Analytical and Thermodynamic Study of Complex of Co(II) Ion with New Azo Reagent2-((3-methoxy phenyl) diazenyl)-4,5-diphenyl imidazole (MBDPI)

Zainab Jassim Khdhair* **Sami WheedRadhi****
*Department of Chemistry, College of science, University of Kufa, Najaf –Iraq.
** Al- Kufa University

الخلاصــة

تضمن البحث تحضير كاشف عضوي جديد 2-((3-methoxy phenyl) diazenyl)-4,5-diphenyl imidazole (MBDPI)كما تم تحضير معقد هذا الكاشف مع أيون الكوبلت((IIوقد جرى تحضير المعقد بعد تثبيت الظروف الفضلى من تركيز ودالة حامضية وذلك من خلال دراسة أطياف الأشعة فوق البنفسجية- المرئية لمحلول مفاعلة أيون الكوبلت((II مع محلول الكاشف ولمدى واسع من الدوال الحامضية pH=5-11)) وضمن التراكيز المطاوعة لقانون بير- لامبرت.شخص الكاشف ومعقد الكوبلت المحضر بالوسائل التحليلية والطيفية المتاحة مثل التحليل الدقيق للعناصر, أطياف الأشعة تحت الحمراء FTIR, التوصيلية المولارية الكهربائية ودراسة السلوك الحراري. لقدبينتنتائجالدراسةانالكاشفيسلككاشف ثنائيةالمخلب(N,N´) يرتبط مع الأيون الفلزي بنسبة مولية (2:1) (فلز: كاشف). وقد بينت دراسة التوصيلية المولارية انعدام الصفة الأيونية للمعقد المدروس. وبالاعتماد على النتائج تم الاستنتاج بان المعقد الكليتي لأيون الكوبلت الثنائي يتخذ الشكل الهندسي الثماني السطوح. كما درست الفعالية البكتيرية للكاشف ومعقده المدروس تجاه نوعين من البكتريا أحدهما موجبة لصبغة كرام(Gram positive)

pseudomonas aeruginosa -1

والاخرى سالبة لصبغة كرام(Gram negative)

Staphylococcus aureus -2

وقد أظهرت الدراسة أن المركبين المدروسين قد أظهرا فعالية تثبيطية عالية لقتل البكتريا من النوع السالب لصبغة كرام دون النوع الموجب.

(II)

Abstract

The new azo reagent 2-((3-methoxy phenyl) diazenyl)-4,5-diphenyl imidazole was designated as (MBDPI) and its metal chelate with Co^{2+}metal ion were prepared. The preparation of the complex have been conducted after fixing the optimum conditions such as pH and concentration. UV-Vis spectrum for reacting of metal solution and the reagent solution. For wide range of pH (5-11) and concentration

which obeys Beer-Lambert Law. The (MBDPI) and its metal chelate were characterized by the analytical and spectroscopic studies of the complex solution such as elemental analysis, FTIR, molar conductivity measurements and thermodynamic study. It has been found that the azo reagent behaves as neutral bidentate (N,N´) reagent forming chelate with 1: 2 (metal : reagent) stoichiometry. The infrared spectra of the chelating complex have been studied and compared with the free reagent spectrum. The Conductivity measurements for solutions in ethanol have shown non-ionic character for the chelate complex.Depending on these results we can conclude that the proposed geometrical structure of the chelate complex is octahedral. The synthesized compounds have been tested in vitro against a two kinds of pathogenic bacteria to assess their antibacterial properties using disk diffusion method, which is aGram negative bacteria:
(*pseudomonas aeruginosa*) and agram positive bacteria (*Staphylococcus aureus*)
The results showed that the reagent, its metal chelate with $Co^{2+}$and the aqueous solution of metal ion had inhibition activity toward the first type of bacteria without the second type.

***Keywords*:**new azo reagent, metal complex, spectroscopic studies, thermodynamic study,disc diffusionandAntibacterial activity.

Introduction

Synthetic azo compounds are widely usedNowadays in different application fields, such as textile fibres dyeing, colouring of different materials in biological–medical studies, advanced organic synthesis, medicines, cosmetics, food, paints, plastics, shipbuilding, automobile industry,cable manufacture and in analytical chemistry[1-6].

Out of different classes of dyes, azo dyes constitute the largest group of dyes used in Industry[7-10]. More than two thousandsazo dyes are known and over half of the commercial dyestuffs are azo dyes.These azo compounds are suitable for the analysis of trace heavy metals[11].

Aromatic compounds, with two phenyl rings separated by an azo (-N=N-) bond are versatilemolecules and have received much attention in research areas both fundamental and application.in general, it possesses groups such as, $–SO_3H$, to increase dyes solubility, and auxochroms groups (like –COOH, –OH, $–NH_2$, –NHR, $–NR_2$) that intensify the colour and increase dyes ability to bind to fibres[12].
Biological importance of azo compounds is well known for their use as antineoplastics, antidiabetics, antiseptics, anti-inflammatory, and other usefulchemotherapeutic agents[13-16]. Azo compounds are known to be involved in a number of biological reactions such as inhibition of DNA, RNA and protein synthesis, carcinogenesis and nitrogen fixation[17].
In this study, the synthesis and characterization of new (imidazole- azo) reagent derived from3-methoxyanilineand 4,5-diphenyl imidazole as coupling component and its Co(II) complex is described.

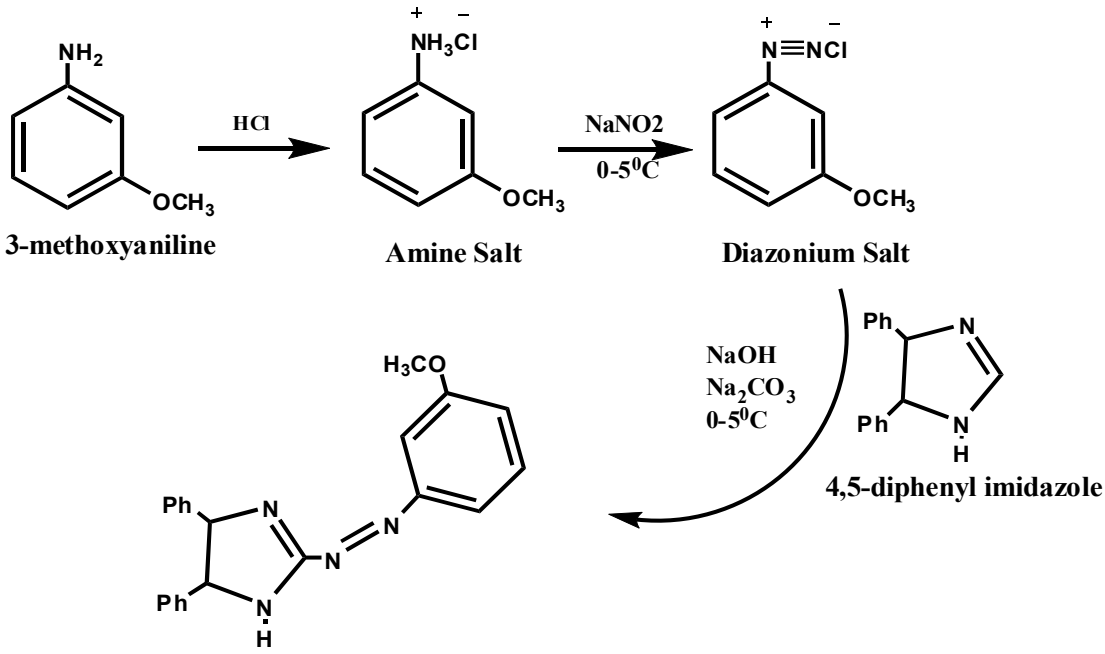
Experimental

Materials and measurements

All chemicals used in the present investigation were of highest purity either from BDH or Fluka companies and used without further purification.
Elemental analyses were carried out by means of Micro analytical unit of (EuRo VECTOR, Italy) C.H.N Elemental analyzer.Absorption spectra were recorded by ShimadzuUV-1650 spectrophotometer, for solution of the complex in aqueous ethanol at room temperature. Using 1cm quartz cell.IR spectra were recorded
using KBr discs at 4000-400 $cm^{-1}$by FT-IR Test scan Shimadzu model 8400. Molarelectrical conductivity measured by digital conductivity meter 720 (WTW) with the prepared complex concentration of 10^{-3}M in ethanol at room temperature. The metal content of the complexes was measured using atomic absorption technique by Shimadzu (AA-2600). pH measurements were carried out using (pH– meter), 720, WTW 82362. The thermal behavior of complex was studied, Water Bath GFL 1083 was employed for this purpose.Electro thermal melting point model 9300 was used to measure the melting points of the reagent and it's complex.

Synthesis of the reagent

(MBDPI) reagent was prepared according to the following general procedure[18](Scheme 1). 3-methoxyaniline(123g) was dissolved in (10ml) of water and (2ml) of concentrated hydrochloric acid. The filtered solution was diazotized below 5°Cwith (69 g) of aqueous (1.0 M) sodium nitrite. The resulting diazonium chloride solution was mixed with 4,5-diphenyl imidazole (220g) dissolved in (150ml) alkaline ethanol cooled below 0°C. After leaving in the refrigerator for 24 hr., the mixture was acidified with dilute hydrochloric acid until pH = 7. The precipitate was filtered off, and recrystallized twice from hot ethanol, and dried in a vacuum desiccator.



Scheme(1) : Preparation of the (MBDPI) reagent

Synthesis of complex

The chelate complex has been synthesized at optimal pH valueby dissolving(354g) of reagent (MBDPI)in (50ml) ethanol and then (237.93g) of Co(II) chloride dissolved in (50ml) distilled water is added drop wise with vigorous stirring to the reagent solution.The reaction mixture was left over nightat room temperature, then the solid complex is filtered off washed with ethanol and dried in air.Table.1

collects the physical properties and analytical data for this complex.

Table 1: Some physical properties and analytical data of the reagent (MBDPI)and Co(II) complex

No.	Compound	Color	m.p °C	Elemental analysis (%) Calc./found			
				C	H	N	Co
1	$C_{13}H_{19}N_4CO$	Red dis -h Ora nge	90 – 92	75.07 6) 74.57 (6	5.19 1) 5.08 (4	14.98 2) 15.81 (9	—
2	$Co(C_{13}H_{19}N][_4CO)_2Cl_2$	vio let	240 – 242	65.03 2) 63.01 (2	4.36 1) 4.29 (6	12.41 4) 13.36 (6	7.094) 7.03 (3

Results and discussion

Calibration curve

Under the experimental conditions described, standard calibration curve for Co(II) complex was constructed by plotting absorbance versus concentration in the linearity range of ($0.1x10^{-5}$– $9x10^{-5}$M) (Fig.1).

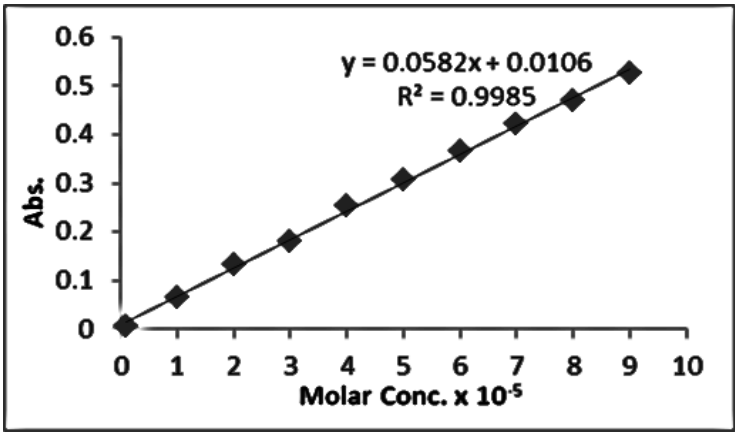


Figure 1: calibration curve of metal complexCo(II) with the reagent(MBDPI) at 455nm and 25°C

Effect of pH

Suitable pH values for metal complex solution Was found to be in the range of
(5 –11). To evaluate the optimal pH values of metal complex solution.The concentrations of metal and the reagent was equal to ($5x10^{-5}$ M)and ($1x10^{-4}$ M) respectively The effect of pH on the absorbance was studied, and the results are shown in Table 2 and Fig.2.

Table 2:Absorbance of Co(II) complex with the reagent(MBDPI) at pH=(5-11)

pH	Abs. of Co(II) complex
5	0.310
6	0.307
7	0.319
8	0.330
9	0.357
10	0.348
11	0.291

Figure 2: The effect of pH on the absorbance of metal complex Co(II) with the reagent(MBDPI)

Effect of time

The optimum reaction time was determined by following the color development at room temperature, the color obtained remained for 180 min (Fig.3).

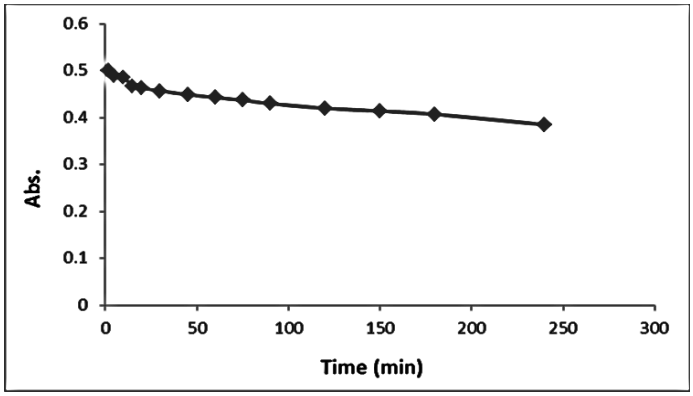


Figure 3: Effect of time on Stability of Co(II)complex with (MBDPI)at λ_{max}=455nm and pH=9

The optimum Concentration of the metal ion

We canget on this by reacting differentconcentrations of metal ion solution in the range of ($1x10^{-5}$– $1x10^{-4}$ M) with the same volume of the reagent solution after fixing the optimum pH for Co(II) ion, the results are shown in Table 3.

Table 3: The absorbanceversusdifferent conc. for Co(II)complex with (MBDPI)

Molar Conc.×10^{-5}	Abs. of Co(II) complex
1	0.362
2	0.352
3	0.354
4	0.403
5	0.474
6	0.478
7	0.473
8	0.467
9	0.435
10	0.422

The optimumconcentration of the reagent (MBDPI)

The optimum concentration of (MBDPI) was getby reacting differentconcentrations of the (MBDPI) solution in the range of ($0.5x10^{-4}$ –$1.3x10^{-4}$ M) with the same volume of the metal ionsolution after fixing the optimum pH for Co(II) ion, the Table 4 and Fig.4are showed the results.

Table4: The absorbance for the Co(II)complex versus different concentrationfor the reagent (MBDPI)

Molar Conc.×10^{-4}	Abs. of Co(II) complex
0.5	0.225
0.6	0.281
0.7	0.347
0.8	0.372
0.9	0.431
1.0	0.463
1.1	0.479
1.2	0.492
1.3	0.515

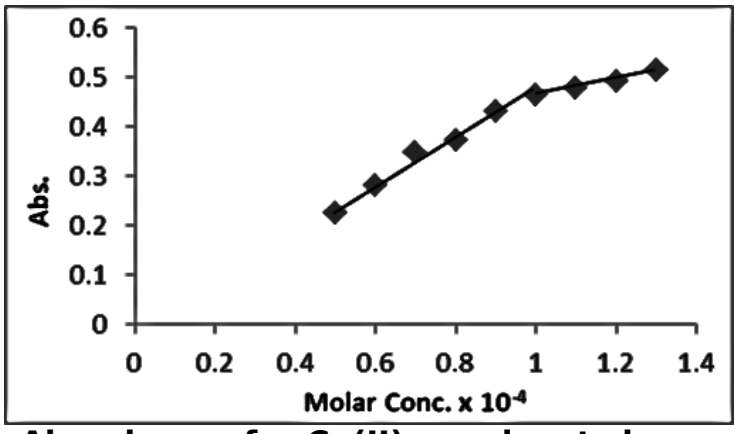


Figure 4: Absorbance for Co(II)complexat change of Conc. of (MBDPI (and constant Conc. of metal ion) 5x10^{-5}M)

Metal: Reagent ratio

The (metal : reagent) ratio(M:R) of complex was determined by moleratio method at fixed concentration and pH at wavelength of maximum absorption. The reagent was found to form (1: 2) chelate with this metal ion. The results are in agreement with the values reported for some azo imidazole complexes[19,20], the Table 5 and Fig.5are showed the results.

Table 5: The relationship between absorbanceand molar ratio for Co(II)complex with (MBDPI)

]:[M] [L	$x\frac{L}{M}$	Abs. of Co(II) complex λ_{max}=455 nm
1:1.0	1.0	0.225
1:1.2	1.2	0.281
1:1.4	1.4	0.347
1:1.6	1.6	0.372
1:1.8	1.8	0.431
1:2.0	2.0	0.463
1:2.2	2.2	0.479
1:2.4	2.4	0.492
1:2.6	2.6	0.529

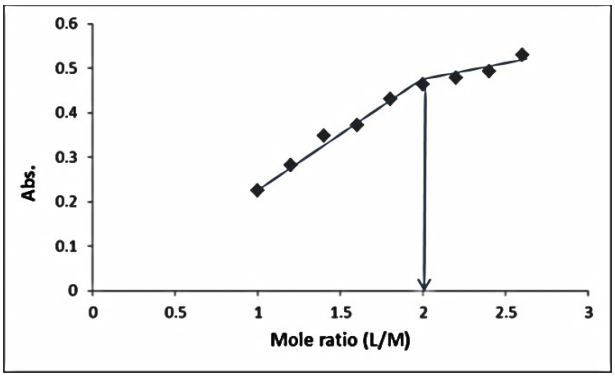


Figure 5:The mole ratio(M:R) of Co(II) with(MBDPI) at Optimal Molar Conc.= 5 x10^{-5} M and pH=9

Continuous variation method

This method include reacting of different volumes for each component (the reagent and metal ion) in constant total volume and molar concentration of each the two component, The results are given in Table6 and Fig.6.

Table 6: Values of absorbance versus to V_M/ V_L for Co(II)complex with (MBDPI)

$V_M/(V_M+V_L)$	$V_L/(V_M+V_L)$	Abs. of Co(II) complex λ_{max}=455 nm
0	1	0.005
0.1	0.8	0.058
0.2	0.6	0.138
0.4	0.5	0.241
0.5	0.4	0.233
0.6	0.2	0.183
0.8	0.1	0.078
1	0	0.010

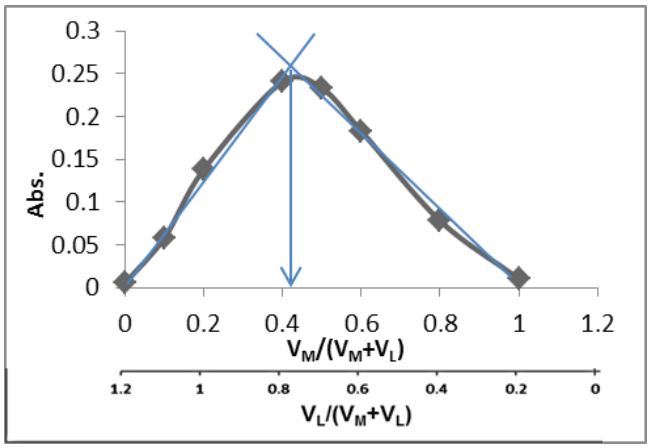


Figure 6: The Continuous variation andthe relationship between
V_M/ V_L for Co(II)complex with (MBDPI) in Optimum Conditions.

Absorption spectra

The absorption spectra of reagent (MBDPI) and its complex were studied and shown in figures (7,8). The (λ_{max}) of the reagent was found at (414nm). The spectra of Co(II) complex was recorded within wavelength (455nm). The absorption maxima (λ_{max}) of the complex shown in Table 3. Three absorption bands[21] were appear at the free reagent (MBDPI) spectrum. The band at (207nm) referring to the ($\pi\rightarrow\pi^*$) transitions of imidazole ring while the band at (289nm)referring to the ($\pi\rightarrow\pi^*$) transitions of benzene

ring[22]and the band at (414nm) is due to the charge transfer characters[23].

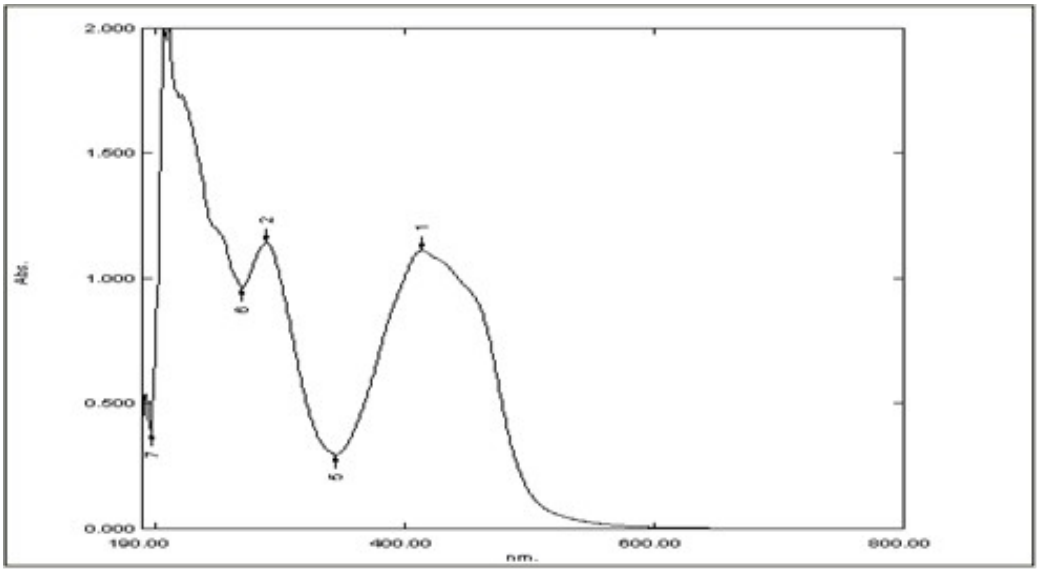


Figure 7: Absorbance spectrum of reagent (MBDPI)

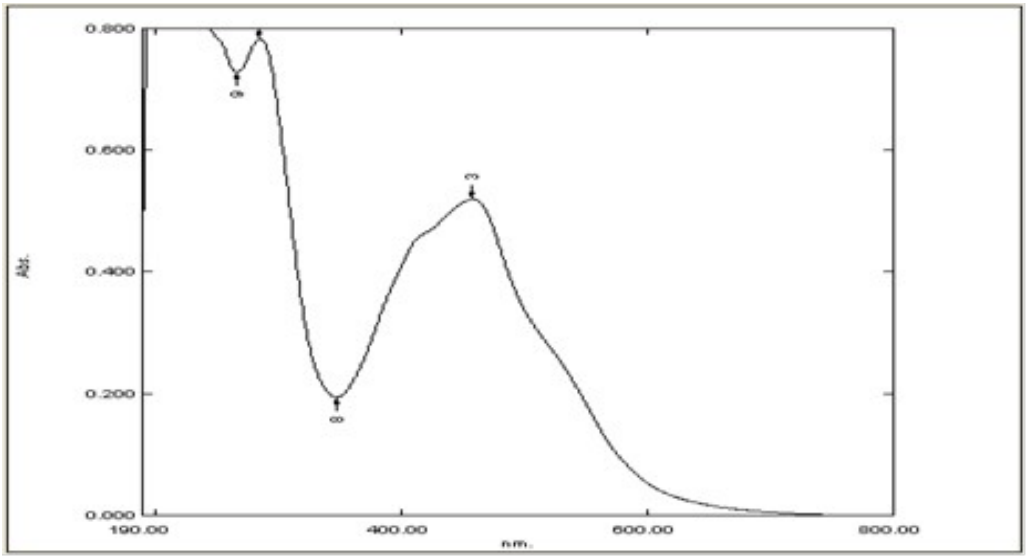


Figure 8: Absorbance spectrum ofreagent (MBDPI)with Co(II)in Molar Conc. 5 x10^{-5}M

Infrared spectra

Selected IR absorption of reagent and its complex are given in Table7. The comparison between the IR spectral data of the free reagent with that of it's complex are discussed as follow:-

1- The medium and broad band at 3404cm^{-1} in the spectrum of the reagentmay be attributed to the ν(N–H) of imidazole ring[24]. This band remains in the same region in free reagent

and in Co (II) complex,that indicates that this band didn'tshare in complexation[25].

2- The spectrum of the reagent shows absorption band at 1643cm^{-1} due to ν(C=N) of the imidazole nitrogen. It is observed with a little change in shape and shifted to lower frequency 1600cm^{-1} in Co(II) complex spectra. These differences suggest the linkage of metal ion with nitrogen imidazole ring[24].

3- The azo group (N=N) appears at 1442cm^{-1} in the free reagent spectrum. This band has been shifted to a higherfrequency 1459 cm^{-1} in complex spectrum;this band shifted and reducedintensity due to complex formation[26].

4- The spectrum of free reagent show absorption band at 1027cm^{-1} due to vibration of ν(OCH_3) group[27].This band is stable in position and intensity in both reagent and it'schelate complex.

5- New weak band in the region 465cm^{-1}was observed in the spectrum of chelate complex. This band was not present in the spectrum of reagent, and it due to ν(M–N)[28,29].

Thus the above FTIR spectrum data lead to suggest that the reagent behavesas a bidentate chelating agent. Figs. 9,10 shows the spectra of reagent(MBDPI) and it's complex.

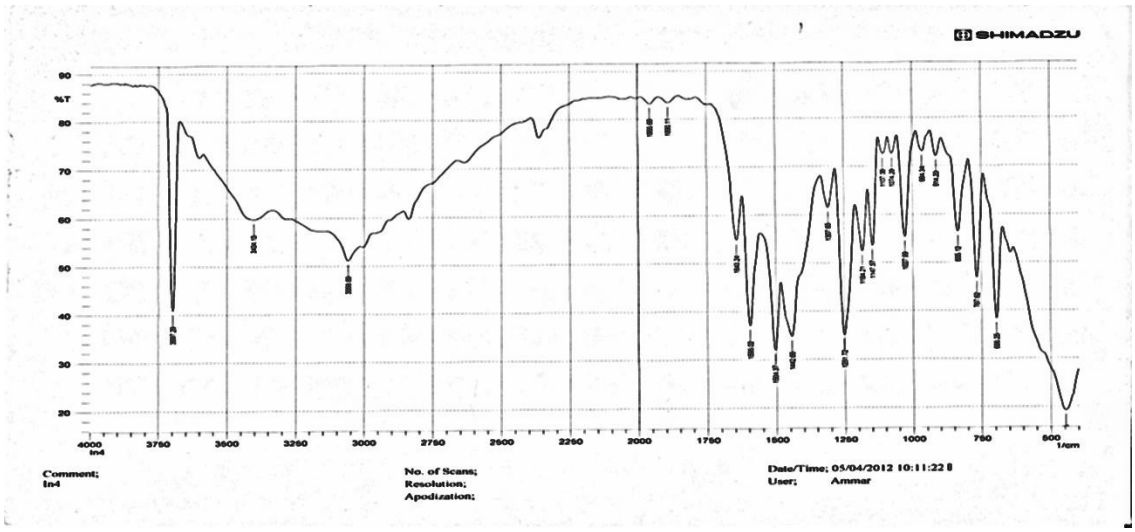


Figure 9: FTIR spectrum of (MBDPI)

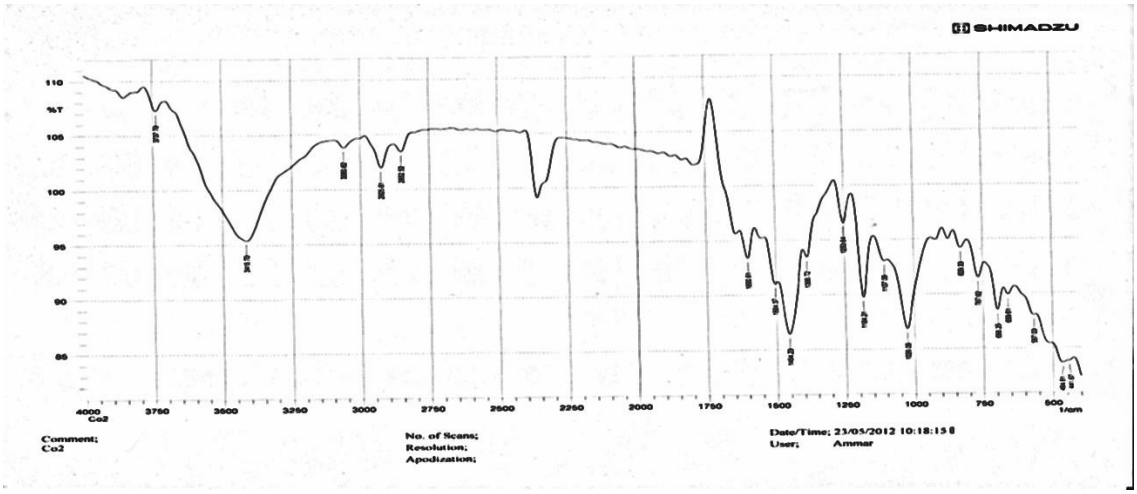


Figure 10: FTIR spectrum of chelate complex of Co(II) with (MBDPI)

Table 7: Characteristic FTIR absorption bands of the reagent (MBDPI) and Co(II) complex in cm^{-1} unit.

Compound	υ(N—H)	υ(C=N)	υ(N=N)	ν(OCH_3)	υ(M—N)
$C_{13}H_{19}N_4CO$	3404w	1643m	1442m	1027w	—
$Co(C_{13}H_{19}N_4C][O)_2Cl_2$	3415s	1600w	1459s	1027w	465w

s =strongw=weak m=medium

Conductivity measurements

This complex show the conductivity measurement value 12.63($S.mol^{-1}.cm^2$) in ethanol solution, this value indicating nonionic structure of this complex[30,31].

Calculation of the metal complex stability constant

Stability constants are obtained spectrophotometrically by measuring the absorbance of solutions of reagent and metal mixture at fixed wavelength (λ_{max}) and pH values. The

degree of formation of the complex is obtained according to the relationship[32], $\beta = (1 - \alpha) / (4\alpha^3 c^2)$, and $\alpha = (A_m - A_s)/A_m$, where A_s and A_m are the absorbance's of the partially and fully formed complex respectively at optimum concentration. The calculated β and Logβ values for the prepared complex are recorded in Table 8, The higher values of the stability constants of the Co(II) complex is expected on the basis of the charge / ionic radius and ionization potential of the metal ion mostly in the octahedral field environment.

Table 8: Stability constants values of the reagent (MBDPI) with Co(II) complex atdifferenttemperatures

T.(K)	A_s	A_m	A	β	Ln β
283	0.495	0.552	0.103	$10^{1} \times 8.229^{0}$	25.133
288	0.481	0.544	0.115	$10^{1} \times 5.855^{0}$	24.787
293	0.431	0.506	0.148	$10^{1} \times 2.630^{0}$	23.993
298	0.398	0.479	0.169	$10^{1} \times 1.720^{0}$	23.568
303	0.376	0.467	0.194	$10^{1} \times 1.104^{0}$	23.125

Thermal studies

Thermal behavior of the azo ligand and its metal complexes were studied by thermo gravimetric techniques in the temperature range of (10-30°C).The thermodynamic activation parameters of decomposition of complex, namely enthalpy (ΔH), entropy (ΔS) and Gibbs free energy (ΔG).The Thermodynamicdata were listed in Table9. The values of Lnβagainst 1/T (shown in Figure 11) gave a slope and ΔH was determined from the intercept. The entropy of activation (ΔS), enthalpy of activation (ΔH) and the free energy change of activation (ΔG) were calculated.

Table 9: Stability constants values and Thermodynamic values of the reagent (MBDPI) with Co(II)

Tem.)K° (	β	β Ln	GΔ J.mol^{-1}	HΔ J.mol^{-1}	SΔ J.mol^{-1}K^{-1}
283	$10^{1} \times 8.2290$	25.133	−59134.48065	-13.267	208.9088821
288	$10^{1} \times 5.8550$	24.787	−59350.78598		206.0330520
293	$10^{1} \times 2.6300$	23.993	58446.99−599		199.4325221
298	$10^{1} \times 1.7200$	23.568	58391.41−690		195.8998319
303	$10^{1} \times 1.1040$	23.125	58255.15−875		192.2174645

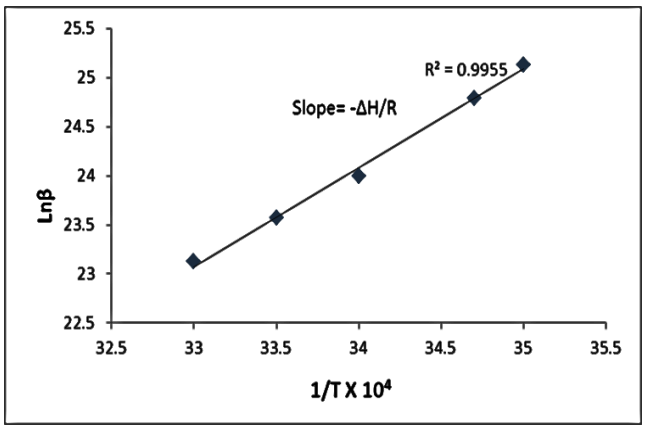


Figure 11: The relationship between valuesof (1 / T) and (Ln(β for reacting thereagent(MBDPI) solution withCo(II)ionsolution

According to these results the structural formula of prepared complex may be proposed in Fig.12.

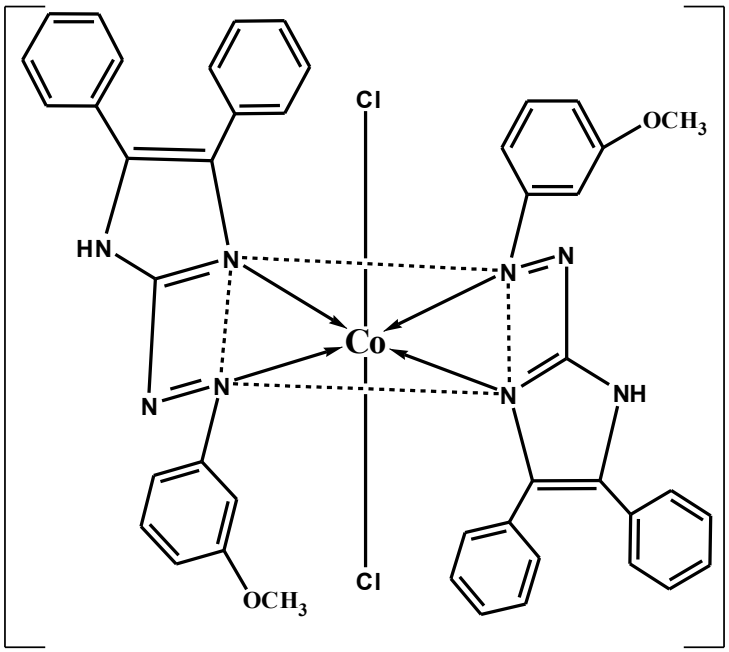


Fig.12: The proposed structural formula of Co(II) with the reagent (MBDPI)

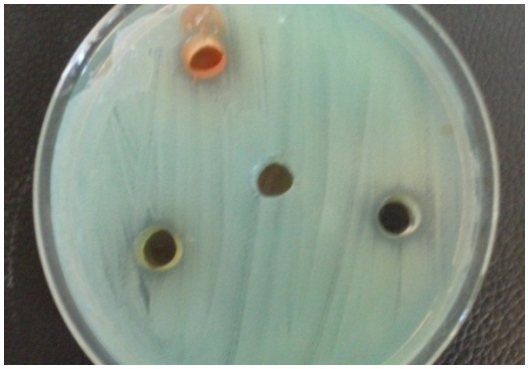
Antibacterial activity

The antibacterialactivity of MBDPI was evaluated in vitro using disc diffusion method. The antimicrobial activity data is given in Table 10. MBDPI and it's complex showed remarkable activity against the first species of bacteria(*pseudomonas aeruginosa*) but were inactive against the second species(*Staphylococcus aureus*).Results were compared with standard drugs. The variation in the effectiveness against two species of bacteria depends either on the impermeability of the cells of the microbes or on differences in ribosome of microbial cells.

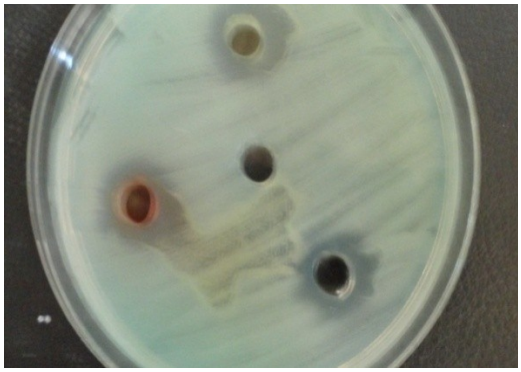
Table 10:Antibacterialscreening data (zone of inhibition in mm) of (MBDPI) and it's complex

Compound	*Pseu.aurginosa*			*Staphy.aureus*		
	10^{-3} M	10^{-4}M	10^{-5} M	10^{-3}M	10^{-4} M	10^{-5}M
$C_{13}H_{19}N_4CO$	15	8	8	–	–	–
$Co(C_{13}H_{19}N_4CO)]$ $[_2Cl_2$	24	18	15	–	–	–

$C_{13}H_{19}N_4CO$ = MBDPI
(_) = no inhibition



Picture 1: effect of the reagent (MBDPI) on growth the (*pseudomonas aeruginosa*)



Picture 2: effect of the reagent (MBDPI) with Co(II) complex on growth the (*pseudomonas aeruginosa*

References

1- A. Mittal, L. Kurup and J. Mittal **;Journal of Hazardous Materials**, 146, 243-248, (2007).
2- A. E. I. Ahmed, J. N. Hay, M. E. Bushell, J. N. Wardell and G. Cavalli**;Reactive & Functional Polymers**, 68, 248, (2008).
3- C. Combellas, F. Kanoufi, H. Pinson, F. Podyorica**; Am. Chem. Soc.**, 34, 429, (2008).
4- M. Wainwright **;Dyes and Pigments**, 76, 582-589, (2008).
5-P. S.Murray, S. F. Ralph**;ElectrochimicaActa**, 51, 2471, (2006).
6-H. A.Dabbagh, A.Teimouri, A. N.Chermahini**;Dyes Pigments**, 73, 239, (2007).
7-K.Hunger **;"Industrial Dyes Chemistry, Properties, Applications"**, 1st Ed., Wiley-VchVerlagGmbh& Co. Kgaa, Weinheim, Germany, (2003).
8-P. A.Ramalho, H.C.Scholze, M. H.Ramalho, M. T. Oliverira and A. M. Campos**;Enzyme and Microbial Technology**, 7, 402–412, (2002).
9-A.Stolz**;Applied MicrobiolBiotechnol.**, 56, 69-80, (2001).
10-S.Nam, V.Renganathan and P. G. Tratnyek**;Chemosphere**, 45, 59, (2001).

11-A. A. M. Ali,H. J. Mohammed and K. J. AL- adely**;J. oF AL-Qadisia for pure science**, 13, 439, (2008).
12-H. S.Rai, M. S. Bhattacharyya, J. Singh, T. K. Bansal, P. Vats and U. C. Banerjee **;Crit. Rev. Env. Sci. Tec.**, 35(3): 219-238, (2005).
13-F. T.Sanjay, M. P.Dinesh, P. P.Manish and G. P.Ranjan**;Saudi Pharm. Journal**, 15(1): 48-54, (2007).
14-J.-S.Bae, H. S.Freeman, A.El-Shafei**;Dyes and Pigments**, 57, 121, (2003).
15-R.G. Child, R.G. Wilkinson and A.Tomcu-Fucik**;Chem. Abstr**, 87, 6031, (1977).
16-H.G. Gargand C. Praksh**; J. Med. Chem.**, 15(4): 435-436, (1972).
17-R. N. Goyal, M. S. Verma and N. K. Singhal**;Croaticachemicaacta**,71(3): 715-726, (1998).
18-S. Shibata, M. Furukawa and K. Toei **;Ana. Chem. Acta**, (1973).
19- A. M. Ali, H. J. Mohammed and A. J. Khadhim**; The Islamic Unvi. J.**,16(1): 85-94, (2008).
20- H. M. Farhan **; M. Sc. Thesis**, Unvi. of Kufa, (2003).
21-H. Sigel**;"Metal ions in biological system"**, Marcel Dekker, New York, (1981).
22-M. R. Mohamoud, A. M. Hammon and S. A. Ibrahim **; Z .Phys. Chem.**, 265, 203, (1984)**.**
23-D. H. Williams and I. Fleming ;"Spectroscopic Methods in Organic **Chemistry"**, 2th ed, Mcgrew-HillBook, (1973).
24-S. Dutta**;Acta Pharm.**,60**,** 229–235**,** (2010).
25-O. Yamauchi, H. Tanaka and T. Uno **; Talanta.**,15, 177**,** (1968).
26-K. R. Anitha, V. Reddy and K. S.V.Rao**; J. Chem. Pharm. Res.**, 3(3): 511-519, (2011).
27-S. G. R. Karnawat, I. K. Sharma and P. S. Verma**;Int. J. of Appl. Bio. and Pharm. Tech.**, 2(2): 332-338, (2011).
28-Q. Fengxian, C. Yonglin, X. Hongliang, J. Yom and Z. Yumine**;Dyes and pigments**, 75, 454-459, (2007).
29-M. S. Masoud, A. E. Ali, M. A. Shaker and M. Abdul Ghani**; SpectroChimicaActa**, part A, 60, 3155-3159, (2004).
30-F. J. Ali **;M. Sc. Thesis**, Unvi. of Kufa, (2009).
31-S. T. Abid AL-Abas**; M. Sc. Thesis**, Unvi. ofKufa, (2010).
32-D. A. Skoog, D. M. West, F. J. Holler and S. R. Crouch **; "Fundamentals of Analytical Chemistry"**, 8th Ed., BROOKS/COLE, 805-807, (2004).