



Synthesis, Spectroscopic characterization and bactericidal valuation of some metal (II) complexes with new Tridentate Heterocyclic Azo Ligand Type (NNO) Donor



Ghusoon Faidhi Hameed¹, Fawzi Yahya Wadday², Murtadha Abd Ali Farhan¹, Sahar Aqeel Hussain³

¹Department of Chemistry, Faculty of Education, Al-Qadisiyah University, Al-Qadisiyah-Iraq

²Department of Chemistry, Faculty of Science, Kufa University, Al-Najaf-Iraq

⁴Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Kufa University, Al-Najaf-Iraq

Abstract

In this article, a synthesized ligand [(1-phenyl-2-pyrazolin-5-one) azo (6-bromo-2-hydroxynaphthalen-1-yl)] has employed for preparing three new complexes of Co(II), Ni(II) and Cu(II). The new azo ligand (PABH) has analyzed by Micro Elemental Analysis (CHNO), UV-visible, Fourier Transform infrared (FTIR) approaches, ¹H, ¹³C-NMR spectroscopy and mass spectroscopy. The consequence has specified that the ligand was represented as N,N,O-tridentate. The preparing of complexes has accomplished after fixing the finest concentration and pH values. UV-Vis spectra of these complexes solutions have been examined for a range of pH (5-9) and concentration (1×10⁻⁴ - 5×10⁻⁴) Molar that comply with Lambert-Beers law. A stoichiometry of the complexes has comprehended in relation to mole ratio which has investigated from a spectroscopic technique. The ratio of metal: ligand was achieved with (1:2) for all complexes. The metal ions complexes were characterized by FTIR, UV-Visible, molar conductance, atomic absorption, magnetic susceptibility and elemental analysis CHNO techniques. From the results of physico-chemical and spectral techniques, octahedral geometry has been proposed for these metal complexes. All these compounds were evaluated against two kinds of human pathogenic bacteria such as *Staphylococcus aureus* (Gram Positive) and *Escherichia coli* (Gram negative).

Keywords: Spectral studies, Complexes, Azo, Antimicrobial activity, Mass spectroscopy

Introduction

Azo compounds have been a highly significant sort of chemical compounds getting a consideration in analytical chemistry field. They can be extremely colored and employed as dyes and pigments extensively [1]. Azo dyes form an imperative type of organic colorants, involving as a minimum a conjugated azo (N=N) chromophore, and are the major and greatest multipurpose category of dyes. It was taken substantial consideration as a result of their remarkable and they have an advantageous in chemical and physical features [2,3]. Azo compounds stand for one of the foremost variations ligand that involving one or more azo moiety with unique pair of electron on the nitrogen atom stimulates a coordinating site for complexation with different metal ions providing these complex compounds a rich physical, biochemical and organic features [4,5]. Consequently, this kind of ligands has prepared stable

chelated complexes of 5 or 6 membered ring as there another coordination site at ortho location to azo moiety. It is provided through the existence of donor atom such as nitrogen, oxygen and sulfur on the aromatic rings or heterocyclic ring [6,7]. pyrazolin-5-one has drawn substantial attention from scholars because of its stimulating natural activities. [8,9]. Pyrazolin-5-one and 5-Pyrazolones are very vital category of heterocycles attributable to their organic and pharmacological actions[10]. They are employed as main initial materials for the synthesizing viable aryl/hetarylazopyrazolone dyes [11], that are used as noble fastness dyes tufts for wool, cotton, leather, silk, rubber and synthetic polymers (Nylons). On the other hand, many azopyrazolone dyes have been employed as indicators for complexometric titrations [12]. Within the subject of azo dyes, phenolic compounds have a foremost role for synthesizing the majority of viable dyes. Most of these commercially existing dyes

*Corresponding author e-mail: fawzi.almuwashi@uokufa.edu.iq; (Fawzi Yahya Wadday).

Receive Date: 20 November 2020, Revise Date: 12 December 2020, Accept Date: 20 December 2020

DOI: 10.21608/EJCHEM.2020.49716.3034

©2021 National Information and Documentation Center (NIDOC)

have the naphthols bearing hydroxyl groups as an auxochrome group. [13,14]. In recent years, a significant number of tridentate azo compounds have a heteroaryl ring systems and have been developed for improving the colouring features and to accomplish more selectivity and specificity in chemical analysis[15]. Metal dye complexes have a highly significant role in dye- stuff technology [16] and find uses in countless fields, particularly in analytical chemistry [17]. In continuance of studies, the interaction of coordination compounds has shown a speedy expansion in varied disciplines because of the potential use of these new compounds in biological uses. The present work explains the preparing and spectral features of [(1-phenyl-2-pyrazolin-5-one) azo (6-bromo-2-hydroxynaphthalen-1-yl)] (PABH) containing naphtholic-OH function and pyrazolin moiety. The azo dye ligand (PABH) and its metal ions Co(II), Ni(II) and Cu(II) complexes were studied by various spectral analysis and screened for their antimicrobial activity in contradiction of *Staphylococcus aureus* and *Escherichia coli*.

Experimental Section

Instruments, Materials and Approaches

Entirely, the chemicals have bought from BDH and Fluka. FTIR spectra have documented based on KBr on Shimadzu- spectrophotometer in (4000-400) cm^{-1} range. Electronic spectrums in ethanol have documented by means of UV-visible spectrophotometer under (200 - 1100) nm range with quartz cell of (1cm) path leangth. pH measurement has done by means of HANNA instruments pH Tester. Melting points have measured with an electro thermal Stuart apparatus, model SMP30. The measurements of electrical conductivity for complexes have noted at (25°C) for 10^{-3} mol.L⁻¹ solution of the samples in dimethyl sulfoxide (DMSO) by means of WTW inolab cond 720 digital conductivity meter. Microelemental analysis (C.H.N) has gained on a(Eure EA 3000 Elemental analyzer) in Ibn Al-Haitham- College of Education For Pure Science Mass spectra in Agilent mass spectrometer 5975 quadropole analyser. ¹H NMR and ¹³C NMR spectrums have measured on a DRX (500-MHz) spectrometer in DMSO and Bruner DRX (500-MHz) were performed at Sharif Sainte University, Tehran, Iran. Chemical shifts are in ppm relative to internal Me₄Si. Elemental microanalyses of the ligand and their complexes have implemented through Euro Vectro-3000A. The solutions and materials are employed in a biochemical analysis sterilized based on Autoclave, Gallen Kamp. The cultivated bacteria dishes incubated via Memmert Incubator, 854 Schwach. Metal content of complexes have determined by means of atomic absorption method through Analytic Jena(A.A350) atomic

absorption Spectrophotometer. Magnetic susceptibility magnitudes have gotten under room temperature via the Gouy process, Johnson Matthey Catalytic system. Thin Layer Chromatography (TLC): the (TLC) has been accomplished on Al coated plates with silica gel (Fluka), and identified through iodine. Preparation of the Ligand (PABH)

The new azo ligand was created in relation to the Gusev method [18] by dissolving (1.75 g, 0.01 mol) of 3-amino-1-phenyl-2-pyrazolin-5-one in a mixture consisting 25 mL of distilled water and 5 mL of concentrated hydrochloric acid. A solution has diazotized at (0–5 °C) 10 mL of aqueous (0.689 g, 0.01 mol) sodium nitrite. Consequentially, diazonium chloride solution was added to the mixture drop by drop with stir then, 6-bromo-2-naphthol (2.23 g, 0.01 mol) was dissolved in 150 mL of alkaline ethanol and then cooled lower than 0 °C. Next to effecting addition, then left the content for two hours, then 150 mL of cooled distilled water was add, the pH of the solution maintained at 6 by HCl, brown molecules were precipitated and left for 24 h. The precipitate has been filtered off and washed up many times with (1:1) (ethanol : water) mix then recrystallized two times from hot ethanol, and dried in a vacuum desicator. Table (1) consist name of ligand, 83.4 % yield ,177-178 m.p °C and ligand color. The reaction is illustrated in scheme (1).

Preparing the Buffer Solution

Buffer solutions with pH magnitudes from 5 to 9, of acetic acid-ammonium acetate-ammonia (0.01M, 0.77gm) have been primed in one litter of deionized twice as distilled water. The requisite pH has gotten by the adding either ammonia solution or acetic acid.

Preparing the Standard Solution

The metal salt solutions (1×10^{-3} - 1×10^{-6} M) have organized through dissolving the fitting weights of every of these salts [CoCl₂.6H₂O, NiCl₂.6H₂O and CuCl₂.2H₂O] in the equipped buffer solutions. Simultaneously, the range of concentration has employed for preparing the ligand solutions obtained via dissolving (0.04 g, 0.001 mole) from the ligand (PABH) in (100) mL of ethanol. The solution was diluted to obtain concentrations (1×10^{-3} - 1×10^{-6} M).

Determination of Optimum Concentrations

The optimum concentrations for the mixing process, the solutions ranged between (1×10^{-3} - 1×10^{-6} M) were determined for both the ligand and metal ions. Equal volumes were mixed for both the ligand and metal ions at rang of acidity functions (5-9) and measured the absorption

value for these solutions. The high-ranged concentrations (1×10^{-3} and 6×10^{-4} M) showed complex precipitation for instant mixing of these solutions. So the concentrations between (1×10^{-4} - 5×10^{-4} M) were chosen because it gave acceptable absorption and some of them comply with the Beer - Lambert law, as for the minimum concentrations (1×10^{-5} and 1×10^{-6} M), they gave a weak measured.

Determination of Optimum Acid Function

The effected of acidic function change was studied at a range of pH (5-9) to reach the optimum acidic function of the metal ions after the optimal conditions were established, which included the determination of the optimal concentrations of ligand and metal ions.

Determination of Standard Calibration Curve of the Metal Complexes

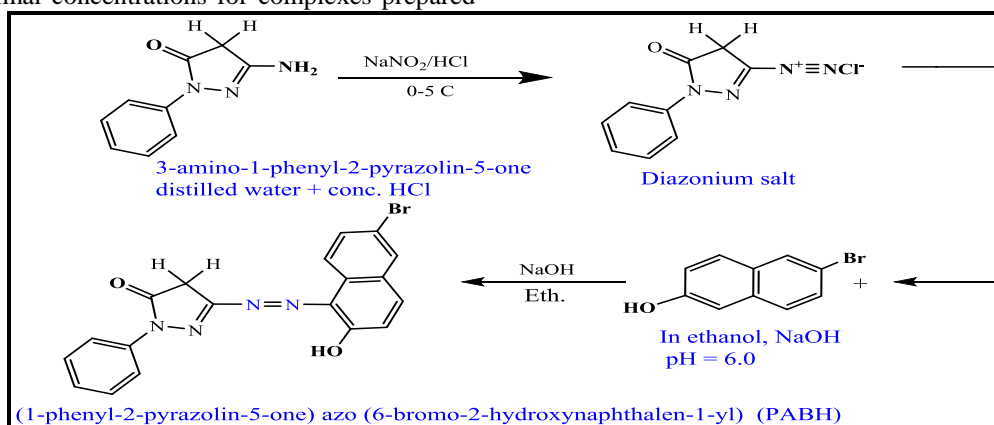
A solutions ranged between (1×10^{-4} - 5×10^{-4} M) for both the ligand and the metal ions studied, the equal volumes of same concentration of metal salts solutions and ligand solutions were mixed at optimal acidic function and the wavelength that gives the highest absorbance (λ_{max}). The absorption values of these solutions in practical experiments showed that the optimal concentrations for complexes prepared

and suitable for spectral measurements fall within the range (1×10^{-4} - 5×10^{-4} M) were obeyed the Lambert-Beers law.

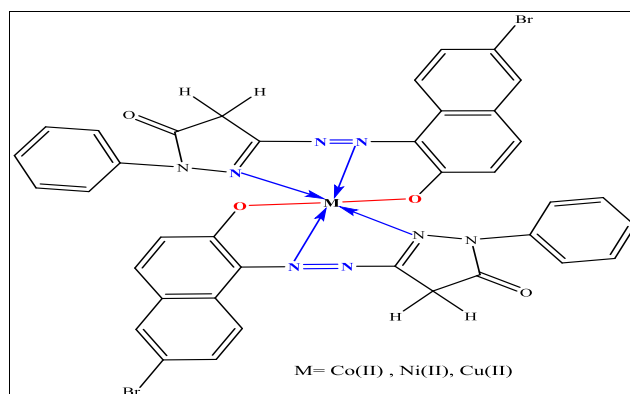
According to the graphs plotted of the standard calibration curve for the metal ions complexes solutions, the relationship between the ligand and metal ions concentrations was obeyed the Lambert-Beers law.

Synthesis of Complexes

The complexes have equipped through adding dropwise based on hot stirring ethanolic solution of (2 mmole) ligand to stoichiometry amount of M:L ratio of (1:2) of [Co(II), Ni(II) as well as Cu(II)] that dissolved in a primed buffer solution at finest pH. A mix has heated up under (60°C) and stirred up to (1 hour). After that, at room temperature, they have left to cool. A colored precipitate has filtered, washed many times with a (1:1) (water:alcohol) mix then left to desiccated in vacuum desiccator. A recommended stoichiometry structure for the complexes has been illustrated in scheme (2), Figure (1) 3D. Table (1) presents, the suggested formula, color, m.p, and yield ratio of formed solid complexes.



Scheme 1: Preparation reaction of the azo ligand (PABH)



Scheme 2: The proposed molecular structure of complexes, M= Co(II), Ni(II) and Cu(II)

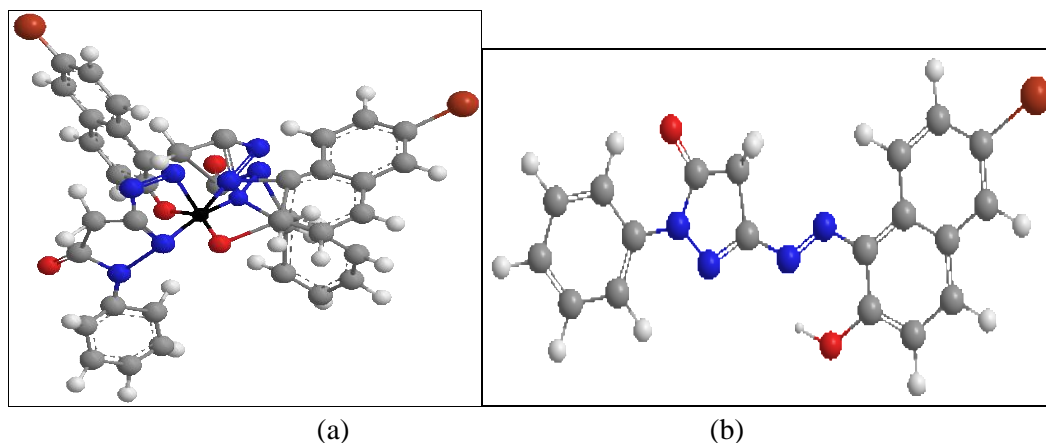


Fig. 1: The proposed molecular structure 3D of (a) azo ligand and (b) complexes, M(II) = Co, Ni and Cu

Antimicrobial Activity Study

Antibacterial activities of the new Azo ligand and its metal complexes were screened by Kirby-Bauer disk diffusion method [19]. This paper employs dual strains for pathogenic bacteria first, a *Staphylococcus aureus* (Gram Positive) and the other strain, *Escherichia coli*. (Gram Negative). The used biochemical solutions were organized through dimethyl sulfoxide (DMSO) as solvent, where a single concentration (C) $1 \times 10^{-3} \text{M}$ is joined. The dishes have been incubated under 37°C for a complete day. Based on the inhibition zones, the diameter (Inhibition area) mm has formed after a complete day as a principle for an intensity of the influence of synthetic chemical compounds on a growth of specific cultivated bacteria strains

Consequences and Discussion

General

The Azo ligand (PABH) is pale brown crystal, which is not soluble in water and soluble in common organic solvents. The reaction of this ligand with the metal ions gives different color crystals. All complexes are quiet water - insoluble, stable in air, while they are soluble in most organic solvents such

as DMSO, DMF, acetone...etc.

Physical Properties and Elemental Analyses

Reacting the organized ligand (PABH) with a selection of metal ions [Co(II), Ni(II) and Cu(II)] at finest concentration and pH, that causes formed complexes with formula $[\text{Co}(\text{PABH})_2]$, $[\text{Ni}(\text{PABH})_2]$ and $[\text{Cu}(\text{PABH})_2]$. The ligand has performance as N,N,O'-chelator, in which N (azo), N (2-pyrazolin-5-one) and O' hydroxyl (6-bromo-2naphthol). The physical features and consequences gotten from C.H.N.O examinations and metal contents of the organized compounds are explained in Table (1). The analytical data have been well accepted with the computed magnitudes. The molecular formula of the ligand and its metal complexes have been suggested according to the data which were resulted from spectral and magnetic susceptibility of metal complexes. In all cases (1:2) metal to ligand solid complexes are isolated.

Table 1: Physical details and analytical information of the made azo ligand (PABH) besides its complexes

Empirical formula (M . Wt) gm/mol	pH	Color	Mp., °C	Yield, %	Found(Calc.)(%)					μ_{eff} , B.M.
					C	H	N	O	M(II)	
Ligand $\text{C}_{19}\text{H}_{13}\text{BrN}_4\text{O}_2$	—	Pale Brown	177-178	83.4	55.76 (56.02)	3.2 (3.23)	13.69 (13.82)	7.81 (7.53)	—	—
$[\text{Co}(\text{C}_{19}\text{H}_{12}\text{BrN}_4\text{O}_2)_2]$	8	Reddish-Brown	196-197	76	52.13 (52.34)	2.76 (2.81)	12.8 (12.71)	7.31 (7.39)	6.73 (6.98)	4.691
$[\text{Ni}(\text{C}_{19}\text{H}_{12}\text{BrN}_4\text{O}_2)_2]$	8	Yellowish-Green	188-189	74.1	52.15 (52.41)	2.76 (2.72)	12.8 (12.89)	7.31 (7.42)	6.7 (6.74)	3.307
$[\text{Cu}(\text{C}_{19}\text{H}_{12}\text{BrN}_4\text{O}_2)_2]$	9	Pale Green	207-208	76.19	51.86 (52.05)	2.74 (2.86)	12.73 (12.65)	7.27 (7.31)	7.22 (7.51)	1.846

Influence of pH

The pH magnitudes intended for metallic ions complexes solutions have been investigated for detecting a finest pH medium for preparing the metallic complexes, the best concentration of complexes has reserved as provided an ascension to study (λ_{max}) at numerous pH magnitudes. The pH influence has been as well examined within (5-9) range. Figure (2) depicted an absorbance for pH curves that had completed at (λ_{max}) and some concentrations under study for every metal ion solution. The huge band of the curves for pH which was reflected as a reference of complex formation and acceptance as finest pH for preparing complexes. Hence, we deduce that all equipped chelate complexes with a selection of metal ions have been prepared in basic medium [20,21].

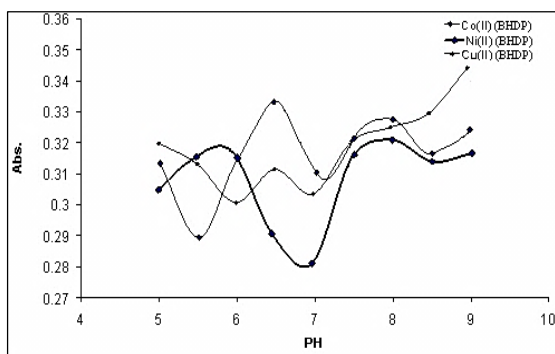


Fig. 2: Influence of the acidic function (pH) on the absorbance for the metal ions-(PABH) complexes solution at concentration (5×10^{-4} M) and λ_{max}

NMR Spectra for the Azo Ligand

The $^1\text{H-NMR}$ spectrum of ligand (PABH) in DMSO-d_6 Figure (4) was shows a single peak appeared at $\delta(10.458)$ ppm which was attributed to chemical shift of OH proton in the 2-naphthol [13],[20]. A multiplet peak at $\delta(8.461 - 6.884)$ ppm that have been as a result of chemical shifts of aromatic protons for aromatic rings of 2-naphthol and phenyl moieties [22,23,24]. The doublet signal observed at $\delta(3.561 - 3.514)$ ppm were assigned to H-C-H protons on the pyrazolin ring moiety in the ligand [22],[25].

Mole Ratio [Metal:Ligand] Ratio

The mole ratio technique was adopted with the intention of identifying spectrophotometrically of a composition of metal chelate complexes and for conclusion of potential structural formula of organized metallic ions complexes, this technique has been in accordance with an range measurement for every complexes solution under fixed finest pH and concentration at a supreme absorbance wavelength. The colors intensity for the solution of equipped metallic ions complexes has raised based on intersection point for [M:L] ratio approach. The continual stability of color beyond an intersection point signifying the formed metallic complexes [21]. The structure of formed complexes in solutions was proven by mole ratio method. Accordingly, the consequences show a mole ratio [M:L] [1:2] for entire metal ions to ligand ratio. The method supports the complexes formation, as explained by Figure (3).

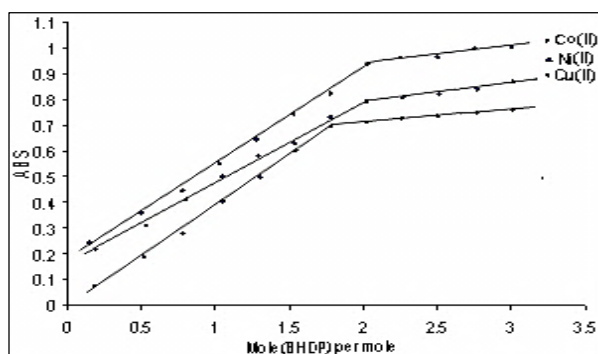


Fig. 3: The mole ratio plot of metal ions(PABH) ligand complexes solution at optimum pH and λ_{max}

$^{13}\text{C-NMR}$ spectrum Figure (5) displayed a peak at $\delta(178.22)$ ppm which is due to pyrazolin carbonyl group[23], while the C=N pyrazolin moiety carbon signal is showed at $\delta(168.26)$ ppm [25]. The multiplet peaks at $\delta(162.16 - 122.23)$ ppm are due to aromatic carbons of 2-naphthol and phenyl moieties [26]. A peak at $\delta(167.13)$ ppm which is assigned to C=C-OH and a signal at (121.81) duo to C=C-Br carbons in 2-naphthol rings [13]. The peak at $\delta(163.74)$ ppm is due to C=C-N site linked of 2-naphthol with azo group for pyrazolin ring [20],[27]. The signals at $\delta(47.34)$ ppm are assigned to the middle and terminal C-C carbon atoms of diaminobutane moiety in the ligand [12],[25],[27].

UV-Vis Spectral Studies

The electronic absorption bands along with the conductivity values have briefed in Table (2). The UV-Visible spectrum of azo ligand Figure (7) in ethanol (5×10^{-4} M) within the range (200-1100) nm appeared two absorptions at (244 and 295) nm (40983 and 33898) cm^{-1} , which is due to $\pi-\pi^*$ transition and a broad low intensity band at (420) nm (23809) cm^{-1} , which was attributed to $n \rightarrow \pi^*$ transition of (C.T) intermolecular charge-transfer taken place through the azo group ($-\text{N}=\text{N}-$), Fig. (1). [16,28,29].

The spectra of the metal ions complexes within (5×10^{-4} M) at optimum pH showed bathochromic transfers of ligand band. The assigned bands to intraligand $\pi \rightarrow \pi^*$ for the Co(II), Ni(II), and Cu(II) complexes were observed at (38055 , 32172), (37811 , 31523) and (37535 , 31014) cm^{-1} respectively. The changing in a color of free ligand solutions and high shift in the (λ_{max}) offers a worthy sign for coordinating and complex forming [28],[30]. The spectra of complexes depicted added low intensity bands in the perceptible regions associated with ligand field d-d transitions [20],[26]. Co-complex spectrum in ethanol solution in Figure (8), revealed dual bands at (510) nm (19607.84) cm^{-1} and (590) nm (16949.15) cm^{-1} , which were attributed to the ${}^4\text{T}_{1g} \rightarrow {}^4\text{T}_{1g}(\text{p})$ (ν_3)

and ${}^4\text{T}_{1g} \rightarrow {}^4\text{A}_{2g}$ (ν_2) transitions correspondingly of octahedral geometrical structure [31],[31]. Based on a ratio of ν_3/ν_2 (1.15), a magnitude of Dq/B (1.73) has been gotten. The magnitude of B' (688.53) in addition to a position of $\nu_1(10Dq)$ (12447.36) cm^{-1} have analyzed by means of Tanaba-Sugano diagram for d^7 structure for octahedral configuration geometry[31,32]. The value of β (0.67) indicates some covalent character. The conductivity measurement in ethanol as solvent, specifies that the Co-complex is non-ionic. Spectrum of Ni(II) complex Figure (9), depicted two bands in a visible region at (573) nm (17452) cm^{-1} and (446) nm (22421.52) cm^{-1} which are due to ${}^3\text{A}_{2g} \rightarrow {}^3\text{T}_{1g}(\text{F})$ (ν_2), ${}^3\text{A}_{2g} \rightarrow {}^3\text{T}_{1g}(\text{P})$ (ν_3) respectively. The ratio of ν_3/ν_2 , (1.28) has applied on Tanaba-Sugano diagram regarding d^8 octahedral complexes,[31,32,33] B_{complex} (774) and β (0.73), Dq/B (1.17), $10Dq(\nu_1)$ (9053.61) cm^{-1} have computed hypothetically. The conductivity has depicted that the Ni(II)-complex has been non-electrolyte. The Cu(II) complex spectrum in Figure (10), has depicted broad band at (650) nm (15384.61) cm^{-1} assigned to ${}^2\text{E}_g \rightarrow {}^2\text{T}_{2g}$ transition that signifies Jahn-Teller distortion of octahedral geometry[27],[31]. A measurement of conductivity for the complex signposts in which a complex has been non-ionic.

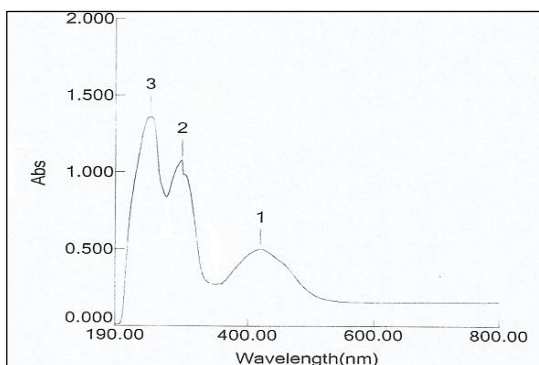


Fig. 7: UV-Vis spectrum of Ligand (PABH)

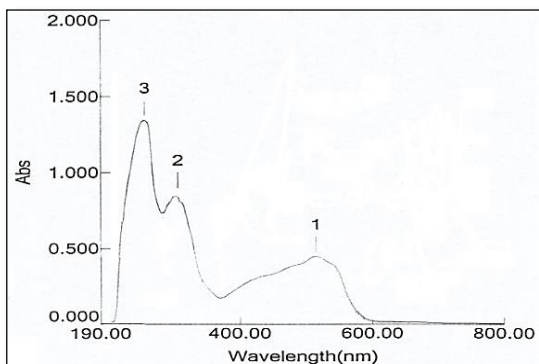


Fig. 9: UV-Vis spectrum of Ni(PABH)₂ complex at pH=8

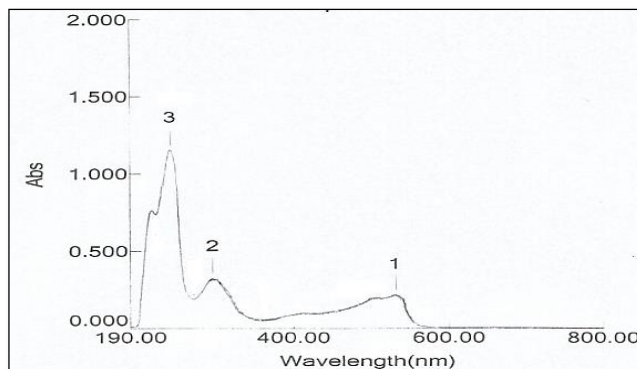


Fig. 8: UV-Vis spectrum of Co(PABH)₂ complex at pH=8

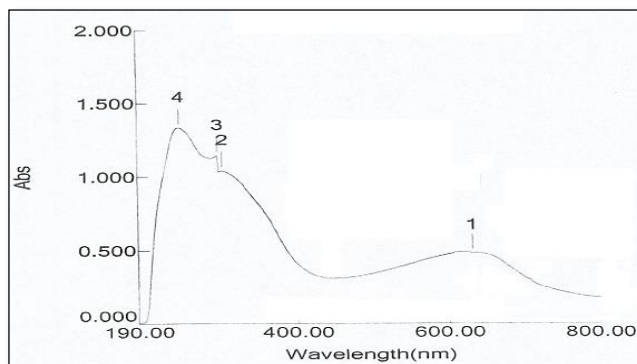


Fig. 10: UV-Vis spectrum of Cu(PABH)₂ complex at pH=9

Table 2: Electronic spectral bands (cm⁻¹) and conductivity for complexes

Complex	Band position, cm ⁻¹	Assignments	B _{complex}	β	Dq/B	10Dq (ν ₁) theoretical, cm ⁻¹	Λ _m Ω ⁻¹ cm ² .mol ⁻¹
[Co(PABH) ₂]	19607.84 ν ₃ 16949.15 ν ₂ 12447.36 (cal.)ν ₁	⁴ T _{1g} → ⁴ T _{1g(P)} ⁴ T _{1g} → ⁴ A _{2g(F)} ⁴ T _{1g} → ⁴ T _{2g(P)}	688.53	0.67	1.73	12447.36	10.14
[Ni(PABH) ₂]	22421.52 ν ₃ 17452 ν ₂ 9053.61(cal.)ν ₁	³ A _{2g} → ³ T _{1g(P)} ³ A _{2g} → ³ T _{1g(F)} ³ A _{2g} → ³ T _{2g(F)}	774	0.73	1.17	9053.61	9.39
[Cu(PABH) ₂]	15384.61	² E _g → ² T _{2g}	—	—	—	—	12.07

The UV-Vis spectra for complexes were studied within the concentration range of (1x10⁻³-1x10⁻⁶) molar, whereas the pH sequence has been (5-9). Not each concentration is appropriate for spectral measurements, while a solution in (1x10⁻⁵ M) concentration go along with Lambert-Beer's law with an obvious color indication. Figure (11) has depicted the finest fit straight lines, as the absorbance plotted vs. molar concentration under (1x10⁻⁴- 5x10⁻⁴) molar range.

IR Spectra of Azo Ligands and Complexes

The infrared spectral data of azo ligand and its complexes have illustrated in Figures (12-15) and Table (3). The IR spectra for complexes have compared with those of the free ligand with the intention of determining the coordination sites that can be included in chelation.

The IR spectrums offer worthy facts about the nature of functional groups in a ligand and some of which involved to the metal ion [22],[26]. The IR spectrum of the free ligand (PABH), exhibited a wide and strong intensity band at (3397 cm⁻¹), which was attributed to the stretching vibration of the hydroxyl group in the naphthol[23,34], this band disappear in the complex spectrum, that indicating the oxygen atom

of the hydroxyl group in the 2-naphthol was attached with the metal ions in the complexes. The ligand showed a strong band at (1628 cm⁻¹) due to (N=N) this band undergo change to lower frequency in the spectrum of the complex signifying an engaged one of the two nitrogen atoms of the azo group in the coordinating with the metal ions[24,32]. The band shoulder at 1725 cm⁻¹ in a spectrum of a free ligand, attributed to ν(C=O) on pyrazolin ring [25],[28] the position of this band remained at approximately the identical frequency in spectrums of metal complexes, signifying the noncoordination of this group [35]. The band appearing at 1650 cm⁻¹ in the free ligand pyrazolin ring, assignable to the ν (C=N) vibration mode[24,35], is shifted to lower wave numbers with a Δν at range (30-19) cm⁻¹ in the complexes spectra, this indicating the involvement of pyrazolin nitrogen atom in coordination[22,25]. A medium intensity band at (1153cm⁻¹) showed in the ligand spectrum returning to the (C-N) (carbon 2-naphthol - nitrogen azo), when compared to the spectrum of the complex, a slight change in its position and shape was observed due to the coordinate the metal ion with the close azo nitrogen atom[35,36]. The presence of dual new bands in the region 482-468 and 444-423 cm⁻¹ in the complexes spectra, as a result of ν(M-N) and ν(M-O) stretching vibrations respectively[23,27,36], also confirmed the formation of metal complexes.

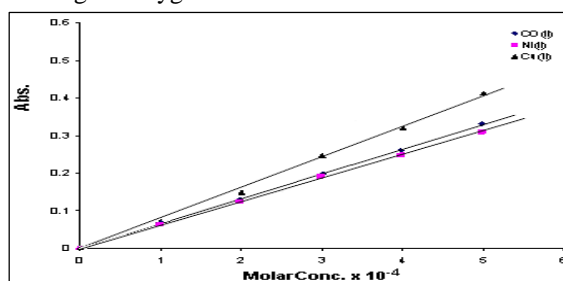
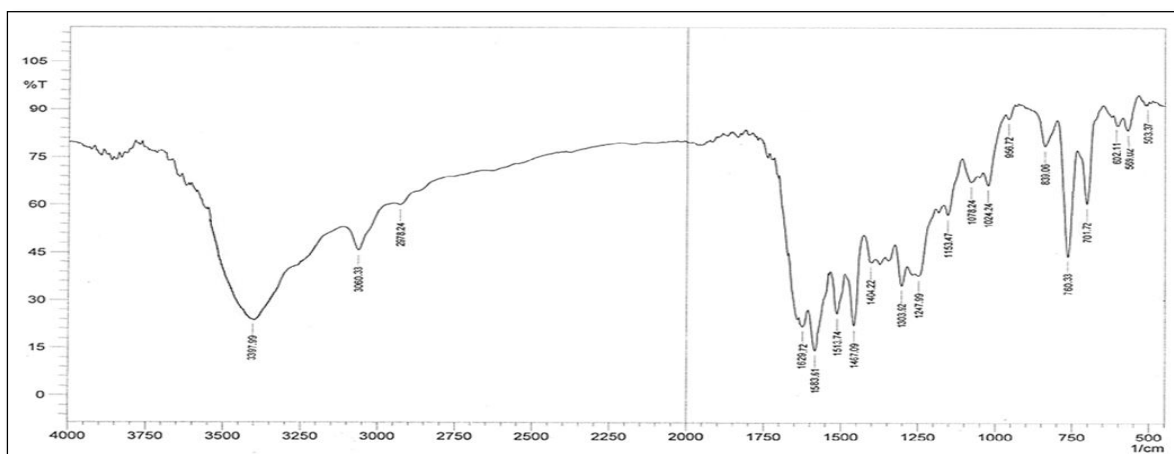
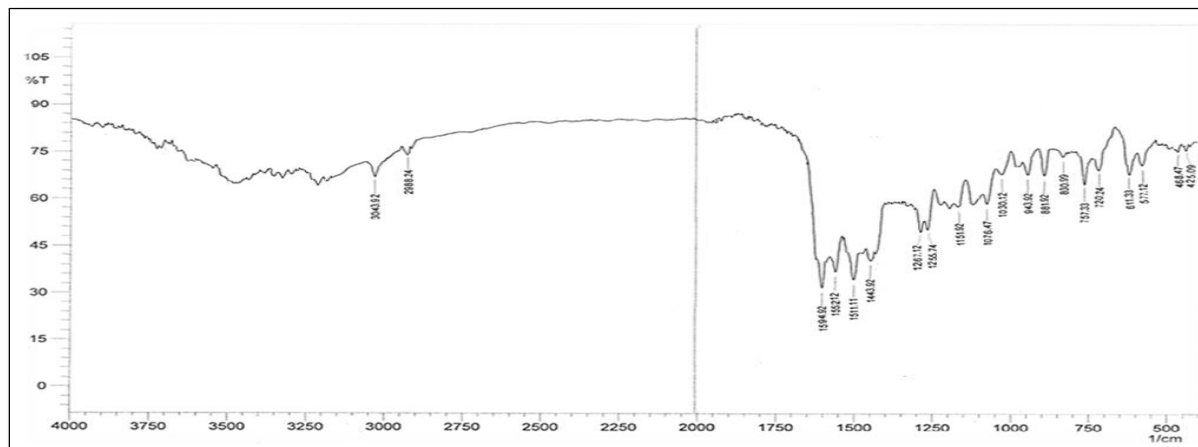


Fig. 11: The linear relationship between absorbance and the range of selected concentrations for metal ions complexes at optimum pH and λ_{max}

Table 3: Most Imperative FTIR spectral bands and their Assignments of the azo ligand and its Complexes (cm⁻¹)

Compounds	ν (O-H) 2-naphth	ν (C=N) pyrazolin	ν (N=N) azo	M-N)(ν)	M-O)(ν)
$C_{19}H_{13}BrN_4O_2=(PABH)$	3397,b	1650,m	1629,m	—	—
$[Co(PABH)_2]$	—	1620,m	1594,m	468,w	425,w
$[Ni(PABH)_2]$	—	1639,m	1596,m	474,w	423,w
$[Cu(PABH)_2]$	—	1631,m	1591,m	482,w	444,w

**Fig. 12: FTIR spectrum of azo ligand (PABH)****Fig. 13: FTIR spectrum of $[Co(PABH)_2]$ complex**

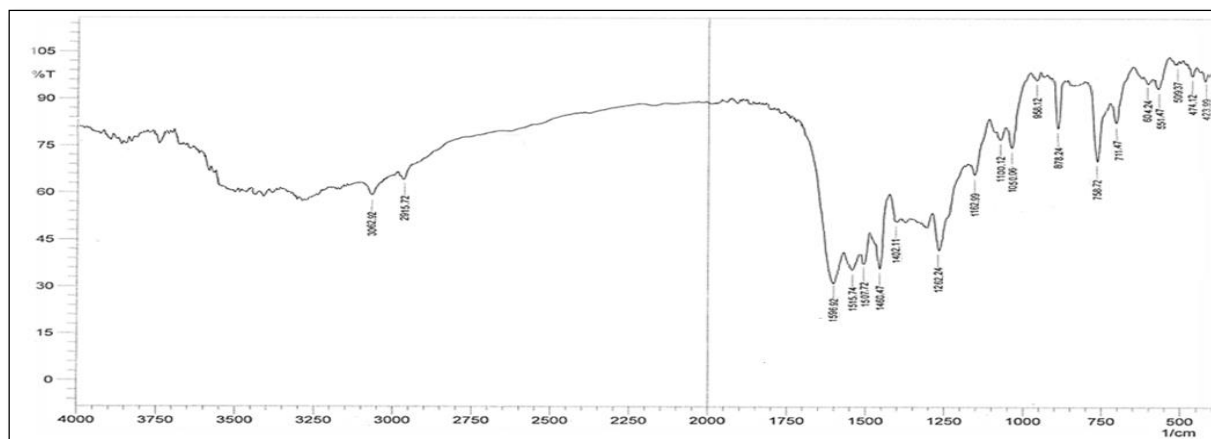


Fig. 14: FTIR spectrum of [Ni(PABH)₂] complex

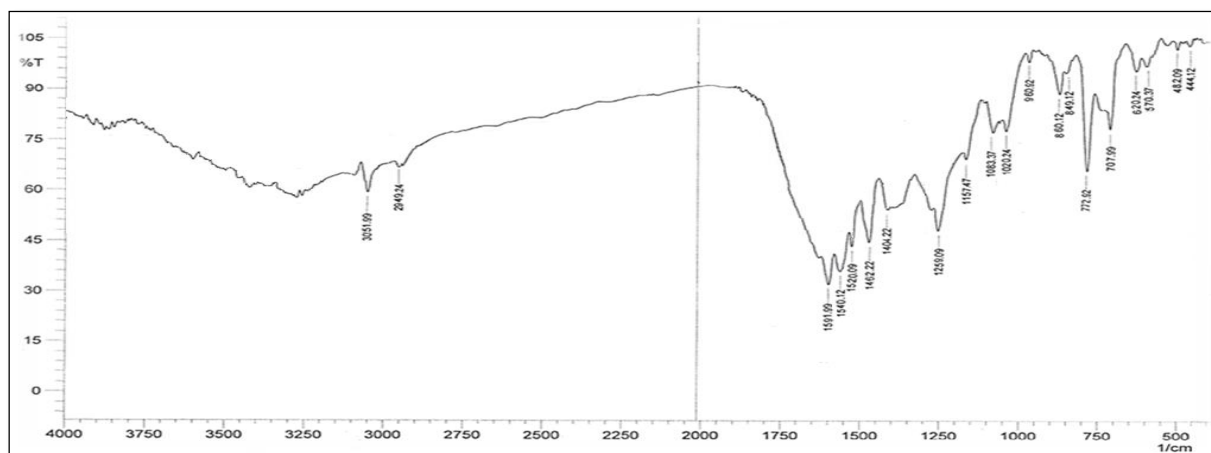


Fig. 15: FTIR spectrum of [Cu(PABH)₂] complex

Thin Layer Chromatography (TLC)

A solution of azo ligand besides its complexes in ethanol as solvent, shown as one spot each, ratifying

that these compounds, entirely, have been pure and possess only single isomer[31]. Table (4) explains the R_f for complexes and the ligand.

Table 4: Shows the R_f for the azo ligand (PABH) and its complexes in ethanol solvent

Compound	Molecular weight (g/mol)	R _f eth. solv.
Ligand C ₁₉ H ₁₃ BrN ₄ O ₂	409.6	0.41
[Co(C ₁₉ H ₁₂ BrN ₄ O ₂) ₂]	875.39	0.28
[Ni(C ₁₉ H ₁₂ BrN ₄ O ₂) ₂]	875.15	0.3
[Cu(C ₁₉ H ₁₂ BrN ₄ O ₂) ₂]	880.00	0.25

Antimicrobial Activity

The response of selected bacteria, we observed the excessive biochemical effect of the azo ligand and its complexes studied at concentration (1x10⁻³ M) with dual varieties of pathogenic bacteria (*Staphylococcus aureus*), its Gram Positive and either with the germ (*Escherichia coli*), Gram negative bacteria, have depicted lower response to ligand besides its

complexes investigated from the other category of bacteria, categorized it's resistant to numerous chemical compounds and antibiotics[19],[29],[35]. The cause for this resistant is the colon bacteria that are found in a distinct bacilli having thick casing surroundings of its cell, this casing has an excessive amount of lipid works to oppose these materials from inflowing a cell, whereas the *Staphylococcus aureus* bacteria don't have this feature. Accordingly, it will

be in lower resistance in an arrival of chemical and antibiotic substances to inner bacterial cell [30,31],[36].

Normally, the prepared complexes have explained biochemical effect more as compared with the ligand (PABH), while the ligand contain nitrogen and oxygen atoms biological retardant[22],[31],[37]. The positive charge ion in the chelated complex has

been partly shared with the donor atoms orbital in a ligand and there has been π -electron delocalization over the entire chelate ring, which decrease in the polarity of the metal ion to a greater level, this in turn increases a lipophilic character of a metal chelate and favors its permeation via the lipid layers of the membrane the microorganisms[31],[37]. The consequences presented in previous activity Tables (5) and figures (16,17).

Table 5. The data of antibacterial activity (zone of inhibition) (mm) of azo ligand (PABH) and its complexes

Compound	G-(<i>E.coli</i>)	G+(<i>St.aureus</i>)
Control (S) DMSO	6	6
Ligand (PABH)	11.5	14
[Co(PABH) ₂]	12.3	15.8
[Ni(PABH) ₂]	13	16.2
[Cu(PABH) ₂]	14.7	19.4

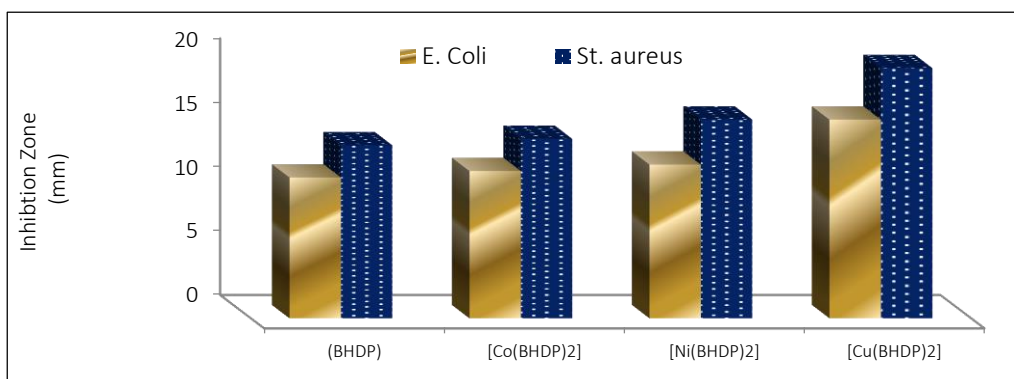


Fig. 16: The biological effects of azo ligand (PABH) and its complexes

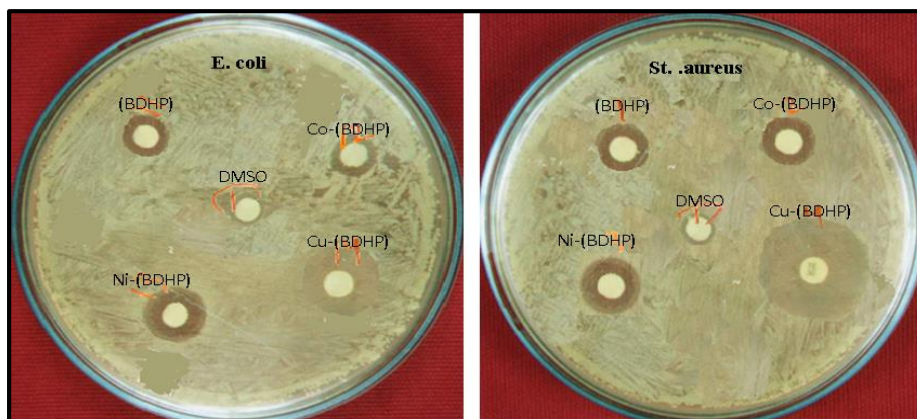


Fig. 17: Photograph of antibacterial activities of the ligand (PABH) and its complexes

Conclusion

The azo ligand (PABH), coordinates with Co(II), Ni(II) and Cu(II) ions based on an tridentate nitrogen atom of the azo, nitrogen atom of pyrazolin ring and oxygen atom of the hydroxyl in the 2-naphthol groups resulting in six-coordinated metal ion, this behavior with M:L mole ratio of 1:2. Each complex has been of octahedral geometry and categorized by numerous

physio-chemical techniques, the analysis ratified the composition and structures for the gained complex jointly. Biological activity results have depicted that all the compounds have variety of antibacterial activities.

Acknowledgment

I would like to state my sincere gratitude to the Department of Chemistry, College of Education,

University of Al-Qadisiyah, and College of Science, Kufa University in Iraq for funding this work.

References

- [1] Koh J. and Greaves A. J., Synthesis and Application of An Alkali-Clearable Azo Disperse Dye Containing A Fluorosulfonyl Group And Analysis of Its Alkali-Hydrolysis Kinetics, *Dyes Pigments*, 50, 117-126 (2001).
- [2] Nabeshima Y.; Shishido A.; Kanazawa A.; Shiono T; Ikeda Tand Hiyama T., Optical Switching by Means of Photoinduced Change in Refractive Index of Azobenzene Liquid Crystals Detected by Reflection-Mode Analysis, *Chem. Mater*, 9, 1480-1487 (1997).
- [3] Yin S.; Xu H.; Shi W.; Gao Y.; Song Y. and Wing J., Synthesis and optical properties of polyacetylenes containing nonlinear optical chromophores, *J. Polymer*, 46(18), 7670-7677 (2005).
- [4] Kareem , I; Waddai, F; Abbas, G., Synthesis, Characterization and biological activity of Some Transition Metal Complexes with New Schiff Base Ligand Type (NNO) Derivative from Benzoin. *J. Pharm. Sci. and Res.*, 11(1), 119-124 (2019).
- [5] Bal S. and Connolly J. D., Synthesis, characterization, thermal and catalytic properties of a novel carbazole derived Azo ligand and its metal complexes, *Arab. J. of Chem.*, 10,761-768 (2017).
- [6] Fu Z. and Viraraghavan T., Fungal decolorization of dye wastewaters: a review, *Bioresour. Technol, A review, Elsevier*, 79, 251-262 (2001).
- [7] Alyakhider A. and Kadhim R. S., Preparation, Spectral and Biological Studies of Azo Ligand Derived from Proline with Cu(II), Ag(I) and Au(III) Metal Ion, *IOSR J. of Appl.Chem*, 9(8), 20-31 (2016).
- [8] Jamwal A.; Javed A. and Bhardwaj V., A review on pyrazole derivatives of pharmacological potential, *J. Pharm. BioSci.*, 3, 114-123 (2013).
- [9] El-Hawash S.; Badawey E. S. and El-Ashmawey I., Nonsteroidal antiinflammatory agents-part 2 antiinflammatory, analgesic and antipyretic activity of some substituted 3-pyrazolin-5-ones and 1,2,4,5,6,7-3H-hexahydroindazol-3-ones, *Eur. J. Med. Chem.*, 41, 155–165 (2006).
- [10] Scheibye S.; El-Barbary A. A.; Lawesson S. O.; Fritz H. and Rihs G., Studies on Organophosphorus Compounds-XLI: Formation of 3-pyrazoline-5-thione disulfides From 3,5-pyrazolidinediones. C-alkylation of 3,5-pyrazolidinediones, *Tetrahedron*, 38(24), 3753-3760 (1982).
- [11] Ran C.; Byunghun C. and Joonseok K., Synthesis and properties of some novel pyrazolone-based heterocyclic azo disperse dyes containing a fluorosulfonyl group, *Indian J. of Chem., Dyes Pigments*, 95(3), 580-586 (2012).
- [12] Zaiter J.; Achibat H.; Amiri O.; Hafid A.; Khouili M.; Rakib E. M.; Neves M.B.; Neves M.S.; Silva M.S.; Cavaleiro A.S.; Almeida F.A. and Faustino M.A.F., An easy synthetic access to new pyrazole spiro derivatives from 3-amino-1-phenyl-2-pyrazolin-5-one, *New J. of Chem.*, 1-3 (2013).
- [13] Himani N. C.; Jyotsna S. M.; Ramakanth P.; Arvind J. M., Synthesis, characterization and antibacterial activity of some novel zo-azomethine dyes of 6 Bromo-2-naphthol, *Intern.l J. of Chem. Tech. Res.*, 2(3), 1823-1830 (2010).
- [14] Jarrahpour A. A.; Motamedifar M.; Pakshir K.; Hadi N. and Zarei M., Synthesis of Novel Azo Schiff Bases and Their Antibacterial and Antifungal Activities, *Molecules*, 9(10), 815-824 (2004).
- [15] Al-Atbi H. S.; Al-Salami B. K. and Al-Assadi I. J., New azo-azomethine derivative of sulfanilamide: Synthesis, Characterization, Spectroscopic, Antimicrobial and Antioxidant activity study, *Phys.: Conf. Ser.*, 1294 052033, 1-12 (2019).
- [16] Kofikye S.; Akaranta O. and Darko G., Synthesis, characterization and antimicrobial activity of peanut skin extract-azo-compounds, *Scient. Afric., Elsevier*, 8, 1-12 (2020).
- [17] Zaubitzer F.; Riis-Johannessen T. and Severin, K., Sensing of peptide hormones with dynamic combinatorial libraries of metal–dye complexes: the advantage of time-resolved measurements, *Org. Biomol. Chem.*, 7(22), 4598-4603 (2009).
- [18] Gusev S. I ; Zhvakina M. V.; Kozhevnikov I. A., Thiazolylazo Dyes and Their Spectrophotometric Applications in Analytical Chemistry, *Zh. Analit.Khim.*, 26, 859-864 (1971).
- [19] Kirby W.A.; Bauer A.W.; Sherris J. and Turk M., Antibiotic Susceptibility Testing by a Standardized Single Disk Method, *American. J. Clin.Pathol.*, 45(4), 493-496 (1966).
- [20] Jarad A. J.; Majeed I. Y. and Hussein A. O., Synthesis and spectral studies of heterocyclic azo dye complexes with some transition metals, *J. Phys.: Conf. Ser.* 1003, 1-14 (2018).
- [21] Masoud M. S.; Mohamed G. B.; Abdul-Razak Y. H., and Ali A. E., Spectral, Magnetic, and Thermal Properties of Some Thiazolylazo Complexes, *J. Korean. Chem. Soc.*, 46(2), 99 (2002).
- [22] Al-Khateeb Z. T.; Karam F. F. and Al-Adilee K., Synthesis and Characterization of Some Metals Complexes With New Heterocyclic Azo Dye Ligand 2-[2- (5- Nitro Thiazolyl) Azo]-4- Methyl -5- Nitro Phenol and Their Biological Activities, *J. Phys.: Conf. Ser.*, 1294(4), 1-18 (2019).
- [23] Vi dya v. G.; Sadasivan V.; Meena S. S. and Bhatt P ., Synthesis, Spectral and Biological Studies of Complexes with Bidentate Azodye Ligand Derived from Resorcinol and 1-Amino-2-Naphthol-4-Sulphonic Acid, *Orient. J. of Chem.*, 34(1), 45-54 (2018).
- [24] Al-Muhanaa S. S.; Al-Khafagy A. H., Preparation and Biological Activities of New Heterocyclic Azo Ligand and Some of Its Chelate Complexes, *J. Nano Biomed. Eng.*, 10(1), 46-55 (2018).
- [25] Apotrosoaei M.; Vasincu I. M.; Dragan M.; Buron F.; Routier S and Profire L., Design, Synthesis and the Biological Evaluation of New 1, 3-Thiazolidine-4-ones Based on the 4-Amino-2, 3-dimethyl-1-phenyl-3-pyrazolin-5-one Scaffold, *J.of molecu.*, 19 (2014).
- [26] Gholamhassan I.; Alireza B.; Taarof F. and Zahra S., Green synthesis of novel isatin thioketal derivatives

- under solvent-free conditions, *J.Green Chem. Lett. and Rev.*, 10 (1), 1-9 (2017).
- [27] AL-adilee K. J. and Hessoon H. M., Synthesis , Spectral Properties And Anticancer Studies of Novel Heterocyclic Azo Dye Ligand Derived From 2-Amino-5-methyl thiazole with Some Transition Metal Complexes, *Conf. Ser.:1243, J. of Phy., 1243*, 1-20 (2019).
- [28] Timothy R. H.; Noah M. B. and Stephen F. M., Syntheses of Gliocladin C and Related Alkaloids, *J.Org. Lett., Americ. Chem. Soci.*, 19 (9), 2254–2257 (2017).
- [29] Kareem I. K.; Waddai F. Y. and Abbas G. J., Synthesis, Characterization and biological activity of Some Transition Metal Complexes with New Schiff Base Ligand Type (NNO) Derivative from Benzoin, *J. of Glob. Pharm. Tech*, 11(1), 119-124 (2019).
- [30] Al-Adilee K., Kyhoiesh H. A., Preparation and Identification of Some Metal Complexes with New Heterocyclic Azo Dye Ligand 2-[2-(1-Hydroxy-4-Chloro phenyl) azo]-Imidazole and their Spectral and Thermal Studies, *Elsevier J. of Molec. Stru.*, 1137, 160-178 (2017).
- [31] Waddai F. Y.; Kareem E. K. and Hussain S. A., Synthesis, Spectral Characterization and Antimicrobial Activity of Some Transition Metal Complexes with New Schiff Base Ligand (BDABI), *Orient. J. of Chem.*, 34(1), 434-443 (2018). [32] Hadi M. A. and Kareem I. K., Synthesis and Characterization of Some Transition Metal Complexes with New Azo- Schiff Base Ligand 3,4-bis(((1E,2E)-2-((2-((4-((Z)-(3-Hydroxyphenyl)Diazenyl)Naphthalen-1-yl)amino)ethyl)imino)-1,2-Diphenyl ethylidene)Amino) Phenyl) (phenyl)Methanone, *Egypt.J.Chem.*, 63(1), 301-313 (2020).
- [33] Abdul Karem L. K. and Waddai F.Y., Ni(II) and Cd(II) complexes derivatives from heterocyclic Schiff base ; Synthesis, spectroscopic and biological studies, *J. of Glob. Pharm. Tech*, 10(8), 201-208 (2018).
- [34] Çanakçı D., and Serin S., Synthesis of New Azo Dye Polymers Based On Naphthol By Oxidative Polycondensation: Antimicrobial Activity and Fastness Studies, *J. of Polym. Res.*, 27(1), 1-23 (2020).
- [35] Abdul karem L. K. and Mahdi S. H., Spectroscopic, Structural and Antibacterial Activity of Mixed Ligand Complexes from Schiff Base with Anthranilic Acid, *J. Phys.: Conf. Ser.* 1234, 1-13 (2019).
- [36] Bal S., A Novel Azo-Schiff Base Ligand and Its Cobalt, Copper, Nickel Complexes: Synthesis, Characterization, Antimicrobial, Catalytic and Electrochemical Features, *J. of Scie. and Tech. A- Appl. Scie. and Eng.* , 17(2), 315-326 (2016). [37] Abd-Ali H. H.; Al-Salami B. K. and Abd M. A., Synthesis, Characterization And Antibacterial studies of some Azomethine and Azo- Compound Derivatives of selected Sulfa Drugs, *Intern. J. of Appl. Chem.*, 7(2), 32-47 (2020).

تحضير و تشخيص طيفي وتقييم التضاد البكتيري لبعض المعقدات الفلزية الثنائية مع ليكاند ازو حلقي غير متجانس ثلاثي السن جديد من نوع مانح (نيتروجين نيتروجين اوكسجين)

الخلاصة

في هذا البحث، تم تحضير ليكاند ازو جديد [1-فنيل-2-بايرازولين-5-اون(6-برومو-2-هيدروكسي نفتالين-1-يل)] استخدم لتحضير ثلاث معقدات جديدة من ايونات (الكوبلت، النيكل والنحاس) ثنائية التكافؤ. شخص ليكاند الازو الجديد بواسطة قياسات التحليل الدقيق للعناصر (كاربون، هيدروجين، نيتروجين و اوكسجين)، واسلوب الأشعة فوق البنفسجية والأشعة تحت الحمراء وتحليل الرنين النووي المغناطيسي البروتوني و كاربون ثلاثة عشر وتحليل طيف الكتلة. اثبتت النتائج ان ليكاند كان ثلاثي السن مانح من نوع (نيتروجين نيتروجين اوكسجين). تم تحضير المعقدات بعد تحديد أفضل تركيز ودالة حامضية. تم دراسة أطراف الأشعة فوق البنفسجية-المرئية لمحاليل المعقدات عند مدى من الدوال الحامضية (5-9) وتراكيز مولارية (1×10^{-4} - 5×10^{-4}) والتي توافقت مع قانون لامبيرت-بير. تم استنتاج تكافؤ المعقدات طبقا الى علاقة النسبة المولية التي تم الحصول عليها من الدراسات الطيفية لمحاليل المعقدات، حيث كانت النسبة (2:1) فلز: ليكاند لجميع المعقدات المحضرة. شخصت معقدات الايونات الفلزية بواسطة اطراف الاشعة تحت الحمراء، واطراف الاشعة فوق البنفسجية-المرئية، التوصيلية المولارية، الامتصاص الذري، الحساسية المغناطيسية و تقنية التحليل الدقيق للعناصر. من نتائج التقنيات الفيزيائية والكيميائية والطيفية، تم اقتراح شكل هندسي ثماني السطح لهذه المعقدات الفلزية. تم تقييم فعالية هذه المركبات ضد نوعين من البكتيريا المسببة للأمراض البشرية المكورات العنقودية (موجبة الجرام) والإشريكية القولونية (سالبة لجرام).