

Synthesis, Characterization and Evaluation Anti-cancer Activity of Fe(III), Co(II), Ni(II) and Cu(II) Complexes Derived from Heterocyclic Schiff bases Ligands

Lubna Khalid Farhan; Mohamed Ahmed Awad and Abdullah Hussein Khash

Department of Chemistry, Education College for Pure Science, University of Anbar /Iraq

Abstract

In this work, two heterocyclic Schiff bases ligands derived from 2-aminopyrimidine with 2-hydroxybenzaldehyde (L_1) and 2,6-diaminopyridine with 4-chlorobenzaldehyde (L_2) were synthesized. These ligands used to synthesis complexes of Fe(III), Ni(II), Co(II), and Cu(II). The synthesized compounds have been characterized by FT-IR, $^1\text{H-NMR}$ and UV-Vis techniques for the ligands and FT-IR, UV-Vis, all reactions monitored by TLC, molar conductivity and magnetic susceptibility measurements for the corresponding complexes. General formula of complexes are $[\text{Cu}(L_1)_2(\text{H}_2\text{O})_2]$, $[\text{Co}(L_1)_2(\text{H}_2\text{O})_2]$, $[\text{Ni}(L_1)_2(\text{H}_2\text{O})_2]$, $[\text{Fe}(L_1)_2\text{Cl}_2]\text{Cl}$, $[\text{Cu}(L_2)_2(\text{H}_2\text{O})_2]$, $[\text{Co}(L_2)_2(\text{H}_2\text{O})_2]$, $[\text{Ni}(L_2)_2(\text{H}_2\text{O})_2]$ and $[\text{Fe}(L_2)_2\text{Cl}_2]\text{Cl}$. All complexes are paramagnetic. The results of the molar conductivity measurements indicated that all complexes are non-electrolytes in (DMF) except the complexes $[\text{Fe}(L_2)_2\text{Cl}_2]\text{Cl}$ and $[\text{Fe}(L_1)_2\text{Cl}_2]\text{Cl}$ are electrolyte with a molar ratio (1:1). An octahedral geometry for all the complexes of. The ligands are bidentate, (L_1) through phenolic (OH) and azomethine nitrogen, and (L_2) through two azomethine nitrogen. Moreover, anticancer activity against ($L_{20}\text{B}$) cell line of metal complexes was tested.

Keywords: Heterocyclic Schiff bases; 2-aminopyrimidine; 2-hydroxybenzaldehyde; 2,6-diaminopyridine; 4-chlorobenzaldehyde; anticancer activity

INTRODUCTION

The importance of heterocyclic Schiff-base ligands and their metal complexes is ascribed to their vast applications in chemical and biological fields^[1,2]. On the other hand, Schiff-base ligands are considered to be the most interesting, due to the presence of hard (N) or (O) and soft (S) donor atoms in their structure and high stability of corresponding complexes^[3]. Schiff-base complexes play as models in many of biological system such as urease^[4]. Recent studies showed that some Schiff bases increased their activity when administrated as metal-complexes rather than as organic compound^[5]. Schiff base complexes played important role in organic chemistry due to their uses as catalysts in many of organic reaction such as oxidation and epoxidation reactions^[6]. Ni(II) and Cu(II) complexes of tetradentate Schiff-base ligand showed calf thymus DNA interaction, anti-bacterial and anti-fungal activities^[7]. Some of Schiff base complexes have been exhibiting a surface activity, Fe(III), Co(II) and Mn(II) complexes of tannic acid Schiff-base surfactants showed surface tension reduction and show activity against bacteria and fungi^[8]. Metal complexes of salicylaldehydes showed liquid crystal properties^[9]. Moreover, the biological studies of interaction between DNA and transition metal complexes have gained more interests due to their use in chemotherapy^[10,11].

In the work we present two Schiff bases ligands derived from 2-aminopyrimidine with 2-hydroxybenzaldehyde (L_1) and 2,6-diaminopyridine with 4-chlorobenzaldehyde (L_2), with some transition metals were synthesized. Also, the structure of prepared ligands were checked by FT-IR, $^1\text{H-NMR}$, UV-Vis techniques and prepared complexes were characterized by FT-IR, UV-Vis, molar conductivity and magnetic susceptibility measurements.

EXPERIMENTAL

Materials

Chemicals and reagents used in this work: 2-aminopyrimidine, 2-Hydroxybenzaldehyde, 2,6-Diaminopyridine, 4-Chlorobenzaldehyde, FeCl_3 , $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{Cu}(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$, were obtained from SIGMA-ALDRICH Chemical Co. which used as received without further purification. Solvents were obtained from SCHARLAU and ROMIL.

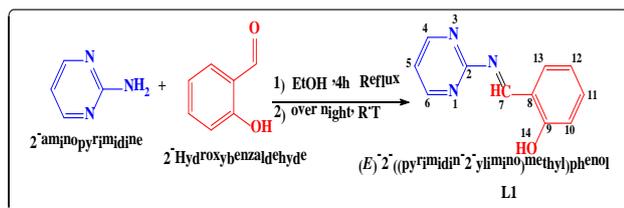
Instrumentation:

Infrared spectra were recorded as KBr pellets on Bruker-Tensor 27 spectrometer (to characterize the ligands), Shimadzu (FT-IR)-8400S spectrophotometer in the range ($4000\text{-}400\text{ cm}^{-1}$) to characterize the complexes. $^1\text{H-NMR}$ spectra were recorded on Bruker-400 MHz spectrometer using DMSO-d_6 as a solvent. UV-Vis Spectrophotometer (23-1884-01-0059) used to record UV-Vis spectra, Conductivity measurements were recorded in (DMF) using Jenway conductivity meter model 4070. Magnetic Susceptibility was determined with a Balance Magnetic Susceptibility, model: MSB-MKI. Melting points were conducted using an Electro-Thermal Stuart Melting Point Apparatus Jenway.

Procedures:

Synthesis of schiff base ligand (L_1):

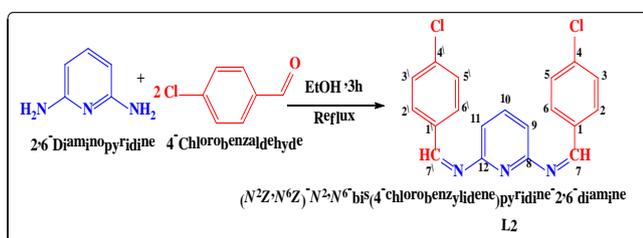
An ethanolic solution (20 mL) of 2-aminopyrimidine (1gm, 10.5mmol) was added to (20 mL) ethanol solution of 2-hydroxybenzaldehyde (1.28gm, 10.4mmol) in 100 mL round bottom flask, the reaction mixture refluxed for 4h then left stirred at r.t. overnight, white precipitate was obtained, filtered off, washed by EtOH and recrystallized from EtOH, yield 63.6%, mp $152\text{-}154\text{ }^\circ\text{C}$, TLC: $R_f = 0.81$ (Toluene: Methanol) (7:3). The synthetic route of (L_1) in scheme 1.



Scheme1. Synthetic route for synthesis of schiff base ligand (L_1)

Synthesis of schiff base ligand (L_2):

An ethanolic solution (20 mL) of 2,6-diaminopyridine (1 gm, 9.1 mmol) was added to (20 mL) ethanol solution of 4-chlorobenzaldehyde (2.55 gm, 18.2 mmol) in 100 mL round bottom flask, the reaction mixture refluxed for 3h, light yellow precipitate was obtained, filtered off, washed by EtOH and recrystallized from EtOH, yield 87.9%, mp 262 (dec.)°C, TLC: $R_f = 0.90$ (Toluene: Methanol) (7:3). The synthetic route of (L_2) in scheme 2.

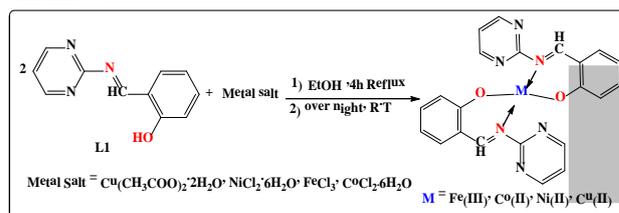


Scheme2. Synthetic route for synthesis of schiff base ligand (L_2)

Synthesis of metal complexes (L_1):

(0.39 gm, 2 mmol) of (L_1) was dissolved in (20 mL) of ethanol in a 100 mL round bottom flask with heating and stirring. After that, (0.80 gm and 2 mmol) of NaOH that was dissolved in (2 mL) ethanol was added, the pH of the solution became (8-9) then, (0.2 gm, 1 mmol) of copper acetate dihydrate (II) dissolved in (10 mL) ethanol was added gradually. The reaction mixture refluxed for 4h, then left overnight with stirring at r.t. The precipitate was filtered off, washed with distilled water, ethanol and diethyl ether. The same procedure has been taken to synthesis the complexes of ($\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$,

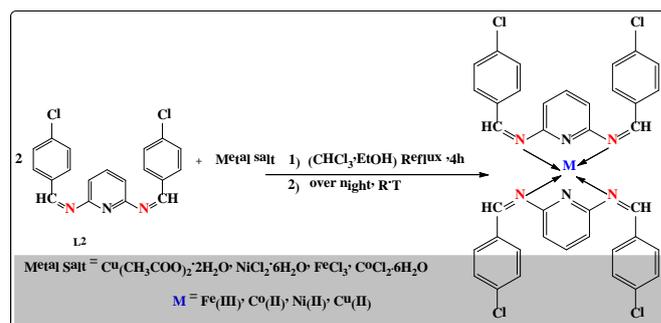
FeCl_3). The synthetic route of metal complexes in Scheme 3.



Scheme3. Synthetic route for synthesis of metal complexes of (L_1)

Synthesis of metal complexes (L_2):

(0.1 gm, 1 mmol) of (L_2) was dissolved in (20 mL) mixture of ethanol/chloroform (1:1) in a 100 mL round bottom flask with heating and stirring, then (0.1 gm, 0.5 mmol) of copper acetate dihydrate (II) dissolved in (10 mL) ethanol was added gradually. The reaction mixture refluxed for 4h, then left overnight with stirring at r.t. The precipitate was filtered off, washed with distilled water, ethanol and diethyl ether. The complexes of ($\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, FeCl_3) were also synthesized with the same steps. The synthetic route of metal complexes in Scheme 4.



Scheme4. Synthetic route for synthesis of metal complexes of (L_2)

RESULTS AND DISCUSSION:

Some of physical properties of Schiff bases ligands and their metal complexes are given in (Table 1).

Table 1: Physical properties of Schiff base ligands (L_1, L_2) and their metal complexes.

Compound	Color	M.P (°C)	Yield (%)	Molar Conductance (Ohm ⁻¹ cm ² mol ⁻¹)	μ_{eff} (B.M)
L_1	White	152-154	63.6	—	—
$[\text{Cu}(L_1)_2(\text{H}_2\text{O})_2]$	Brown	150(dec.)	63.5	10.5	1.8
$[\text{Co}(L_1)_2(\text{H}_2\text{O})_2]$	Bright Green	145(dec.)	40.7	13	4.97
$[\text{Ni}(L_1)_2(\text{H}_2\text{O})_2]$	Greenish Yellow	350	69.5	10.6	2.81
$[\text{Fe}(L_1)_2 \text{Cl}_2]\text{Cl}$	Reddish Brown	122	58.4	72.4	5.5
L_2	Light yellow	262(dec.)	87.9	—	—
$[\text{Cu}(L_2)_2(\text{H}_2\text{O})_2]$	Light Brown	245(dec.)	46.6	13	1.9
$[\text{Co}(L_2)_2(\text{H}_2\text{O})_2]$	Light Green	178(dec.)	81.9	14.5	4.93
$[\text{Ni}(L_2)_2(\text{H}_2\text{O})_2]$	Dark Green	250(dec.)	64.2	21.5	2.97
$[\text{Fe}(L_2)_2 \text{Cl}_2]\text{Cl}$	Dark Olive Green	190(dec.)	49.7	69.5	6.1

Table 2: IR data (cm⁻¹) of Schiff base ligand (L₁) and its corresponding metal complexes.

Comp.	ν C-H _{arom.}	ν C-H _{aliph.}	ν C=N _{imine}	ν C=N _{Pyr.}	ν C-O _{phenolic}	ν C=C _{ring}	ν M-N	ν M-O
L ₁	3053	2833	1661	1567	1222	1531	1409	—
[Cu(L ₁) ₂ (H ₂ O) ₂]	3051	2873	1616	1566	1246	1527	1404	532
[Co(L ₁) ₂ (H ₂ O) ₂]	3051	2850	1639	1570	1238	1527	1404	536
[Ni(L ₁) ₂ (H ₂ O) ₂]	3020	2858	1639	1577	1238	1523	1404	520
[Fe(L ₁) ₂ Cl ₂]Cl	—	2908	1612	1585	1265	1562	1442	536

Pyr. : Pyrimidine ring**Table 3:** IR data (cm⁻¹) of Schiff base ligand (L₂) and its corresponding metal complexes.

Comp.	ν C-H _{arom.}	ν C-H _{aliph.}	ν C=N _{imine}	ν C=C _{ring}	ν M-N
L ₂	3053	2968	1583	1410	1476
[Cu(L ₂) ₂ (H ₂ O) ₂]	3051	2839	1697	1411	1593
[Co(L ₂) ₂ (H ₂ O) ₂]	3059	2870	1608	1450	1485
[Ni(L ₂) ₂ (H ₂ O) ₂]	—	2870	1608	1450	1485
[Fe(L ₂) ₂ Cl ₂]Cl	—	2843	1647	1404	1489

Table 4: ¹H-NMR data (δ, ppm) for synthesized Schiff bases ligands (L₁,L₂)

	Chemical shift	Assignment		Chemical shift	Assignment
L ₁	7.83-7.85(d)	H ₄ ,H ₆	L ₂	7.84-7.86 (d)	H ₂ ,H ₂ ¹ ,H ₆ ,H ₆ ¹
	7.76-7.80 (t)	H ₅		8.15-8.17 (d)	H ₃ ,H ₃ ¹ ,H ₅ ,H ₅ ¹
	6.97 (s)	H ₇		10.36 (s)	H ₇ ,H ₇ ¹
	7.38-7.40(d)	H ₁₀ ,H ₁₃		7.59, (s)	H ₉ ,H ₁₁
	7.28-7.31 (t)	H ₁₁ ,H ₁₂		7.20 (s)	H ₁₀
	5.77(s)	OH ₁₄			

FTIR Spectra:**FTIR of Schiff base ligand (L₁) and its metal complexes:**

FTIR spectra of (L₁) show band at 3242 cm⁻¹ (ν O-H), (1661cm⁻¹) (ν C=N) azomethine, while in their metal complexes this band were shifted to low wave number because of the effect of coordination were in the range (1639-1612cm⁻¹), 1222 cm⁻¹ (ν C-O)_{phenolic}, this band in the complexes appeared at (1265-1238cm⁻¹), 1531-1409 cm⁻¹ (ν C=C)_{ring},

the new bands in the range (536-532cm⁻¹), (470-432cm⁻¹) these are for the coordination of (ν M-N) and (ν M-O) respectively^[12,13]. (Table 2).

FTIR of Schiff base ligand (L₂) and its metal complexes:

FTIR spectra of (L₂) show bands in the range (1583cm⁻¹) (ν C=N)_{azomethine}, (1476-1410cm⁻¹) (ν C=C_{ring}), while in their metal complexes this bands appeared in the range (1697-1608cm⁻¹), (1489-1404 cm⁻¹) (ν C=N), (ν C=C_{ring}) respectively, the new bands in the range (482-420cm⁻¹) for the coordination of metal-ligand (ν M-N)^[12,13] (Table 3).

¹H-NMR Spectra of Schiff bases ligands :

The¹H-NMR spectra of the ligands (L₁,L₂) were recorded using DMSO-d₆ as a solvent.

¹H-NMR (δ, ppm) of (L₁):

The ¹H-NMR of the ligand (L₁) showed signal at 5.77(s,OH₁₄) attributed to proton for phenolic group, singlet signal 6.97 (s,H₇) attributed to proton for azomethine group, 7.28-7.31 (t,H₁₁,H₁₂),7.38-7.40 (d,H₁₀,H₁₃), 7.76-7.80 (t,H₅), 7.83-7.85 (d,H₄,H₆) attributed for aromatic protons^[14]. ¹H-NMR data summarized in (Table 4)

¹H-NMR (δ, ppm) of L₂:

The ¹H-NMR of the ligand (L₂) showed signal at 7.20

(s,H₁₀), 7.59, (s,H₉,H₁₁), 7.84-7.86 (d,H₂,H₂¹,H₆,H₆¹), 8.15-8.17, (d, H₃,H₃¹,H₅,H₅¹) attributed for aromatic protons, 10.36 (s, H₇,H₇¹) attributed to protons for azomethine group^[15,16]. ¹H-NMR data summarized in (Table 4).

Molar conductance measurements:

The conductivity measurements of the complexes were recorded for (10⁻³ M), the solution of the samples was in (DMF) at R.T. The molar conductance values of the complexes showed in (Table 4). We concluded from the results that the Fe(III), Ni(II), Co(II), Cu(II) complexes of the ligands (L₁, L₂) have a molar conductivity values in the range (10.5 - 72.4 Ω⁻¹ mol⁻¹ cm²). Which indicates that complexes are non-ionic therefore, they are considered as non-electrolytes, except the complexes [Fe(L₁)₂Cl₂]Cl and [Fe(L₂)₂Cl₂]Cl were found to have molar conductance value of (72.4 - 69.5 Ω⁻¹ mol⁻¹ cm²), that indicate the ionic nature and they are an electrolytes with molar ratio (1:1)^[17].

Magnetic susceptibility:

The values of effective magnetic moment of metal complexes were summarized in (Table 1), which were measured at (r.t). The complexes [Cu(L₁)], [Cu(L₂)] have (μ_{eff}) in the range (1.8-1.9 B.M) this value is within the range of octahedral geometry. The complexes [Co(L₁)], [Co(L₂)] have (μ_{eff}) in the range (4.97-4.93 B.M) which indicate the octahedral geometry. The complexes [Ni(L₁)], [Ni(L₂)] their (μ_{eff}) exhibited in the range (2.81-2.97 B.M) this value indicated for octahedral geometry. The complexes [Fe(L₁)], [Fe(L₂)] exhibited (μ_{eff}) value in the range (5.5-6.14B.M) which indicate for octahedral geometry^[18].

Electronic Spectra:

The electronic spectra of Schiff base ligand (L_1) show absorption bands at (42553cm^{-1} and 27027cm^{-1}) which are attributed to ($\pi \rightarrow \pi^*$) and ($n \rightarrow \pi^*$) transitions respectively, the complex of Cu(II) shows band at (42553cm^{-1}) due to ($\pi \rightarrow \pi^*$) and band at (23529cm^{-1}) due to charge transfer (C.T). The complex of Co(II) shows bands at (42553cm^{-1} , 22727cm^{-1}) due to the transitions ($\pi \rightarrow \pi^*$ and C.T) respectively. The complex of Ni(II) shows band at (42553cm^{-1}) which due to ($\pi \rightarrow \pi^*$) transition, the band at (23255cm^{-1}) is due to charge transfer (C.T), the band at (14814cm^{-1}) is due to the transition ${}^3A_{2g} \rightarrow {}^3T_{2g}$. The complex of Fe(III) shows bands at (42553cm^{-1} , 25316cm^{-1}) due to the transitions ($\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$) respectively, (14925cm^{-1} , 13071cm^{-1} , 12738cm^{-1}) due to the transitions (C.T, ${}^6A_{1g} \rightarrow T_{2g}$ (G), ${}^6A_{1g} \rightarrow {}^5T_{1g}$) respectively. The Schiff base ligand (L_2) shows absorption bands at (42553cm^{-1} , 23809cm^{-1}) which are attributed to ($\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$) transitions respectively, the complex of Cu(II) shows band at (42553cm^{-1}) assignable to the transitions ($\pi \rightarrow \pi^*$), (21276cm^{-1}) due to charge transfer (C.T). The complex of Co(II) shows band at (42553cm^{-1}) assignable to the transition ($\pi \rightarrow \pi^*$), the band at (20833cm^{-1}) is due to charge transfer (C.T) and the band at (14925cm^{-1}) is due to (${}^4T_{1g}^{(F)} \rightarrow {}^4T_{2g}^{(F)}$) transition. The complex of Ni(II) shows band at (42553cm^{-1}) assignable to the transition ($\pi \rightarrow \pi^*$), the band at (21505cm^{-1}) is due to charge transfer (C.T) and band at (15625cm^{-1}) is due to (${}^4T_{1g}^{(F)} \rightarrow {}^4A_{2g}^{(F)}$) transition. The complex of Fe(III) shows band at (42553cm^{-1}) assignable to the transition ($\pi \rightarrow \pi^*$), the band at (21505cm^{-1}) is due to charge transfer (C.T) and the band at (15503cm^{-1}) which is due to (${}^6A_{1g} \rightarrow T_{2g}$ (G)) transition.^[19], all complexes are octahedral geometry and shown in (figure 1).

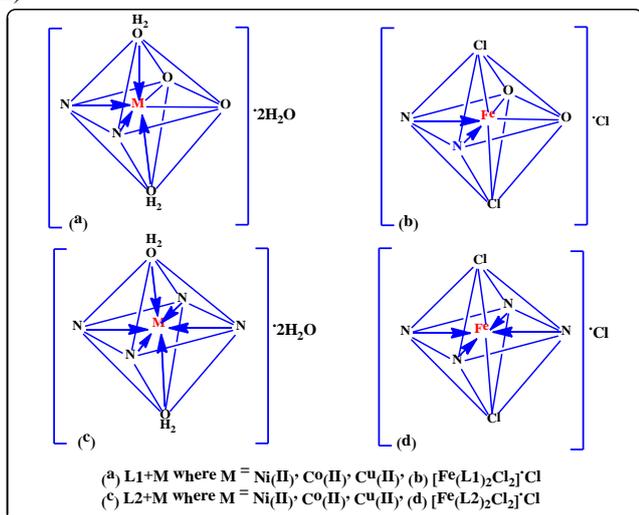


Figure 1: The octahedral geometry of metal complexes

Anti-cancer activity:

The in vitro Anti-cancer activity of metal complexes on ($L_{20}B$) cell line was tested, the inhibitory effect of the complexes on $L_{20}B$ cell line was tested using the (MTT) assay^[20] for (24h) using the concentration of metal complexes $4000 \mu\text{g} / \text{mL}$ and $2000 \mu\text{g} / \text{mL}$, the complexes

were dissolved in DMSO. The protocol described in the literature^[21, 22] was used. The results showed that the concentration $4000 \mu\text{g} / \text{mL}$ had a greater inhibitory effect than the concentration $2000 \mu\text{g} / \text{mL}$, the value of inhibition ratio detailed in (Table 5) and (Figure 2).

Table 5: The Inhibition ratio of metal complexes against ($L_{20}B$) cell line

No.	Compound	%IR of the Conc.2000 $\mu\text{g} / \text{mL}$	%IR of the Conc.4000 $\mu\text{g} / \text{mL}$
1	$[\text{Cu}(L_1)_2(\text{H}_2\text{O})_2]$	10.1	15.6
2	$[\text{Co}(L_1)_2(\text{H}_2\text{O})_2]$	9.1	11.4
3	$[\text{Ni}(L_1)_2(\text{H}_2\text{O})_2]$	9.1	15.3
4	$[\text{Fe}(L_1)_2\text{Cl}_2]\text{Cl}$	7.3	8.7
5	$[\text{Cu}(L_2)_2(\text{H}_2\text{O})_2]$	13.2	16
6	$[\text{Co}(L_2)_2(\text{H}_2\text{O})_2]$	18.4	20.9
7	$[\text{Ni}(L_2)_2(\text{H}_2\text{O})_2]$	8.7	12.5
8	$[\text{Fe}(L_2)_2\text{Cl}_2]\text{Cl}$	19.1	22.9

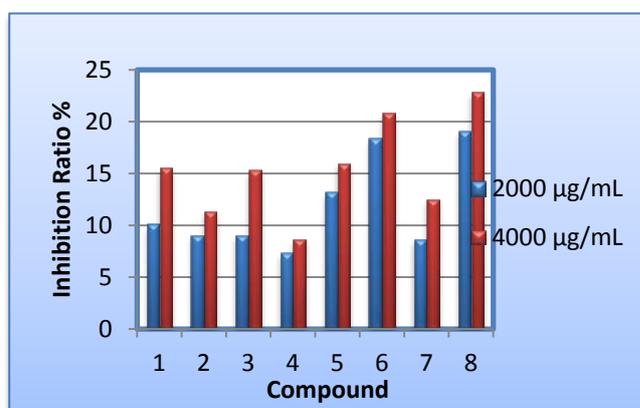


Figure 1: The Inhibition ratio of metal complexes against ($L_{20}B$) cell line

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