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Novel bioactive Ni (II)-Isatin-bishydrazone complexes; Synthesis, characterization, antibacterial and antifungal activity investigation

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ABSTRACT

On a strategy to create new complexes based on bioactive organic ligands can be used as antifungal and antibacterial reagents, we synthesized new Ni(II) complexes based on bioactive isatin-bishydrazone compounds. The Ni(II) complexes characterized on the bases of elemental analysis, molar conductance, IR, electronic spectra, magnetic measurements and thermal analysis (TGA & DTA). Moreover, the stiochiometry of the synthesized complexes have been determined spectrophotometrically. The results suggested that Isatin-bishydrazone ligands exhibited neutral tridentate ligand with ONN sites coordinating to the metal ion via Isatin-carbonyl (C=O), azomethine-Nitrogen (-CR=N) and pyridine-Nitrogen (C=N) forming the general formula $[Ni(L)\ H_2O]Cl_2.2H_2O$, (where L = neutral tridentate Isatin-hydrazone ligand). The bioefficacy of the ligands and their complexes have been examined for their in-vitro antibacterial and antifungal activity against many types of bacteria and anti fungal cultures which are common contaminants of the environment in Egypt and some of which are involved in human and animal diseases or in plant diseases or frequently reported from contaminated soil, water and food substances. The results of these studies indicate that the ligands and their metal complexes possess notable antimicrobial properties.

Keywords: Isatin-bishydrazone ligands, Ni(II) complexes, antibacterial, antifungal, Thermal analysis.

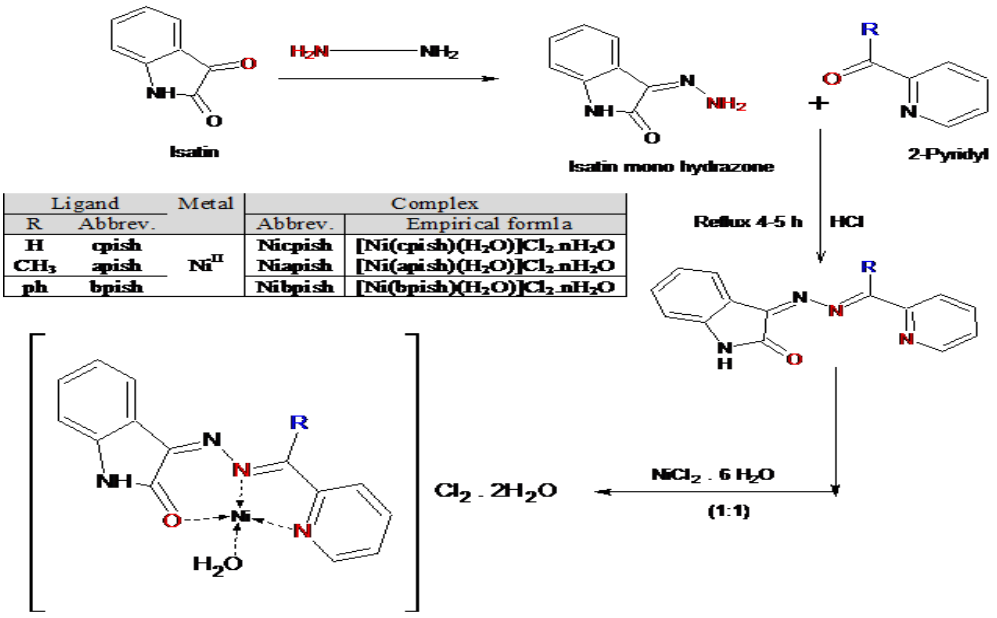
1. INTRODUCTION

Hydrazones form an interesting class of chelating ligands which contain an azomethine group linked to a nitrogen atom and find extensive application in various fields [1,2], and also form an important class of organic compounds with a wide variety of biological properties [3-6]. Recently, the development of new chemotherapeutic hydrazones is attracting the attention of medicinal chemists [7]. Many studies reported the biological activities of hydrazones, including their anticancer [8], antibacterial [9], antifungal, and herbicidal activities [10,11].

Isatin (1-H-indole-2, 3-dione) and its derivatives possess a broad range of biological and pharmacological properties [12-16] and are widely used as starting materials for the synthesis of broad range of heterocyclic compounds and as substrates for drug synthesis [17]. Variety of biological activities are associated with isatin-hydrazones including the central nervous system (CNS) activities as potentiation of pentobarbitone induce nercosis [18], Analgesic [19], anticonvulsant [20], antidepressant [21], anti-inflammatory [22], and antimicrobial [23] , moreover isatins are capable of crossing the blood–brain-barrier [24]. Although metal complexes of monohydrazones derived from isatin have been extensively investigated, those formed from bishydrazones received comparatively less attention [25,26].

On the other hand, hydrazones of 2-pyridyl derivatives (i.e. 2-acetyl pyridine, 2-benzoyl pyridine and pyridine-2-carboxaldhyde) and their metal complexes have good biological applications such as antimicrobial [27,28], anti-inflammatory [29-31] and anticancer [32-34] reagents.

While much attention has been directed to study the metal complexes derived from isatin-hydrazones [35,36], however, no investigations have appeared in the literature to describe the metal complexes of the hydrazones derived from isatin-monohydrazone and 2-pyridyl derivatives. Therefore the need to design new isatin-bishydrazone derivatives for emerging drug targets is an active area of medicinal chemistry.



Ligand		Metal	Complex	
R	Abbrev.		Abbrev.	Empirical formla
H	cpish	Ni^{II}	Nicpish	$[Ni(cpish)(H_2O)]Cl_2.nH_2O$
CH_3	apish		Niapish	$[Ni(apish)(H_2O)]Cl_2.nH_2O$
ph	bpish		Nibpish	$[Ni(bpish)(H_2O)]Cl_2.nH_2O$

Scheme - 1: Schematic diagram of the Isatin-bishydrazone ligands and their Ni(II) complexes.

In our previous work [37] we synthesized and characterized new isatin-bishydrazone compounds derived from isatin monohydrazone and 2-pyridyl which showing notable biological activity. Thus in the present work, we aim to synthesis new Ni(II) complexes with the synthesized isatin-bishydrazone ligands, characterization by different physicochemical methods. Also, spectrophotometric determination of the complex stoichiometry. Furthermore, Testing the in-vitro antibacterial and antifungal activity of the free ligands and their Ni(II) complexes against many types of bacteria and anti fungal cultures, which are common contaminants of the environment in Egypt and some of which are involved in human and animal diseases, plant diseases or frequently reported from contaminated soil, water and food substances in order to assess the antimicrobial activity of the synthesized compounds.

The structures of the prepared ligands and their Ni(II) complexes in this investigation are shown in scheme 1.

2. Experimental

2.1. Reagents

All chemicals were used as produced without further purification. Isatin, 2-acetyl pyridine, 2-benzoyl pyridine and pyridine-2-carboxaldhyde were obtained from Sigma-Aldrich Company Ltd. Hydrazine hydrate and hydrated Nickel chloride ($NiCl_2.6H_2O$) was obtained from BDH Company. All other reagents and solvents (methanol, ethanol and DMF) were purchased from commercial sources and were of analytical grade.

2.2. Synthesis of the ligands and their complexes.

2.2.1. Synthesis of the ligands.

Isatin-bishydrazone ligands, namely [(pyridine-2-Carboxaldhyde)-3-isatin]-bishydrazone (cpish), [(2-acetyl pyridine)-3-isatin]-bishydrazone (apish) and [(2-benzoyl pyridine)-3-isatin]-bishydrazone (bpish) were prepared as described before [37] in two steps: the first step is the synthesis of Isatin-monohydrazone, followed by condensation with the 2-pyridyl, giving the Isatin-bishydrazone Ligands.

2.2.1.1. Synthesis of Isatin-monohydrazone.

Isatin (1.47 g, 10 mmol) was dissolved in methanol (40 mL) and was added to a solution of hydrazine hydrate (0.05 g, 10 mmol) dissolved in hot methanol (5 mL). The resulting mixture was refluxed for 3 h on a water-bath. On cooling, the yellow compound that formed was filtered, washed with cold methanol, dried and recrystallized from methanol [38].

2.2.1.2. Synthesis of Isatin-bishydrazone ligands.

A (1.0 mmol of 2-pyridyl (2-acetyl pyridine or 2-benzoyl pyridine or Pyridine-2-carboxaldhyde) was added drop wise to a hot methanolic solution of Isatin-monohydrazone (1.0 mmol) then the resulting mixture was refluxed for 1h with constant stirring, then 2-3 drops of glacial acetic acid was added, with continue refluxing for 4h under constant magnetic stirring. On cooling, the formed ligand was filtered, washed with cold methanol, dried and finally recrystallized from methanol [37].

2.2.2. Synthesis of Ni (II) complexes

The solid Ni(II)-complexes of Isatin-bishydrazone ligands were prepared as the following; a solution of the metal salt in minimum amount of water [$NiCl_2.6H_2O$, 1.0 mmol] was added drop wise to a hot methanolic solution of the ligand [cpish or apish or bpish, 1.0 mmol]. The resulting mixture was refluxed at 70 ^{0}C for 10 h under constant stirring. By evaporation over night, the resulted solid product was filtered, washed with water-methanol solvent, drying, and finally recrystallized from water-methanol mixed solvent. The yield and melting point of each product were determined.

2.3. Analyses of the complexes

2.3.1. Physico-chemical measurements

The stoichiometric analysis (C, H and N) of the new compounds was performed using elemental analyzer Perkin-Elmer model 40c, at the Micro-analytical Centre at Cairo-University, Egypt. The molar conductance of 10^{-3} molar dilution was measured by JENWAY conductivity-meter model 4320 at 298 K. The IR spectra were recorded on Shimadzu FTIR model 8101 in the region 4000–400 cm^{-1} using dry KBr discs. The electronic spectra of the complexes in methanol were recorded in the region 200–800 nm using a 10 mm matched quartz cells on Jasco UV-Visible spectrophotometer model V-530. Rigaku model 8150 thermo-analyzer was used for simultaneous recording of TG–DTA curves at a heating rate of 10 min^{-1}.

2.3.2. Determination of the stoichiometry and formation constant of the complexes.

The stoichiometries of the prepared complexes were determined by applying the spectrophotometric molar ratio [39-41] and continuous variation [42,43] methods. All the spectrophotometric measurements were carried out in 10 mm cells in the thermostatted cell Jacket compartment of Jasco UV-Visible spectrophotometer (model V-530). The thermostatted cell holder was supplied by an ultrathermostate water circulator (CRIOTERM model 190) to control the temperature at 25ºC ± 0.1 ºC. Magnetic susceptibilities of the complexes were determined at room temperature by Gouy method using $Hg[Co(NCS)_4]$ as calibrant [44]. Consideration the magnetic contribution of various atoms and structural units [45,46]. The effective magnetic moments (in Bohr magneton) were calculated from the corrected susceptibilities using the equation: $\mu_{eff} = 2.828\ (xT)^{1/2}$, where x is the molar susceptibility (in cm^3/mole) and T is the absolute temperature.

2.4. In-vitro biological activity

2.4. 1. Antibacterial and anti-fungi screening

The antimicrobial activity of all the synthesized Isatin-bishydrazone ligands and their corresponding Ni(II) complexes were tested against 6 bacterial (three gram positive bacteria and three gram negative bacteria) and 6 fungal strains. These strains are common contaminants of the environment in Egypt and some of which are involved in human and animal diseases (Trichophyton rubrum, Candida albicans, Geotrichum candidum, Scopulariopsis brevicaulis, Aspergillus flavus and Staphylococcus aureus (+ve)) or in plant diseases (Fusarium oxysporum) or frequently reported from contaminated soil, water and food substances (Escherichia coli (-ve), Bacillus cereus (+ve), Pseudomonas aeruginosa (-ve), Serratia marcescens (-ve) and Micrococcus luteus (+ve)). To prepare inocula for bioassay, bacterial strains were individually cultured for 48h in 100 ml conical flasks containing 30 ml nutrient broth medium. Fungi were grown for 7 days in 100 ml conicals containing 30 ml Sabouraud's dextrose broth. Bioassay was done in 10 cm sterile plastic Petri plates in which microbial suspension (1ml/plate) and 15 ml appropriate agar medium (15 ml/plate) were poured. Nutrient agar and Sabouraud's dextrose agar were respectively used for bacteria and fungi. After solidification of the media, 5 mm diameter cavities were cut in the solidified agar (3 cavities/plate) using sterile cork borer. Chemical compounds dissolved in dimethyl sulfuxide (DMSO) at 100 ppm were pipetted in the cavities (20 ul /cavity). Cultures were then incubated at 28 °C for 48 h in case of bacteria and up to 7 days in case of fungi. Results were read as the diameter (in mm) of inhibition zone around cavities [47].

2.4.2. Determination of minimum inhibitory concentration (MIC) value.

To determine the minimum inhibitory concentrations (MICs), chemical compounds giving positive results were diluted with DMSO to prepare a series of descending concentrations down to 5 ppm. Diluted chemicals were similarly assayed as mentioned before and the least concentration (below which no activity) was recorded as the MIC.

2.4.3. Determination of the Activity Index (%) for the complexes.

The antibacterial and antifungal activity of a common standard antibiotic Chloramphenicol as antibacterial standard and Clotrimazole as antifungal standard were also recorded maintaining the same protocol as above at the same concentrations and solvent. The antibacterial and antifungal results of the compounds were compared with the standard and

% Activity Index for the complexes was calculated by using the formula as under [48]:

$$\%\text{Activity index} = \frac{\text{Zone of inhibition by test compound(diamter)}}{\text{Zone of inhibition by standard(diamter)}} X100$$

3. Results and discussion

3.1. Identification of the prepared compounds.

3.1.1. Microanalysis and molar Conductance Measurements

The results of the microanalysis of the prepared Isatin-bishydrazone ligands and their Ni(II) complexes in addition to the molar conductance measurements, table 1 suggested that: the subject ligands act as neutral tridentate and form complexes in 1:1 molar ratio (metal : ligand), with 1:2 electrolytic nature of all the complexes[48,50]. Thus, the general formula of the prepared complexes is suggested to be $[Ni(L)(H_2O)]Cl_2 .2H_2O$ according to the following reaction:

$$NiCl_2.nH_2O + L \longrightarrow [Ni(L)(H_2O)]Cl_2.nH_2O$$

Where L is the Isatin-bishydrazone ligand. The satisfactory results of analytical data table 1 and spectral studies revealed that the ligands and their complexes were of good purity.

Table - 1: Analytical and physical data of ligands and their complexes.

	Empirical Formula (Formula Weight)	color	m.p (°C)	yield (%)	Elemental Analysis found (calculated)					μ_v ($\Omega^{-1}.Cm^2.mol^{-1}$)		
					C %	H %	N %	Cl %	M %	Methanol	DMF	Ethanol
cpish	$C_{14}H_{10}N_4O$ (250.255)	red	245 °C	89 %	67.19 (67.20)	4.03 (4.33)	22.39 (22.30)	-------	-------	-------	------	-------
Ni-cpish complex	$[Ni(cpish)H_2O]Cl_2.2H_2O$ (433.89)	Dark brown	> 300	65 %	38.84 (38.75)	3.49 (3.72)	12.94 (12.91)	16.38 (16.34)	13.56 (13.53)	170	140	85
apish	$C_{15}H_{12}N_4O$ (264.28)	red	250 °C	85 %	68.17 (68.50)	4.58 (4.25)	21.20 (20.98)	-------	-------	-------	------	-------
Ni-cpish complex	$[Ni(apish)H_2O]Cl_2.2H_2O$ (447.93)	Dark brown	> 300	60 %	40.31 (40.22)	3.83 (4.05)	12.54 (12.51)	15.87 (15.83)	13.13 (13.10)	166	137	82
bpish	$C_{20}H_{14}N_4O$ (326.35)	red	265 °C	85 %	73.61 (73.55)	4.32 (4.28)	17.17 (17.05)	-------	-------	-------	------	-------
Ni-cpish complex	$[Ni(bpish)H_2O]Cl_2.2H_2O$ (509.99)	Dark brown	> 300	70 %	47.29 (47.10)	3.57 (3.95)	11.03 (10.99)	13.96 (13.90)	11.55 (11.51)	160	133	80

m.p; melting point, μ_v; molar conductance

3.1.2. Spectroscopic studies

3.1.2.1. I.R. spectra and mode of bonding

In the absence of a powerful technique such as single X-ray crystallography, infrared spectra has proven to be the most suitable technique to give enough information to elucidate the nature of bonding of the subject ligands to the metal ion. Thus a detailed interpretation of IR spectra of the free ligands and the effect of binding of Ni(II) of the vibration frequencies of the free ligands is discussed to determine the coordination sites which may involve in chelation. The significant infrared bands of the subject ligands and their metal complexes are given in table 2. The observed bands may be classified into those originating from the ligands and those arising from the bonds formed between metal ions and the coordinating sites. The IR spectrum of the Isatin-bishydrazone ligands exhibited Characteristic band due to (-NH) and lactonyl carbon ν(C=O) at ≈ 3180-3200 cm^{-1} and ≈ 1722 cm^{-1}[51] respectively. In addition, the strong band at ≈ 1460 cm^{-1} and a characteristic high intensity band at ≈ 1621 cm^{-1} in the IR spectrum of the Isatin-bishydrazone ligands are assigned to ν(C=N) and ν(HC=N) respectively. In comparison with the spectra of the Isatin-bishydrazone ligands, all the Ni(II) complexes exhibited the band of ν(HC=N) in the region ≈ 1590 cm^{-1}; showing the shift of the band to lower wave numbers indicating that, the azomethine nitrogen is coordinated to the metal ion[52,53]. The band of ν(C=O) in the region 1670-1680 cm^{-1} in the metal complexes showing the shift to lower wave numbers confirms that, the carbonyl oxygen of Isatin is coordinated to the metal ion [54,55]. The unaltered position of a band due to ν(NH) and ν(C=N) in all the metal complexes indicates that, these groups are not involved in coordination. The new bands in the region of 500-510 and 630-650 cm^{-1} in the spectra of the complexes are assigned to stretching frequencies of (M-N) and (M-O) bonds respectively [56]. Thus the IR spectral results provide strong evidences for the complexation of Isatin-bishydrazone ligands with metal ion in tridentate mode via Isatin-carbonyl(C=O), azomethine-N(C=N) and pyridine-N (C=N).

Table - 2: The infrared absorption frequencies (cm^{-1}) and electronic spectra of the investigated Isatin-hydrazone ligands and it's metal complexes

no	Compound	υ (OH)[1]	υ (N-H)[2]	υ (C=O)[3]	υ (HC=N)[4]	υ (C=N)[5]	υ (M-O)	υ (M-N)	Electronic spectra λ_{max}, nm	ε_{max} ($dm^3.mol^{-1}.cm^{-1}$)	Assignment
1	Cpish	3421.2	3276.5	1721.7	1613.7	1460.3	.....	.....	237	1744.05	π - π *
									324	865.49	n - π *
2	Ni- cpish	3430.8	3287.1	1685.0	1611.7	1409.0	656.8	510.2	627 nm	320.12	d-d band
										293.02	d-d band
									676 nm		
3	Apish	3393.2	3180.0	1721.7	1618.5	1458.4	.....	.....	254	538.30	π - π *
									274	580.97	π - π *
									325	339.05	n - π *
4	Ni- apish	3357.5	3159.8	1672.5	1592.4	1463.2	637.6	504.4	625 nm	723.22	d-d band
										256.57	d-d band
									680 nm		
5	Bpish	3424.0	3181.0	1721.7	1607.9	1452.6	.....	.....	252	1096.47	π - π *
									271	882.56	π - π *
									327	653.87	n - π *
6	Ni- bpish	3359.4	3160.7	1671.5	1594.4	1463.2	636.6	503.5	636 nm	268.80	d-d band
										295.95	d-d band
									682 nm		

[1]. Vibrations of the water molecules, [2]. Vibrations of the Indol ring (N-H), [3]. Vibrations of the Lactonyl group (C=O), [4]. Vibrations of the azomethine group (C=N), [5]. Vibrations of group from the β-hydrazone of Isatin (C=N). λ_{max}; maximum absorbance wave Length , ε_{max} ; molar extension coefficient.

3.1.2.2. Electronic Spectra

Electronic spectra are a valuable tool for coordination chemists to draw important information about the structural aspects of the complexes. The ligands, which are organic compounds, have absorption bands in the ultraviolet region and in some cases these bands extend to higher wavelength region due to conjugation. Upon complexation with metal ions, changes will take place in the electronic properties of the system. New bands in the visible region due d-d absorption and charge transfer spectra from metal to ligand (M→L) or ligand to metal (L → M) can be observed and this data can be processed to obtain information regarding the structural and geometry of the complexes. The Electronic spectra of the Isatin-bishydrazone ligands and its Ni(II)-complexes were recorded in MeOH (≈ $1x10^{-3}$ mol/dm^3) in the range 200 – 800 nm at 298K. The absorption maxima bands are listed in table 2 and the spectra are given in Figure 1. The UV-Vis spectrum of the Isatin-bishydrazone ligand show important strong bands at ≈ 388 and ≈ 230 nm due to π - π * and n - π * transitions[57] respectively. These bands are altered to a greater extent on complexation. The spectrum of Ni(II)-complexes shows characteristic bands at at λ_{max} ≈ 220-250 nm, λ_{max} ≈ 320-400 nm and abroad band at λ_{max} ≈ 670-680 nm due to Intra-ligand band, Ligand-Metal Charge Transfer (LMCT) and d-d transition band respectively.

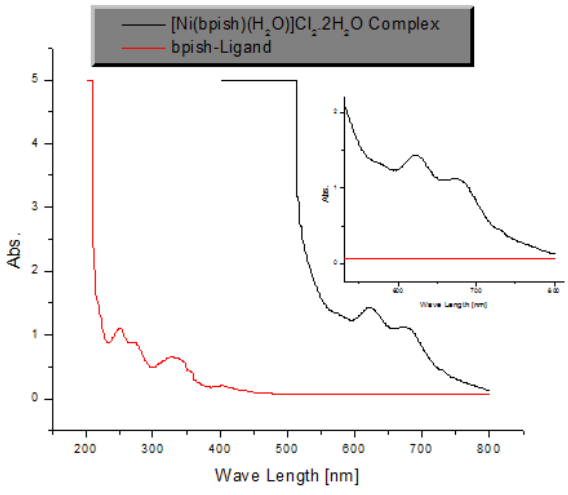


Figure - 1: UV VIS Spectrum of the Ni(II) Isatin-hydrazone Complexes.

3.2. Stoichiometry and formation constant of the complexes.

3.2.1. Determination of the stoichiometry of the complexes.

The stoichiometry of the various Ni(II) Isatin-bishydrazone complexes was determined by applying the spectrophotometric molar ratio[58-60] and continuous variation[61-62] methods, which suggested the possible formation of 1:1 complexes. The curves of continuous variation method (Figure 2) displayed maximum absorbance at mole fraction X ligand ≈ 0.5, indicating, the formation of the complex between the metal ion and the ligands in 1:1 (metal: Ligand) molar ratio. Moreover, the data resulted from applying the mole ratio method support the same metal ion to ligand ratio of the prepared complexes (Figure 3).

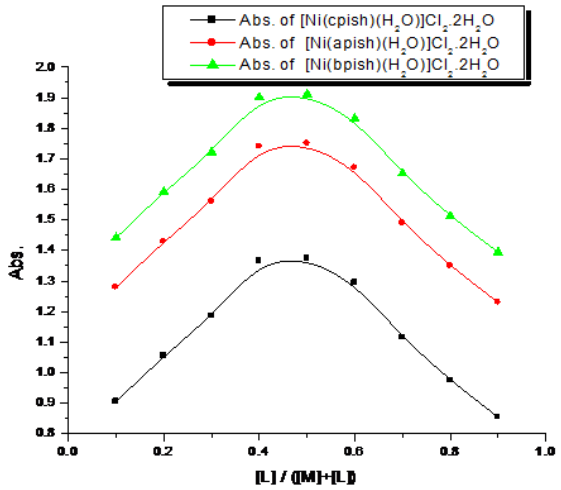


Figure - 2: Continuous variation plot of Ni(II)-Isatin-bishydrazone complexes.

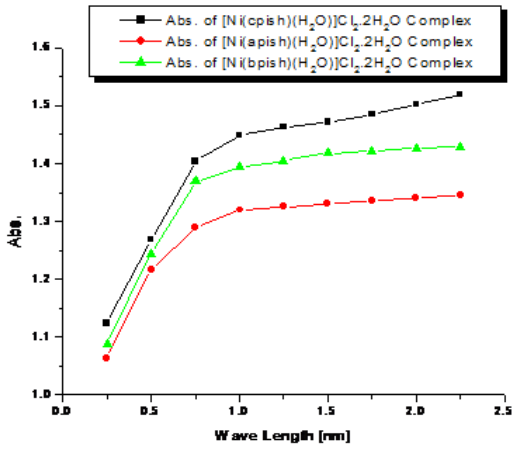


Figure - 3: Molar ratio plot of Ni(II)-Isatin-bishydrazone complexes.

3.2.2. Evaluation of the apparent formation constants of the synthesized complexes

The formation constants (K_f) of the studied Ni(II) Isatin-bishydrzone complexes formed in solution were obtained from the spectrophotometric measurements by applying the continuous variation method according to the following relations[63].

$$K_f = \frac{A/A_m}{\left(1 - A/A_m\right)^2 C}$$

Where A_m is the absorbance at the maximum formation of the complex, A is the actual absorbance of the complex and C is the initial concentration of the metal. As mentioned in Table 3, the obtained K_f values indicate the high stability of the prepared complexes. The values of K_f for the studied complexes increase in the following order: Ni-bpish > Ni-apish > Ni-cpish.

Table - 3: Stability constants and Thermodynamic parameters

Complex	Temp. (°C)	K_f (x 10^9)	Log(k_f)	$\Delta G^{\neq}$ (KJ/mol)
Ni-cpish complex	25	4.09	9.61	-54.8212
Ni-apish complex	25	4.47	9.65	-55.0321
Ni-bpish complex	25	5.43	9.73	-55.5348

$\Delta G^{\neq} = -RT \log K_f$; K_f is the formation constant, and $\Delta G^{\neq}$ is the free energy

3.3. Thermo-Gravimetric analysis (T.G, Dr.T.G) of the prepared Isatin-bishydrazone complexes.

The importance of this study on the Ni(II) complexes of the Isatin-bishydrazone ligands (cpish, apish and bpish) stems from their possible biological activities. Therefore, they are widely subjected to investigation by thermal analysis and other physicochemical methods. And also for perfect deduction of the complex structure, by determining the number of hydrated and coordinated water molecules. Thermal data of the complexes are given in (Table 4). The Ni (II) complexes of the Isatin-bishydrazone ligands exhibited thermal stability in the range ≈ 25 – 50 ^{0}C, and then degrade in three steps Figure (4(a), 4(b) and 4(c)). The first degradation step in the temperature range ≈ 350–360 K may account for the loss of the hydrated water molecules (two water molecules). The second degradation step in the temperature range ≈ 470–540 K may be attributable to the loss of the coordinated water molecule (one water molecule) and the third step of decomposition occurs within the temperature range ≈ 730–790 K corresponds to the loss of organic moiety leaving $NiCl_2$ as metallic residue.

Table - 4: Thermo analytical data. (TG, DTG)

Complex	Step	TG_{range} (°C)	DTA_{max} (°C)	Thermal effect	Mass loss: Obs. (Calc.) (%)	Assignment	Metallic residue
	I	53.11–106.55	78.38	Endo	8.98 (8.30)	$2H_2O$ (hydrated)	
Ni-cpish	II	106.55–426.69	209.42	Endo	5.13 (4.14)	$1H_2O$ (coordinated)	
complex	III	426.69–600.53	506.16	Endo	58.10 (57.68)	Organic moiety	$NiCl_2$
	I	54.07–219.16	78.71	Endo	8.55 (8.03)	$2H_2O$ (hydrated)	
Ni-apish	II	219.16–389.07	196.61	Endo	5.22 (4.02)	$1H_2O$ (coordinated)	
complex	III	389.07– 497.8	460.33	Endo	75.11 (72.86)	Organic moiety	$NiCl_2$
	I	56.07–210.55	87.68	Endo	7.35 (7.06)	$2H_2O$ (hydrated)	
Ni-bpish	II	210.55–404.91	265.57	Endo	4.05 (3.53)	$1H_2O$ (coordinated)	
complex	III	404.91-580.93	519.39	Endo	65.78 (63.99)	Organic moiety	$NiCl_2$

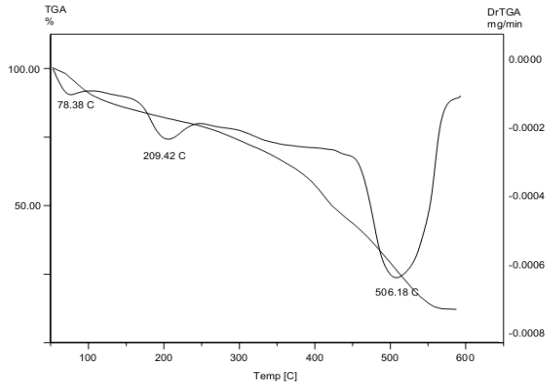


Figure 4(a): Thermal decomposition of $[Ni(cpish)(H_2O)]Cl_2.2H_2O$ Complex.

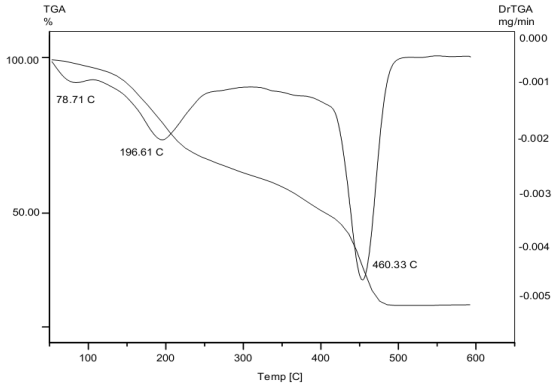


Figure - 4(b): Thermal decomposition of $[Ni(apish)(H_2O)]Cl_2.2H_2O$ Complex.

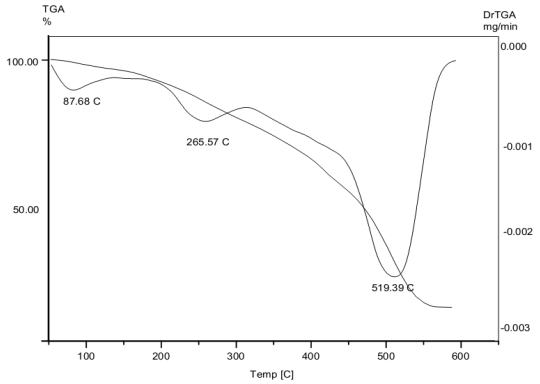


Figure - 4(c): Thermal decomposition of $[Ni(bpish)(H_2O)]Cl_2.2H_2O$ Complex.

3.4. Magnetic measurements

The magnetic measurements of the prepared Ni(II) Isatin-bishydrazone complexes showing the diamagnetic character, which suggested the square planar structure.

3.5. In-vitro biological activity (Antibacterial and anti-fungi screening)

The free ligands and there Ni(II) complexes were screened as anti-bacteria activity against three gram positive (Staphylococcus aureus (+ve), Micrococcus luteus (+ve), Bacillus cereus (+ve)), and three gram negative (Escherichia coli (-ve), Pseudomonas aeruginosa (-ve), Serratia marcescens (-ve)) and also antifungal activity against (Candida albicans, Geotrichum candidum, Trichophyton rubrum, Fusarium oxysporum, Scopulariopsis brevicaulis, Aspergillus flavus) to assess their potential antimicrobial activity. The susceptibilities of certain strains of bacteria and fungal cultures to Isatin-bishydrazone complexes were evaluated by measuring the diameter (in mm) of the inhibition zone around cavities. The antimicrobial activity data of all synthesized Isatin-bishydrazone Ligands [37] and there Ni(II) complexes are summarized in tables 5 and 6 and show that the newly synthesized ligands and their Ni(II) complexes possess notable biological activity. The antibacterial screening results exhibited marked enhancement in activity on coordination with the metal ions against most the testing bacterial strains cf. Figure 5 and 6. This enhancement in the activity can be rationalized to the basis of the structures of the ligands by possessing an additional azomethine (-C=N) linkage which is important in elucidating the mechanism of transamination and resamination reaction in biological system[64]. It has also been suggested that the ligands with nitrogen and oxygen donor systems might inhibit enzyme production[65], since the enzymes which require these groups for their activity appear to be especially more susceptible to deactivation by the metal ions upon chelation[66]. The polarity of the metal ion is

reduced by chelation [67] and this mainly because of the partial sharing of its positive charge with the donor groups and possibly with the delocalized π-electrons within the whole chelation ring, which is formed because of the coordination. This process of chelation increases the lipophilic nature of the central metal atom, which in turn favors its permeation through the lipoid layer of the membrane[68]. This is also responsible for the increasing of the hydrophobic character and liposolubility of the molecules in crossing the cell membrane of the microorganism and hence enhances the biological utilization ratio and activity of the testing drug/compound. The results are quite promising. It is evident from the results that, the biological activity of the metal complexes is higher than the corresponding ligands. This enhancement in the activity of the metal complexes can be explained on the basis of chelation theory [69]. It is, however, known that the chelating tends to make the Schiff base act as more powerful and potent bactereostatic agents, thus inhibiting the growth of bacteria and fungi more than the parent ligand [70-72], and also, noticed that the activity of Ni(II) complexes were more than the paraent ligands.

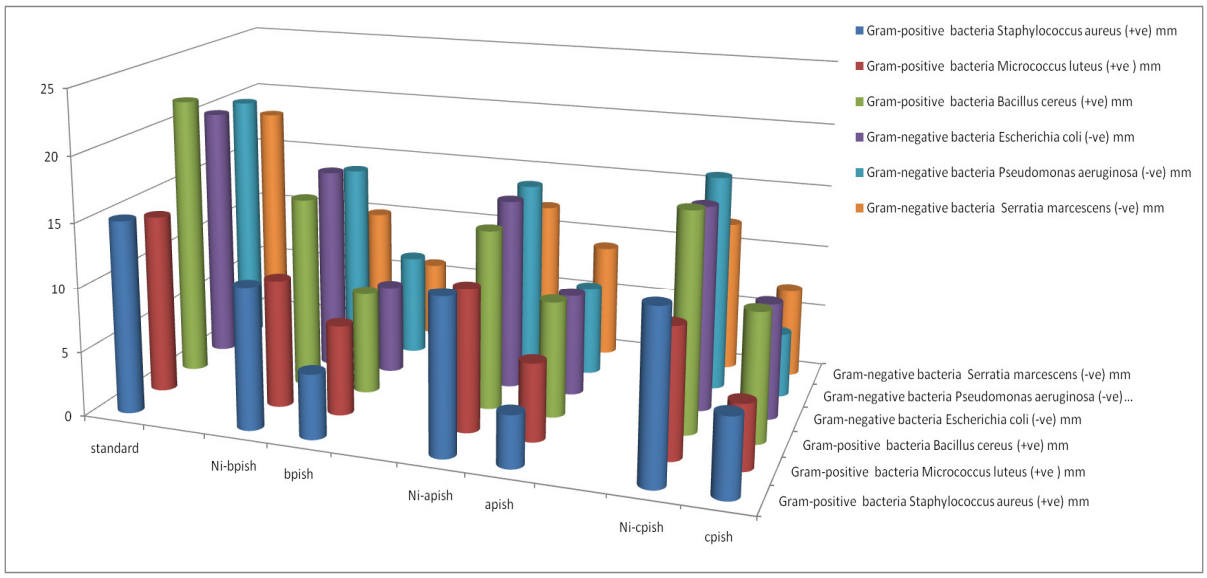


Figure - 5: Antibacterial activity of the synthesized Isatin-hydrazone ligands and their Ni(II) complexes.

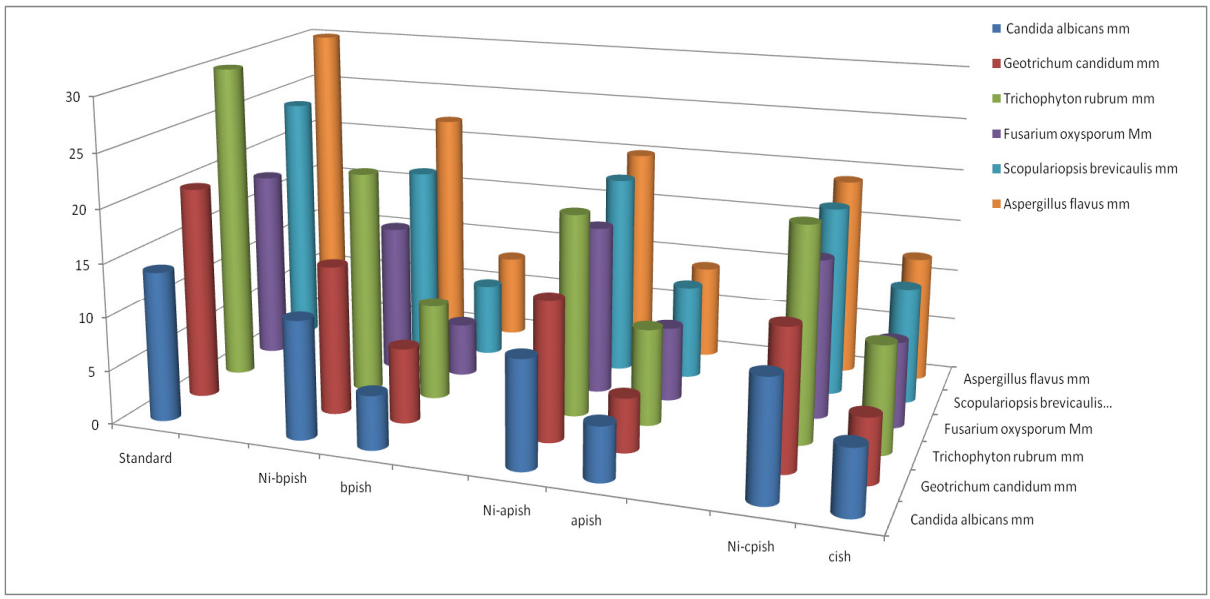


Figure - 6: Antifungal activity of the synthesized Isatin-hydrazone ligands and their Ni(II) complexes.

Table - 5: Antibacterial activity of the synthesized Isatin-hydrazone ligands and their metal complexes.

Compound Conc. (ppm)		Diameter of inhibition zone in mm , Minimum Inhibition Concentration (MIC) and Activity Index (%) for Some Bacteria																	
		Gram-positive bacteria									Gram-negative bacteria								
		Staphylococcus aureus (+ve)			*Micrococcus luteus (+ve)*			*Bacillus cereus (+ve)*			*Escherichia coli (-ve)*			*Pseudomonas aeruginosa (-ve)*			*Serratia marcescens (-ve)*		
		mm	*MIC*	*%*	*mm*	*MIC*	*%*	*mm*	*MIC*	*%*	*Mm*	*MIC*	*%*	*mm*	*MIC*	*%*	*mm*	*MIC*	*%*
cpish	100	6	45	40	5	50	35.7	10	40	45.4	9	50	45	5	45	25	7	50	38.8
apish	100	4	40	26.7	6	45	42.8	9	55	40.9	8	45	40	7	50	35	9	55	50
bpish	100	5	50	33.3	7	40	50	8	45	36.4	7	45	35	8	50	40	6	50	33.3
Ni-cpish	100	13	20	86.7	10	15	71.4	17	20	77.3	16	25	80	17	20	85	12	30	66.7
Ni-apish	100	12	25	80	11	20	78.6	14	20	63.6	15	25	75	15	15	75	12	25	66.7
Ni-bpish	100	11	25	73.3	10	25	71.4	15	15	68.2	16	20	80	15	25	75	10	25	55.6
Chloramphenicol As antibacterial standard		15		100	14		100	22		100	20		100	20		100	18		100

mm; Diameter of inhibition zone in mm, MIC; Minimum Inhibition Concentration (ppm), %; Activity Index.

Table – 6: Anti-fungi activity of the synthesized Isatin-hydrazone ligands and their metal complexes.

Compound Conc. (ppm)		Diameter of inhibition zone in mm, Minimum Inhibition Concentration (MIC) and Activity Index (%) for Some fungi																	
		Candida albicans			*Geotrichum candidum*			*Trichophyton rubrum*			*Fusarium oxysporum*			*Scopulariopsis brevicaulis*			*Aspergillus flavus*		
		mm	*MIC*	*%*	*mm*	*MIC*	*%*	*mm*	*MIC*	*%*	*Mm*	*MIC*	*%*	*mm*	*MIC*	*%*	*mm*	*MIC*	*%*
cish	100	6	40	42.8	6	55	30	10	40	33.3	8	30	44.4	11	40	45.8	12	45	40
apish	100	5	50	35.7	5	45	25	9	30	30	7	35	38.9	9	50	37.5	9	55	30
bpish	100	5	55	35.7	7	50	35	9	35	30	5	40	27.8	7	45	29.2	8	40	26.7
Ni-cpish	100	11	30	78.6	13	25	65	20	20	66.7	15	30	83.3	18	20	75	19	30	63.3
Ni-apish	100	10	25	71.4	13	30	65	19	20	63.3	16	25	88.9	19	20	79.2	20	20	66.7
Ni-bpish	100	11	25	78.6	14	25	70	21	25	70	14	20	77.8	18	25	75	22	15	73.3
Clotrimazole as antifungal standard		14		100	20		100	30		100	18		100	24		100	30		100

mm; Diameter of inhibition zone in mm, MIC; Minimum Inhibition Concentration(ppm), %; Activity Index.

4. REFERENCES

1. Mariar CR, Belicchi FM, Franco B, Corrado P, Giorgio P, Silvana P and Monica S. **J. Inorg. Biochem.,** 2004; 98: 313.
2. Sridhar SK and Ramesh A. **Biol. Pharm. Bull.,** 2001; 24: 1149.
3. Abdel Aziz AA, Salem ANM, Sayed MA and Aboaly MM. **J. Mol. Str.,** 2012; 1010: 130.
4. Garnovskii AD, Nivorozhkin AL and Minkin VI. **Coord. Chem. Rev.,** 1993; 1: 126.
5. Costamagna, Vargas J, Latorre R, Alvarado A and Mena G, **Coord. Chem. Rev.** 1992; 67: 119.
6. Walsh CT and Orme-Johnson WH. **Biochemistry,** 1987; 26: 4901.
7. Katia B, Simon L, Anne R, Gerard C, Francoise D and Bernard M. **Inorg. Chem.**, 1996; 35: 387.
8. Solomon EI and Lowery MD. **Science,** 1993; 259: 1575.

9. Gerdemann C, Eicken C and Krebs B. **Chem. Res.,** 2002; 35: 183.
10. Mallikarjun SY and Sangamesh AP. **Transition Met. Chem.,** 1997; 22: 220.
11. Yang GW, Xia XP, Tu H and Zhao XC. **Chem. Res. Appl.,** 1995; 7: 41.
12. Daisley RW and Shah VK. **J. Pharm. Sci.,** 1984; 73: 407.
13. Pandeya SN, Sriram D, Declecq E, Pannecouque C and Mitvrouw M. **Indian J. Pharm. Sci.**, 1999 60, 207.
14. Pandeya SN and Dimmock JR. **Pharmazie.** 1993; 48: 659.
15. Boon R. **Antiviral Chem. Chemother.,** 1997; 8: 5.
16. Pandeyab SN, Siram D, Nath G and Declercq E. **Eur. J. Pharm. Sci.** 1999; 9: 25.
17. Khan KM, Khan M, Ali M, Taha M, Rasheed S, Perveen S and Choudhary MI. **Bioorg. Med. Chem.** 2009; 17: 7795.
18. Sarangapani M, Reddy NA, Jayamma Y and Reddy VM. Indian Drugs. 1998, 35, 336.
19. Sarangapani M and Reddy VM. **Indian Drugs.** 1999, 36, 357; (b) M. Sarangapani, V. M. Reddy, Indian J. Pharm. Sci. 1996, 58, 147.
20. Sarangapani M and Reddy VM. **J. Pharm. Sci.** 1997; 59: 105.
21. Singh GS, Singh T and Lakhan R. Indian **J. Chem., Sect. B.** 1997; 36: 951.
22. Lingaiah N, Narendra R and Dattatray AM. **Indian J. Chem., Sect. B.** 1998; 37: 1254.
23. Medvedec AE, Clow A, Sandler M and Glover V. **Biochem. Pharmacol.** 1998; 52: 385.
24. Panova NG, Zemskova MA, Axenova LN, Medevedev AE. **Neurosci. Lett.** 1997; 223: 58.
25. Murukan B and Mohanan K. **J. Enzyme Inhib. Med. Chem.** 2007; 22: 65.
26. Hassan AMA. **J. Chem.** 1997; 36A: 241.
27. Vicini P, Zani F, Cozzini P and Doytchinova I. **Eur. J. Med. Chem.** 2002; 37: 553.
28. Despaigne AAR, Vieira LF, Mendes IC, Da-Costa FB, Speziali NL and Beraldo H. **J. Braz. Chem. Soc.** 2010; 21: 1247.
29. Moldovan CM, Oniga O, Parvu A, Tiperciuc B, Verite P, Pirnau A, Drişan O, Bojita M and Pop R. **Eur. J. Med. Chem.** 2011; 46: 526.
30. Eissa AAM, Farag NAH and Soliman GAH. **Bioorg. Med. Chem.** 2009; 17: 5059.
31. Hoonur RS, Patil BR, Badiger DS, Vadavi RS, Gudasi KB, Dandawate PR, Ghaisas MM, Padhye SB and Nethaji M. **Eur. J. Med. Chem.** 2010; 45: 2277.
32. Lovejoy DB and Richardson DR. **Blood.** 2002; 100: 666.
33. Richardson DR, Tran EH and Ponka P. **Blood.** 1995; 86: 4295.
34. Savini L, Chiasserini L, Travagli V, Pellerano C, Novellino E, Cosentino S and Pisano MB. **Eur. J. Med. Chem.** 2004; 39: 113.
35. Garg BS, Singh PK and Garg SK. **Indian J. Chem,** 1991; 30A: 979.
36. Abu El-Reash GM, Taha F, Shallaby AM and El-Gamal OA. **Indian J. Chem,** 1991; 30A: 286.
37. Mostafa. K. Rabia, Ahmad Desoky M Mohamad, Nabawia M. Ismail and Ali Abdo M. Ahmed. **Russian journal of general chemistry,** (accepted manuscript).
38. Kulkarni AD, Patil SA and Badami PS. **Int. J. Electrochem. Sci.,** 4 (2009) 717 – 729.
39. Yoe JH and Jones AL. **Ind. Eng. Chem.** (Analyst. Ed.), 1944; 16: 111-115.
40. El Shiekh R, Akl M, Gouda A and Ali W. **J. Am. Sci.** 2011; 7(4): 797-807.
41. Shah SS and Parmar RG, **Der Pharma Chemica**, 2011; 3(1): 318-321.
42. Job P. **Ann. Chem.,** 1928; 9: 113-203.
43. Issa RM, Hassanein AA, El-Mehasseb I M and Abed El-Wadoud RI. Spectrochimica **Acta Part A, 2006;** 65: 206–214.
44. Figgis BN and Lewis J. Progress in Inorganic Chemistry, Inter Science, New York 1964; 6.
45. Earnshaw A, King EA and Larkworthy LK. **J. Chem. Soc., Sect. A,** 1968: 1048.
46. Figgis BN and Nyholm RS. **J. Chem. Soc.,** 1959; 388.
47. Kwon- Chung and Bennett. **1992**
48. Singh VP and Katiyar A. **Pesticide Biochemistry and Physiology** 2008; 92: 8–14.
49. Geary WJ. **Coord. Chem. Rev.** 1971; 7: 81.
50. Olchoubian HG, Nazaria O and Ariuki BK. **J. Chin. Chem. Soc.,** 2011; 58:1.
51. Neena S, Sunita H, Jyoti S, Vanita P and Agarwala BV. **Synth. React. Inorg. Met.-Org. Chem.,** 1992; 22:1283.
52. Nakamoto K. Infrared spectra of Inorganic and Coordination Compounds, **Wiley-Interscience:** New York, 1970.
53. Murukan B, Bhageerethi S.K and Kochukittan M. **J Coord. Chem.,** 2007; 60; 15:1607.

54. Murukan B, Bhageerethi SK and Kochukittan M. **J Coord. Chem.,** 2007; 60; 15: 1607.
55. Cerchiaro G, Saboya PL, Ferreira AMC, Tomazela DM and Eberlin MN. **Tran. Met. Chem.,** 2004; 29: 495.
56. Hatzidumitrious A and Bolos CA. **Polyhedron,** 1998; 7: 1779.
57. Nakamoto K. Infrared spectra of Inorganic and Coordination Compounds, **Wiley-Interscience:** New York, 1970.
58. Yoe JH and Jones AL. **Ind. Eng. Chem.** (Analyst. Ed.), 1944; 16: 111-115.
59. El Shiekh R, Akl M, Gouda A and Ali W. **J. Am. Sci.** 2011; 7(4): 797-807.
60. Shah SS and Parmar RG. **Der Pharma Chemica,** 2011; 3(1): 318-321.
61. Job P. **Ann. Chem.,** 1928; 9:113-203.
62. Issa RM, Hassanein A.A, El Mehasseb IM and Abed. El Wadoud RI. **Spectrochimica Acta Part A**, 2006; 65: 206–214.
63. Abdel-Rahman LH, El-Khatib RM, Nassr LAE and Abu-Dief AM. **Journal of Molecular Structure**, 2013; 1040: 9–18
64. Nishat N, Hasnain S and Asma SD. **J. Coord. Chem.** 2010; 63(21): 3859-3870.
65. Chohan ZH, Supuran CT and Scozzafava A. **J. Enz. Inhib. Med.Chem.,** 2004; 19: 79-84.
66. Avajia PG, Kumar CHV, Patilc SA, Shivanandad KN and Nagaraju C. **Eur. J. Med. Chem.** 2009; 44(9): 552–3559.
67. Rehman SU, Chohan ZH, Naz F and Supuran CT. **J. Enzy. Inhib. Med. Chem.** 2005; 20: 333-340.
68. Chohan ZH, Arif M, Shafiq Z, Yaqub M and Supuran CT. **J. Enz. Inhib. Med. Chem.** 2006; 21: 95-103.
69. Thimmaiah KN, Lloyd WD and Chandrappa GT. **Inorg. Chim. Acta.,** 1985; 106: 81.
70. Kulkarni A, Avaji PG, Bagihalli GB, Badami PS and Patil SA**. J. Coord. Chem.,** 2009; 62;3: 481.
71. Kulkarni AD, Patil SA and Badami PS. **J. Sulf. Chem.,** 2009; (In Press).
72. Chohan ZH, Pervez H, Rauf A, Khan KM and Supuran CT. **J. Enz. Inhib. Med. Chem.,** 2004; 19: 417.