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# SYNTHESIS OF INDOLE BORATE LIGAND WITH Ni(II) AND Cu(II) COMPLEXES

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**ABSTRACT**. A new borate derivative (2,2',2'',2'''-((ʎ$^4$-boranetetrayl)tetrakis(1H-indole-1,3-diyl))tetraacetic acid)(BIN) was synthesized from the reaction of indole-3-acetic acid (IAA) with sodium tetrahydroborate $NaBH_4$ at a 4:1 mole ratio. This ligand was characterized by $^1$HNMR, FTIR, electronic spectra (UV-Vis.) thermal analysis (TG and DTG), elemental analysis (CHN) and melting point. Nickel and copper complexes were also synthesized and characterized by CHN, UV-Vis, atomic absorption, FTIR, melting point, magnetic susceptibility and molar conductivity. Results of analyses were in agreement with suggested structures. The results showed that the structure of nickel complex was octahedral while the copper complex was square planar and both complexes were nonelectrolytes. Antimicrobial activity against four kinds of bacteria *Staphylococcus aureus* and *Streptococcus* ($G^+$), *E. coli* and *Pseudomonas aeruginosa* ($G^-$) as well as one fungus *Candida albican* exhibited that the copper complex has higher antimicrobial activity. Total antioxidant results showed that the BIN ligand has higher activity at 50 and 100 µg/mL while the copper complex has more antioxidant activity at 150 µg/mL.

**KEY WORDS**: Antimicrobial agents, Antioxidant agents, Borate derivative, Indole-3-acetic acid (IAA), Nickel and copper complexes

## INTRODUCTION

Polyborate ligands are a class of chelates compounds, have a wide spectrum of applications in various areas such as coordination, bioinorganic and organometallic chemistry. General characteristics of these compounds are ease of synthesis, formation of stability chelated cage and bridgehead boron compounds when coordinated with metal centers [1]. The poly(pyrazolyl)borate or poly(imidazolyl )borate ligands are well known and have a wide range of applications. Bis, tris and tetrakis(pyrazolyl)borate ligands were prepared by increasing the pyrazole ring and studied their ability of complexation [2]. Three ligands were prepared by using pyrazolyl and 3,5-dimethylpyrazolyl, then investigated their interactions with $Fe(BF_4)_2$, $Cu(BF_4)_2$, and $Ag(BF_4)$ [3]. The substituted imidazolium poly(azolyl)borate $[R_1R_2im][B(H)4\text{-}n(azolyl)n]$ ($R_1$ = n-butyl, methyl, 2-(diethylamino)ethyl; $R_2$ = methyl; 1,2,4-triazolyl, tetrazolyl azolyl = pyrazolyl, imidazolyl; n = 2, 3) have been synthesized [4]. N-(Alkyl/aryl)imidazolium-borates were prepared with imidazolium-trihydridoborate adducts [5]. This kind of ligand has multi coordinate arms, this leads to ease and flexibility of coordination with metal ions. The binding with metal complexes and biomolecules is affected by the nature of the ligands [6]. Also these ligands form bridges which is a necessary requirement to interaction between the metal center and boron and this gives these compounds importance in potential catalytic applications [7]. Indole is a bicyclic molecule containing a benzene ring fused with a pyrrole ring in position 2 and 3. Indole regraded an electron-rich aromatic compound with characteristic binding properties and this is due to the presence of pyrrole ring, also the Indole is regarded as a biomolecules and natural product. The importance of indole is attributed to its existence in a wide field of pharmacological molecules. Some indoles have anti-tumor, antiprotozoal, monooxime inhibition, antibacterial and anti-tubercular activity. Research reported the effect of N-H indole in the interaction with specific receptors [8]. The heterocyclic compounds of indoles have ability to improve the solubility and

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*Corresponding authors. E-mail: asmaa.m@sc.uobaghdad.edu.iq bioavailability via complexation with metal ions. The coordination behavior of indole compounds led to the publication of many research papers in diverse style of coordination indole with metal ions.

One of the most important indole derivatives is indole-3-acetic acid (IAA) a plant hormone to regulate growth, it is a type of auxin, contains an indole ring with carboxy methyl substituent [9]. The anti-inflammatory role of IAA may be associated with its anti-oxidative activity [10]. IAA has been used in human health applications, metabolites obtained from a variety of body parts and fluids, including urine, feces, blood, saliva and cerebrospinal fluid. Recent studied suggest that IAA can be used as a scavenger of free radicals, inhibit oxidative stress, and mitigate pro-inflammatory cytokine production [11]. The coordination chemistry of IAA was extensively studied with applications [12-14].

In the present work, a new poly indolyl borate ligand (2,2',2'',2'''-(($\lambda^4$-boranetetrayl) tetrakis(1H-indole-1,3-diyl))tetraacetic acid)(BIN) was prepared from the reaction of (IAA) with sodium tetrahydroborate $NaBH_4$ at a 4:1 mole ratio. Nickel and copper complexes were also prepared at a 2:1 mole ratio (L:M). The new ligand and its complexes were characterized by spectral and physical methods. The antimicrobial and antioxidant activities of the synthesized compounds were evaluated.

## EXPERIMENTAL

### *Chemicals and reagents*

Indole-3-acetic acid ($C_{10}H_9O_2N$, 99%, Merck), toluene ($C_6H_5CH_3$, 99%, Gold Label), sodium borohydride ($NaBH_4$, 98%, Fieser), nickel chloride hexahydrate ($NiCl_2.6H_2O$, 99.9%, Sigma-Aldrich), copper chloride ($CuCl_2$, 99%, Merck), and dimethyl sulfoxide DMSO ($(CH_3)_2SO$, 99.9%, Gold Label) were used without further purification.

### *Instrumentation*

$^1H$ NMR was carried out by using Bruker UltraShield 300MHz NMR spectrophotometer. FTIR spectroscopy to analyze the structures and functional groups with Shimadzu FTIR-8400S, Fourier Transform Infrared spectrophotometer using KBr and CsI discs at (4000 - 400 cm$^{-1}$) and (4000-250 cm$^{-1}$), respectively. Ultra violet spectra in DMSO were acquired in the range 200-1100 nm using Shimadzu 1800-UV spectrophotometer. Thermal analyses (TG and DTG) were obtained under Helium gas by using METTLER TA 4000 SYSTEM. Elemental analysis CHN was carried out by using Elemental Analyzer EuroEA 3000. Melting points were determined using the Gallenkamp melting point apparatus. The conductivity of complexes at (10$^{-3}$ M) in DMSO was determined by WTW Cond 7300 digital conductivity meter. The metal content was determined by atomic absorption spectroscopy using Nova350 spectrophotometer.

### *Synthesis of (2,2',2'',2'''-((λ-boranetetrayl)tetrakis(1H-indole-1,3-diyl))tetraacetic acid) (BIN) ligand*

The solution of indole-3-acetic acid (IAA) (09262 g, 0.0052 mol) in 10 mL toluene was heated in a round flask, a solution of $NaBH_4$ (0.05 g, 0.0013 mol) in 6 mL toluene was added and the mixture was heated under reflux at 110 °C for 15 hours. The color was changed from yellow to green, filtered and washed with toluene, recrystallized with (an acetone:$H_2O$ 5:1) mixture and washed with petroleum ether and dried to get a light green product (Figure 1a).

### *Synthesis of nickel(II) and copper(II) complexes*

The solutions of $NiCl_2.6H_2O$ (0.0325 g, 0.00013 mol) and $CuCl_2$ (0.0184 g, 0.00013 mol) in 5 mL ethanol was added to a solution of BIN ligand (0.2 g, 0.00027 mol) in 5 mL ethanol, the mixture was heated under reflux at 78 °C for 4 hours. The color changed from green to brown, filter the brown products and wash with ethanol followed by petroleum ether and dry (Figure 1b and 1c).

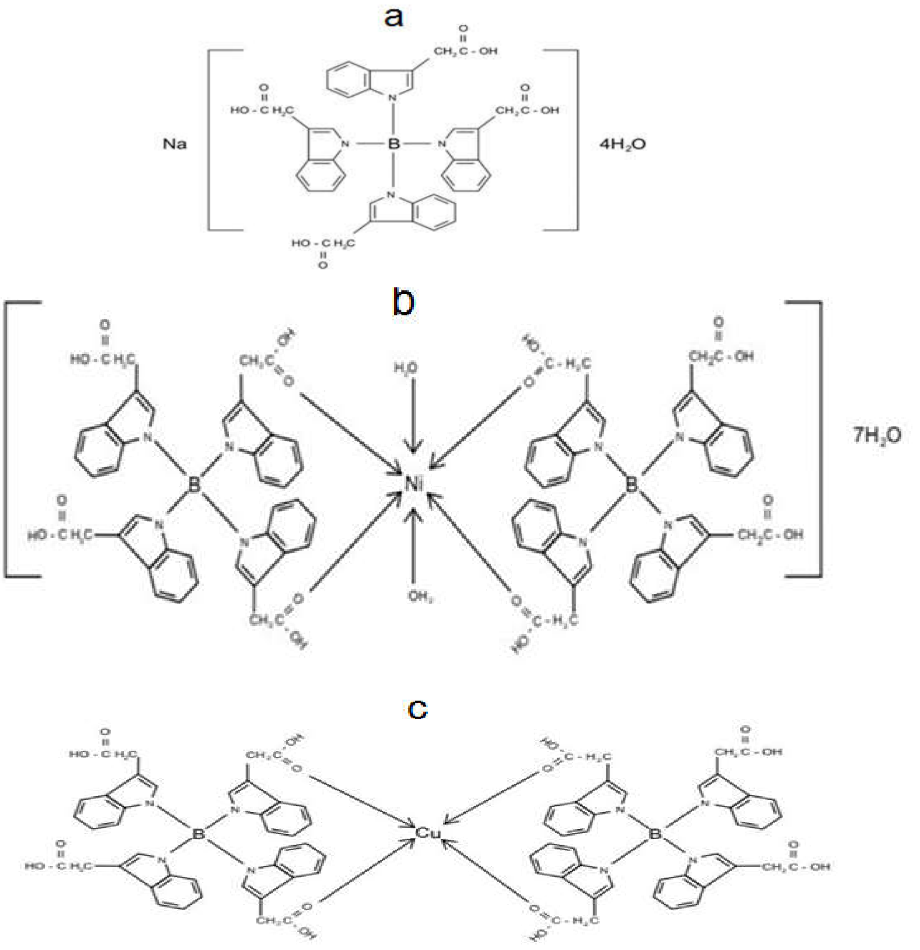

Figure 1. Suggested structures of a- BIN ligand, b- nickel complex, and c- copper complex.

## RESULTS AND DISCUSSION

The elemental analysis, physical properties and metal content percent data are illustrated in Table 1.

### *FTIR spectroscopy of the ligand and its complexes*

The FTIR spectrum of the ligand (Figure 2) showed a band at 3390 cm$^{-1}$ which was attributed to stretching vibration of lattice $H_2O$ , this band also appeared in the nickel complex at 3300 cm$^{-1}$ as well as the coordinated $H_2O$ showed stretching vibration at 3380 cm$^{-1}$, in addition to $p_r$ at 750 cm$^{-1}$ and $p_w$ at 680 cm$^{-1}$ [15]. The stretching vibration of the OH group in carboxylic moiety appeared at 3549 cm$^{-1}$ [16, 17], this band did not change in complexes. The band appeared at 1701 cm$^{-1}$ in the spectrum of the ligand which assigned to stretching vibration of carboxylic C=O group [16, 18], this band changed in complexes (1720 and 1712 cm$^{-1}$ for Ni and Cu complexes, respectively) due to coordination of C=O group with metal ions through the oxygen atom. The band of $\upsilon_{\mathbf{B-N}}$ appeared at 1458 cm$^{-1}$ [19], no change was observed in this band when compared with ligand spectrum (1450 and 1456 cm$^{-1}$ for Ni and Cu complexes respectively) indicating that it did not enter into the coordination. Low frequency bands for υ M-O appeared at 440 cm$^{-1}$ and 485 cm$^{-1}$ in the spectra of Ni and Cu complexes, respectively [20].

Table 1. The physical properties and elemental analysis of BIN ligand with its nickel and copper complexes.

| Compounds | M.Wt. (g/mol) | Color | Yield % | m.p. (°C) | Calculated % (found) % | | | M% calculated (found) |
|---|---|---|---|---|---|---|---|---|
| | | | | | C% | H% | N% | |
| Ligand (BIN) | 802.00 | Greenish gray | 80.46 | 152-154 | 59.85 (60.52) | 4.98 (5.26) | 6.98 (6.81) | - |
| $C_1$(Ni) | 1634.29 | Brown | 41.82 | > 280 | 58.74 (58.77) | 5.01 (5.18) | 6.85 (6.03) | 3.59 (3.74) |
| $C_2$(Cu) | 1477.10 | Brown | 71.05 | > 280 | 64.99 (65.81) | 4.33 (4.89) | 7.58 (7.01) | 4.30 (3.69) |

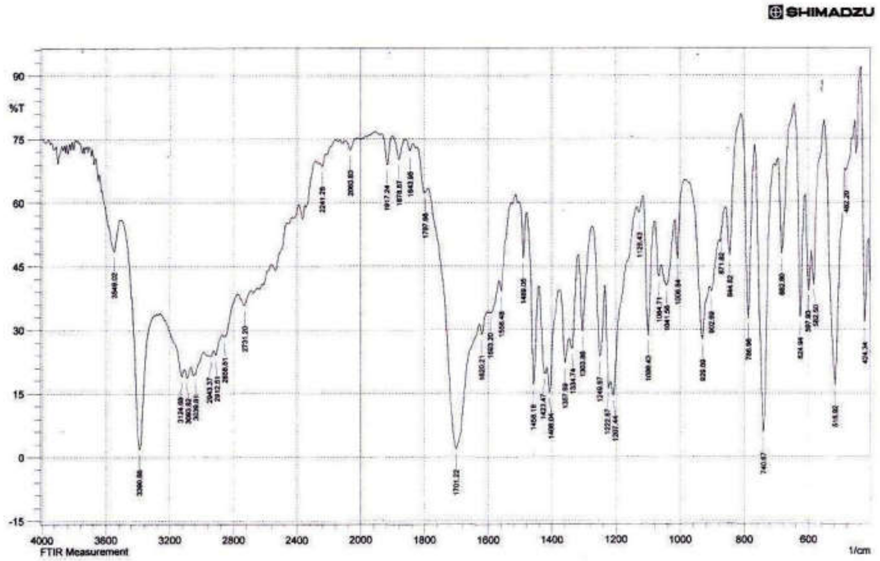

Figure 2. FTIR spectrum of BIN ligand.

*Thermogravimetric analysis TGA and DTG*

Thermal analysis TG and DTG of the ligand (Figure 3) were carried out in the temperature range 25-900°C at the rate 20°C/min. under helium as inert gas. Thermal stability of the ligand was studied through the remaining part of ligand (residue) after thermal degradation which was 17.58% at 830 °C and this indicates that the ligand has good stability. High agreement between found and calculated mass loss percent indicates the validity of the ligand proposed structure. (Table 2) shows steps of the thermal degradation with suggested assignment, decompose temperature and mass loss %.

Table 2. Thermogravimetric data (TG and DTG) of BIN ligand.

| Formula and M.Wt. of ligand | Steps | Temp.of decom. °C (DTG) | Suggested assignment | Mass loss% calculated (found) |
|---|---|---|---|---|
| $Na[C_{40}H_{32}N_4O_8B]4H_2O$<br>802 g/mol | 1 | 77.21 | 0.5 $H_2O$ | 1.12 (0.09) |
| | 2 | 223.56 | 2.5 $H_2O$ | 5.61 (5.58) |
| | 3 | 339.95 | $H_2O+Na+C_8H_{12}O_8+C_{18}H_{10}N_2$ | 66.21 (66.97) |
| | 4<br>Residue | 636.11<br>830.00 | $C_6H_4$<br>$BN+C_8H_5N$ | 9.47 (9.76)<br>17.43 (17.58) |

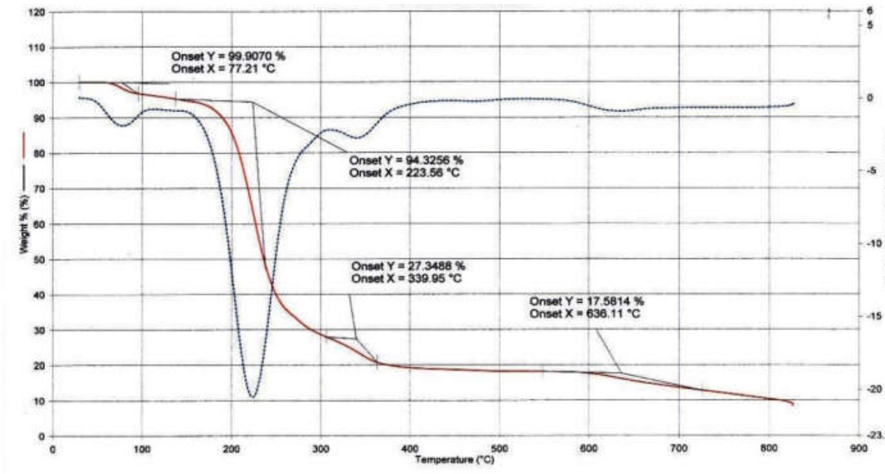

Figure 3. Thermogram of BIN ligand.

### *$^1$H NMR spectroscopy*

The $^1$H NMR technique is useful for studying the content of protons. $^1$H NMR analysis of the ligand (BIN) was carried out in $d_6$-DMSO as a solvent. (Figure 4) showed a peak at δ = 2.50 ppm due to the chemical shift of DMSO [21]. A high intensity peak appeared at range δ = 3.42-3.83 ppm which was assigned to protons of $H_2O$ and $CH_2$ groups [14, 21]. Aromatic protons appeared as multiple peaks at range δ = 6.96-7.52 ppm [14]. The δ = 8.43 ppm refers to the chemical shift of NCH protons [22]. Finally the peak appeared at low field δ = 10.88 ppm due to the chemical shift of the acid OH proton [23].The $^1$H NMR results showed that the total protons of the ligand were 40 protons and this agreement with the suggested structure of BIN ligand.

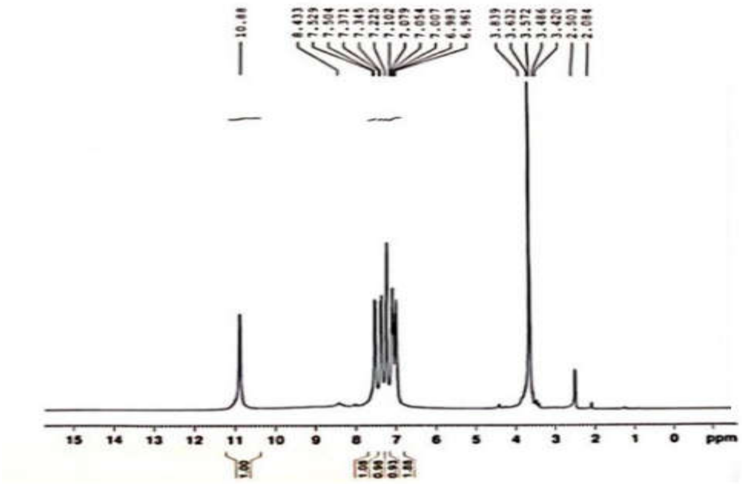

Figure 4. $^1$H NMR spectrum of BIN ligand.

*Electronic spectra*

The UV-Vis spectra of the ligand and its complexes were recorded in DMSO as solvent. The spectrum of BIN ligand showed two peaks at 282 and 338 nm (35460 and 29585 cm$^{-1}$, respectively) which refer to $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ transitions as a result of the presence of the aromatic system with donor atoms, these peaks exhibited a frequency shift in the spectra of nickel and copper complexes because of the coordination of ligand with metal ions. The spectrum of Ni complex showed two bands at 974 and 654 nm (10266 and 15290 cm$^{-1}$, respectively) which are assigned to $^3A_2g \rightarrow {}^3T_2g$ and $^3A_2g \rightarrow {}^3T_1g(F)$ transitions, respectively (Table 3), these bands represent $\upsilon_1$ and $\upsilon_2$ transitions for octahedral geometry [20, 24]. The spectrum of the copper complex exhibited two bands at 810 and 700 nm (12345 and 14285 cm$^{-1}$, respectively) which refer to $^2B_1g \rightarrow {}^2A_1g$ and $^2B_1g \rightarrow {}^2B_2g$ transitions for square planar Cu(II) complex [20].Table 3 illustrates the suggested structures of Ni and Cu complexes.

Table 3. Electronic transitions of BIN ligand and its metal complexes.

| Compounds | Band positions nm (cm$^{-1}$) | Assignment | Molar conductivity (S cm$^2$ mol$^{-1}$) in DMSO | μeff. (B.M.) | Suggested geometry |
|---|---|---|---|---|---|
| Ligand (BIN) | 282(35460)<br>338(29585) | $\pi - \pi *$<br>$n - \pi *$ | - | - | - |
| $C_1$(Ni) | 293(34129)<br>340(29411)<br>974(10266) $\upsilon_1$<br>654(15290) $\upsilon_2$ | Intra ligand<br>$^3A_2g \rightarrow {}^3T_2g$<br>$^3A_2g \rightarrow {}^3T_1g(F)$ | 23 | 3.76 | Octahedral |
| $C_2$(Cu) | 296(33783)<br>317(31545)<br>810(12345)<br>700(14285) | Intra ligand<br>$^2B_1g \rightarrow {}^2A_1g$<br>$^2B_1g \rightarrow {}^2B_2g$ | 39 | 1.92 | Square planar |

*Antimicrobial activity*

The antimicrobial activity was evaluated by disc diffusion method at $10^{-3}$ M in DMSO of BIN ligand and its Ni and Cu complexes against four kinds of bacteria *Staphylococcus aureus* and *Streptococcus* ($G^+$) , *E. coli* and *Pseudomonas aeruginosa* ($G^-$ ) as well as one fungus *Candida albican*. The synthesized compounds showed various activities (Table 4). Copper complex has higher inhibition of tested microbial strains except *Pseudomonas aeruginosa* and the order of activity is Cu = Ni > BIN (*Staph.*), Cu > (Ni, BIN) (*Strep.*), Cu > BIN > Ni (*E. coli*), BIN > (Ni, Cu) (*Pseud.*) , Cu > Ni > BIN (*Cand.*).

Table 4. Inhibition zone of BIN ligand and its complexes.

| Gram positive | | | | | |
|---|---|---|---|---|---|
| Bacteria | Sample No. | Size (mm) | Bacteria | Sample No. | Size mm |
| *Staphylococcus aureus* | BIN ligand | -- | *Streptococcus* | BIN ligand | -- |
| | Ni($C_1$) | 13 | | Ni($C_1$) | -- |
| | Cu($C_2$) | 13 | | Cu($C_2$) | 12 |
| Gram negative | | | | | |
| Bacteria | Sample No. | Size mm | Bacteria | Sample No. | Size mm |
| *E. coli* | BIN ligand | 12 | *Pseudomonas aeruginosa* | BIN ligand | 12 |
| | Ni($C_1$) | 11 | | Ni($C_1$) | 11 |
| | Cu($C_2$) | 13 | | Cu($C_2$) | 11 |
| Fungi | | | | | |
| *Candida albican* | | | | | |
| Sample No. | Size mm | | Sample No. | Size mm | |
| BIN ligand | 11 | | Cu($C_2$) | 13 | |
| Ni($C_1$) | 12 | | -- | -- | |

*Antioxidant activity*

*Phosphomolybdate assay (total antioxidant capacity)*

The antioxidant database is an essential research tool to explain the health effects of chemical antioxidants. The phosphomolybdate assay is based on reducing the phosphate Mo(VI) to phosphate Mo(V) by the sample and forming a bluish-green colored phosphate Mo(V) complex at acid pH [25]. Total antioxidant capacity of the ligand and its complexes at three concentrations 50, 100 and 150 µg/mL was studied by the phosphomolybdate method with ascorbic acid as a standard . The absorbance was measured at 765 nm against a blank [26]. The antioxidant capacity was estimated by using the following equation:

$$\text{Antioxidant effect (\%)} = \left[\frac{\text{control absorbance} - \text{sample absorbance}}{\text{control absorbance}}\right] \times 100 \qquad (1)$$

All synthesized compounds have good antioxidant activities spatially at 150 µg/mL (Figure 5) where the antioxidant activities are greater than ascorbic acid as a control while other concentrations have varying effects (Table 5).

Table 5. Total antioxidant data of BIN ligand and its complexes.

| Sample | 150 µg/mL | | 100 µg/mL | | 50 µg/mL | |
|---|---|---|---|---|---|---|
| | Absorbance | Total antioxidant | Absorbance | Total antioxidant | Absorbance | Total antioxidant |
| Blank | 0.004 ± 0.002 | | | | | |
| Control | 0.133 ± 0.002 | 20.11 | 0.124 ± 0.002 | 18.75 | 0.144 ± 0.002 | 17.31 |
| BIN ligand | 0.690 ± 0.002 | 104.57 | 0.450 ± 0.005 | 67.96 | 0.307 ± 0.003 | 46.55 |
| Ni complex | 0.179 ± 0.002 | 27.04 | 0.098 ± 0.003 | 14.85 | 0.053 ± 0.002 | 8.07 |
| Cu complex | 0.960 ± 0.008 | 145.48 | 0.194 ± 0.003 | 29.35 | 0.085 ± 0.003 | 12.80 |

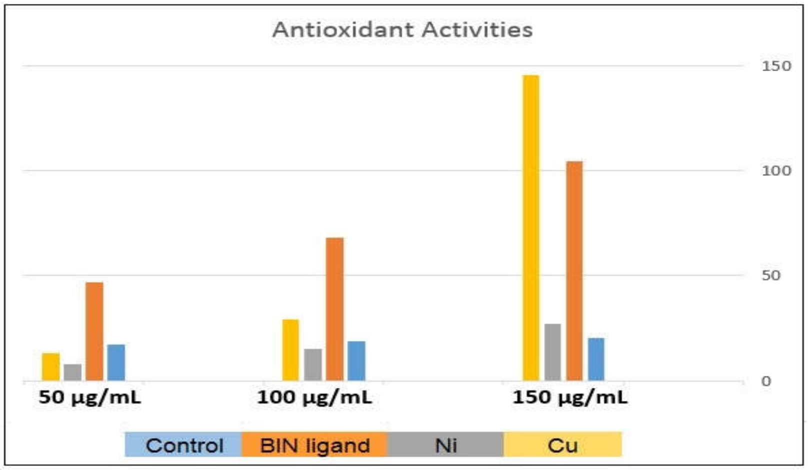

Figure 5. Antioxidant effects of ascorbic acid (control), BIN ligand and its Ni(II) and Cu(II) complexes.

## CONCLUSION

A new borate ligand (BIN) and its metal complexes have been synthesized and the structures were confirmed through different characterizations with high agreement between the found and calculated results of analyses. The geometry of Ni and Cu complexes was octahedral and square planar, respectively, both complexes exhibited nonelectrolyte behavior through measurement of molar conductivity. Antimicrobial activity was evaluated for BIN ligand and its complexes against $G^+$ and $G^-$ bacteria as well as against fungi, the results showed that all synthesized compounds possess good activity as antimicrobial. Also new compounds tested as antioxidant materials in three concentrations where the results showed the following activity orders: at 50 µg/mL was BIN ligand > control > Cu complex > Ni complex, at 100 µg/mL was BIN ligand > Cu complex > control > Ni complex and at 150 µg/mL was Cu complex > BIN ligand > Ni complex > control.

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