to bind the synthesized complexes. The complexes (A-D) had binding energy values of -7.1, -9.3, -8.1, and -6.2 kcal/mol, in that order. Of all the created complexes, complex 2 exhibits superior activity toward the protein molecule. Complex A intermingle with Glu357 and Arg208 residues as represented sturdily. Nonetheless, the other complexes (B-D) interact with Glu125, Leu582 and Gln416 with strong degree of interaction. The interaction values were

Table 1 — Elemental analysis and molar conductance of the Schiff base ligand and their complexes

S. No.	Compd	Mol. Formula	Found (Calcd) %					Mol. Wt.	Molar conductance Λ_m ($ohm^{-1}cm^2mol^{-1}$)
			C	H	N	O	M		
1	Ligand	$C_{22}H_{26}N_2O_4$	69.09	6.85	7.32	16.73		381.45	–
			Metal Complexes						
2	[Mn(CBMN)] (A)	$C_{22}H_{24}N_2O_4Mn$	60.69	5.56	6.43	14.70	12.62	435.38	5.85
3	[Ni(CBMN)] (B)	$C_{22}H_{24}N_2O_4Ni$	60.17	5.51	6.38	14.57	13.37	439.13	3.40
4	[Cd(CBMN)] (C)	$C_{22}H_{24}N_2O_4Cd$	59.27	5.43	6.28	14.36	14.67	444.82	3.80
5	[Zn(CBMN)] (D)	$C_{22}H_{24}N_2O_4Zn$	53.61	4.91	5.68	12.99	22.81	492.85	4.95

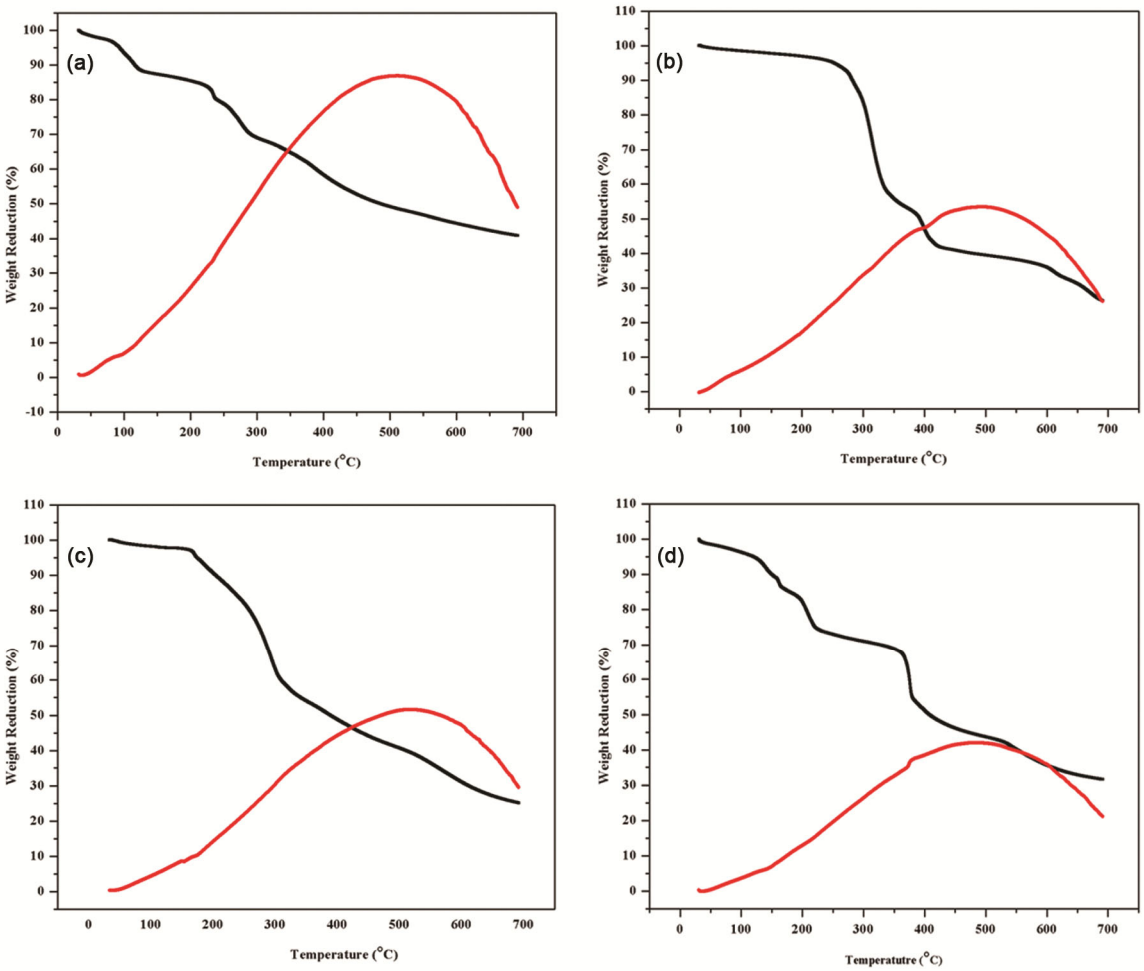


Fig. 5 — TGA graph of synthesized metal complexes (a) complex A, (b) Complex B, (c) Complex C and (d) Complex D

tabulated in Table 2 and the docking images were as shown in Fig. 6.

Docking with DNA

Using the Autodock Vina program, the as-prepared metal complexes (A–D) were docked with the DNA molecule (PDB ID: 1Z3F)[17]. The DNA residue's nucleotide is utilized in the docking process, and Fig. 7 shows the final image. The results' binding interaction values are tabulated in Table 3. The binding energies of the produced metal complexes (A-D) are -6.3, -5.8, -7.1, and -6.3 kcal/mol, respectively. The molecular docking results revealed that metal complexes are capable of interacting with both protein and DNA molecules effectually.

Antibacterial activity

Antibacterial activity is one of the essential properties for the development of medications and pharmaceuticals. The *in vitro* antibacterial activity of

Table 2 — BSA docking studies interactions

S. No.	Complex	Interaction	Distance (Å)
1	[Mn(CBMN)] (A)	Glu357, Phe205, Ala209, Arg208	3.62, 5.05, 4.10, 4.08
2	[Ni(CBMN)] (B)	Glu125, Leu122, Lys132, Lys136	2.63, 5.49, 4.52, 5.16
3	[Cd(CBMN)] (C)	Ala583, Leu582, Ala538, Lys535, Lys537, His534, Pro498	4.05, 3.33, 5.43, 4.65, 5.14, 4.65, 4.35
4	[Zn(CBMN)] (D)	Lys499, Val497, Gln416, Val468	5.16, 3.55, 2.34, 3.91

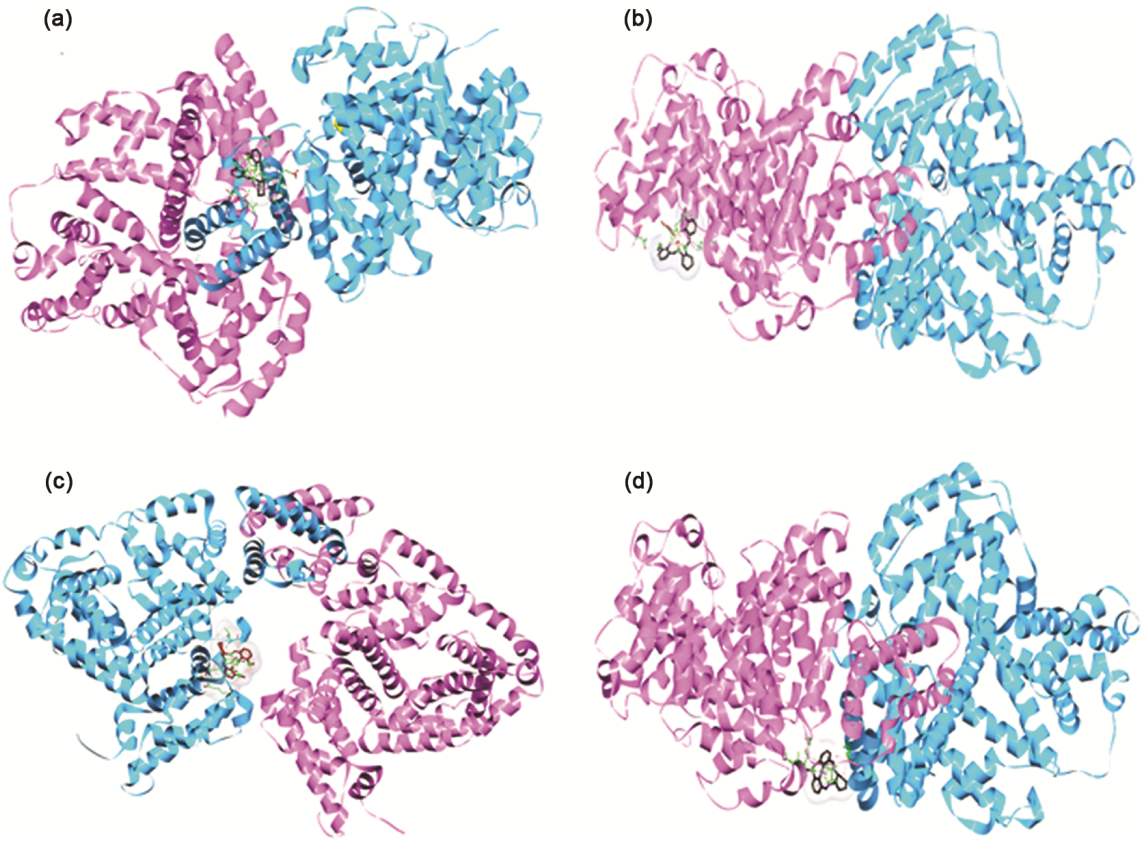


Fig. 6 — The molecular docking interaction images of BSA protein and metal complexes

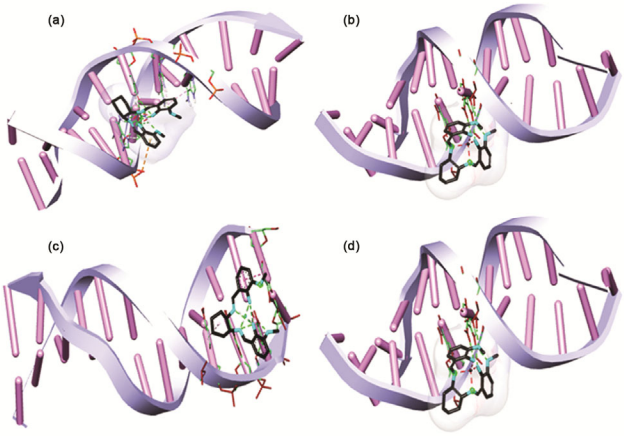


Fig. 7 — The molecular docking interaction images of DNA and metal complexes

the Schiff base metal complexes was evaluated against both Gram-positive (*S. aureus*) and Gram-negative (*E. coli*) bacteria. Clotrimazole and streptomycin were used as controls[18]. The antibacterial results for the metal complexes are summarized in Table 4. The findings from the antibacterial study indicate that the complex B exhibits superior activity against both Gram-positive and Gram-negative bacteria compared to the other metal complexes under investigation, as shown in Fig. 8. The complex B also demonstrated high antibacterial activity and a strong tendency to interact with DNA and BSA proteins, showing significant binding affinity at their respective binding sites.

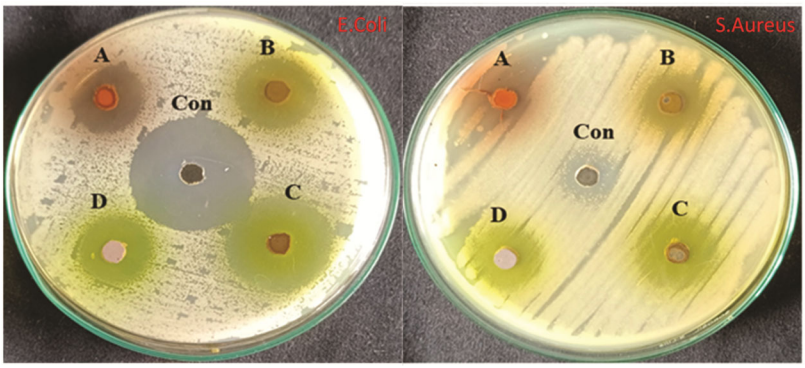


Fig. 8 — The antibacterial activity of the ligand and metal complexes measured by viable count method against *E. coli* and *S. Aureus* bacterial strains

Table 3 — DNA docking studies interactions

S.No.	Complex	Interaction	Distance (Å)
1	[Mn(CBMN)] (A)	Ade6, Ade17, Ade18	2.22, 2.32, 2.54
2	[Ni(CBMN)] (B)	Ade5, Ade6, Ade17, Ade18	4.97, 5.17, 3.14, 2.07
3	[Cd(CBMN)] (C)	Ade5, Ade6, Ade17, Ade18, Thy7	5.24, 4.94, 4.94, 3.14 2.57
4	[Zn(CBMN)] (D)	DC21, DG22, DC23, DG24, DC1	4.86, 3.01, 5.09, 3.65, 4.80

Table 4 — Antibacterial activity of compound A-D

Name of metal complex	Zone of Inhibition (mm) (*E. coli*)		Zone of Inhibition (mm) (*S aureus*)	
		Control		Control
[Mn(CBMN)] (A)	21	15	12	18
[Ni(CBMN)] (B)	21	15	8	16
[Cd(CBMN)] (C)	21	15	11	14
[Zn(CBMN)] (D)	21	15	11	15

Conclusions

In an effort to develop an effective and safe antimicrobial agent, we focused on synthesizing a novel Schiff base ligand using *o*-vanillin and trans-1,2-cyclohexanediamine, along with the corresponding metal complexes. The appearance of the azomethine peak at 1630 cm^{-1} confirms the successful formation of the ligand from *o*-vanillin and trans-1,2-cyclohexanediamine. Spectral studies were employed to synthesize and characterize transition metal complexes of Mn(II), Ni(II), Cd(II) and Zn(II) in addition to the Schiff base ligand. The multidentate core, with multiple atoms involved, significantly enhances the variety of metal coordination. The antimicrobial effectiveness of the complexes was demonstrated in this study, showcasing their potential for *in vitro* applications.

References

1 Kulkarni Y D & Rowhani A, *J Indian Chem Soc*, 67 (1990) 46.
2 West D X, Liberta A E, Padhye S B, Chikate P B, Sonawane A S, Kumbhar O & Yerande R G, *Coord Chem Rev*, 49 (1993) 123.
3 Singh A, Lalita, Dhakarey R & Saxena G C, *J Indian Chem Soc*, 73 (1996) 339.
4 Molina R. H & Mederos A, *In Comprehensive Coordination Chemistry II, A.B.P. Lever (Ed.), Vol. 1*, (Elsevier-Pergamon Press, Amsterdam-Oxford- New York), (2003) 411.
5 Garnovskii A D & Kharisov B I, *Synthetic Coordination and Organometallic Chemistry*, (Marcel Dekker, New York-Basel), (2003) 513.
6 Hossain A M S, M´endez-Arriaga J M, Xia C & Gomez-Ruiz J X S, *Polyhedron*, 217 (2022) 115692.
7 Guo Z, Xing R, Liu S, Zhong Z, Ji X, Wang L & Li P, Res, 342 (2007) 1329.
8 Manimalathi S & Madheswari D, *Indian J Chem A,* 61 (2022) 965
9 Priya J & Madheswari D, *J Biosci*, 47 (2022) 29.
10 Sridevi N & Madheswari D, *Indian J Chem Sec A*, 59 (2020) 1768.
11 Gupta K C & Sutar A K, *Coord Chem Rev*, 252 (2008) 1420.
12 Guo Y, Wang J, Ma X, Wei Z, Wang W, Zhang C & Lei Y, *J Energy Storage*, 48 (2022) 103956.
13 Devi N, Sarma K, Rahaman R & Barman P, *Dalton Trans*, 47 (2018) 4583.
14 Sacconi L, Ciampolini M & Campigli U, *Inorg Chem*, 4 (1965) 407.
15 Palmer R A & Piper T S, *Inorg Chem*, 864 (1966). (Kindly provide valid page number)
16 Akinremi C A & Obaleye J A, *Int J Med Biomed Res*, 1 (2012) 24.
17 Khan M, Ali M & Juyal D, *Pharma Innov J*, 6 (2017) 73.