Spectral and cytotoxic study of complexing mixed ligand formed by sodium [5-(P-nitro phenyl)-4-phenyl-1,2,4-traizole-3-dithiocarbamato hydrazide] and 1,10 phenanthroline with Fe(II...

Article · January 2016

CITATION
1

READS
33

5 authors, including:

Nibras Mohammed
King Faisal University
6 PUBLICATIONS **8** CITATIONS
SEE PROFILE

Mahasin F Alias
University of Baghdad
72 PUBLICATIONS **204** CITATIONS
SEE PROFILE

Farah Saadoon Jaafar
Ibn Sina University of Medical and Pharmaceutical Sciences Iraq Baghdad
9 PUBLICATIONS **6** CITATIONS
SEE PROFILE

International Journal of Pharmacy
Journal Homepage: http://www.pharmascholars.com

Original Article **CODEN: IJPNL6**

Spectral and cytotoxic study of complexing mixed ligand formed by sodium [5-(*P*-nitro phenyl)-4-phenyl-1,2,4-traizole-3-dithiocarbamato hydrazide] and 1,10 phenanthroline with Fe(III) and Mn (II) ions

Mahasin Alias[a], Farah Saadoon[a], Maha Faisal[b], Nibras Mohammed[a], Caroline Shaker[c]

[a] Department of Chemistry/ College of Science for Woman/ University of Baghdad, Iraq
[b] College of Dentistry/ University of Baghdad, Iraq.
[c] Central Organization for Standardization and Quality Control, Ministry of Planning, Iraq.

***Corresponding author e-mail:** alias_mahasin@yahoo.com

Received on: 06-02-2016; Revised on: 15-03-2016; Accepted on: 19-03-2016

ABSTRACT

Mixed ligands complexes have been prepared by reaction of Sodium [5-(p-nitrophenyl)-'4-phenyl-1,2,4-traizole-3-dithiocarbamato hydrazide] (L_1) and 1,10-phenanthroline (L_2) with Fe(III) and Mn(II) ions. The newly prepared complexes were isolated and characterized by FT-IR and UV-Vis spectroscopy, C.H.N., FAA technique, in addition to magnetic susceptibility and conductivity measurements. The nature of bonding between the metal ion and the donor atoms of the ligands were demonstrated by calculating of the ligand field parameters using suitable Tanaba-Sugano diagrams. Cytotoxic effect of the prepared complexes were evaluated against Hep-2 cell line using different concentrations (3000, 1500, 750 and 375 µg/ml) respectively in an exposure time 72 hrs comparing this effect with control positive Cis-Pt as reference drug.

***Keywords*:** Dithiocarbamates, N,N-bidentate ligand ,Cytotoxic effect, Mixed ligand

INTRODUCTION

Dithiocarbamate (DTC) formed an important class of biologically active ligands **[1]**. An extremely large number of dithiocarbamate complexes with transition and non transition metal ions have been known **[1,2]**.The chelating property of DTC arises from the π-electron flow from the nitrogen atom to the sulphur atom via a plane delocalized π-orbital system. The net effect of such an electron flow is strong donation to metal, resulting in a high electron density on the metal **[2,3]**. The use of antitumour drugs based on platinum(II) metal complexes as ; cisplatin and its analogues, carboplatin and oxaliplatin is limited by two factors: installation of tumour drug resistance and severe adverse effects, so therapeutic strategies are oriented towards the development of new platinum- and non-platinum based antitumour drugs with higher efficiency, reduced general toxicity and broader spectrum of activity. Dithiocarbamates and their complexes have attracted particular attention for the use of chemical modulation of cisplatin nephrotoxicity **[4]**.

1,10-Phenanthroline has been extensively used as a classical *N,N* - bidentate ligand to prepare mixed-ligand complexes in coordination chemistry because it has varity properities such as high stability in chemical form , red.-ox. properties in addition to its capability to good coordination and its metal complexes can appear numerous biological properties**[5,6]**.

EXPERIMENTAL

Equipments: Melting point apparatus of Gallen Kamp M.F.B-60 was used to measure melting points of all compounds. FT-IR spectra were recorded as CsI discs using FT-IR3800 Shimadzu in the range of

(4000-200 cm^{-1}). Electronic spectra were obtained using UV-1650PC Spectrophotometer at room temperature in DMF solvent. Conductivity was measured by capacitor analyzer in DMF solution (10^{-3} M) at room temperature using (WTW) Conductometer. Elemental analysis was performed by using EM-017mth instrument. Magnetic susceptibility measurements were obtained at 25 ^{o}C Magnetic Susceptibility Balance of Johnson matty catalytic system division, England. The metal percent of the complexes were determined by using GBS-933 Flam plus Atomic Absorption Spectrophotometer.

Preparations:

a) The ligand Sodium [5-(p-nitro phenyl)-4-phenyl-1,2,4-traizole-3-dithio- carbamato hydrazide] as L1 (TRZ.DTC) was prepared according to previously literature **[7]**.

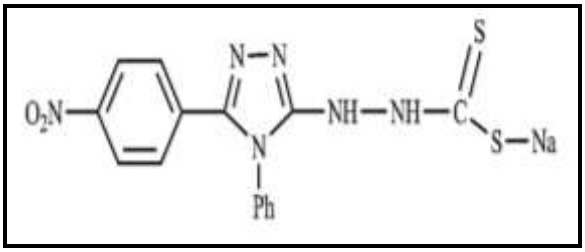

Figure (1): Structure of TRZ.DTC ligand.

b) Fe (III) and Mn (II) complexes were prepared conventionally by the reaction of (0.1 mole) of L_1 in 15 mL ethanol with (0.1 mole) of phenanthroline (Phen.) L_2 in the same solvent, then a warm solution of each of the metal salts (0.1mole) were added in 5 mL of absolute ethanol in molar ratio 1:1:1. The mixture was heated and refluxed with stirring for 45 min. The colored precipitates were filtered, washed several times with mixture of solvents (ethanol:water) and dried under vacuum. The obtained results showed a dark orange crystals with 85% for Mn (II) complex, and reddish orange crystals with 89% for Fe (III) complex.

Cytotoxic assay: Cancer cells were grown in DMEM medium containing 2 mM L-glutamine supplemented with 1000 U/L penicillin, streptomycin and 10% FBS. Briefly, cell lines suspended in DMED containing 10 % FBS were seeded with 1×10^4 cells (100 µL) per well in a 96-well plate. The incubation was performed at 37 °C under humidified (95% air, 5% CO_2) for 24 hrs. Additional medium (100 µL) containing the test compounds was added to a final concentrations of 3000, 1500, 750 and 375 µg/ml, further incubated for three days. All of the procedures concerning the cell culture maintenance, drug dissolution, and treatment were carried out in sterile conditions. After elapsing the incubation period, the culture medium was discarded from micro titer plates, 50 µl/well of neutral red dye solution was added to wells and the plates were incubated for 2 hrs. Plates were washed gently with phosphate buffered saline (PBS) and 100 µl/well of elution buffer was added, the absorbance was measured at 492nm by ELISA reader.The inhibition percentage was measured according to Gao *et al* equation **[8]** as follows:

$$\text{Inhibition rate\%} = \frac{\text{Absorbance of negative control - Absorbance of test}}{\text{Absorbance of negative control}} \times 100$$

RESULT AND DISCUSSION

Physio-chemical properties: Table (1) summarized some physical and the analytical data of the prepared complexes.The experimental values are approximate agreemant with theoretically calculated values correspond to the suggested composition.The isolated complexes in solid state are stable at room temperature , soluble in most organic solvents and their general formula $[M(L_1)(L_2)(Cl)x(H_2O)y].nH_2O$ where M = Mn(II) and Fe(III) ; x= 1, 2 ; y = 1,0 and n = 1,2, respectively.

FTIR spectral analysis: As shown previously by Bonati and Ugo **[9]** the presence of a solitary band in the range (950-1050) cm^{-1} is due to symmetrical bidentate coordination of the dithiocarbamato group while the splitting of this band within a narrow range of (±20) cm^{-1} is due to the asymmetrical ansiobidentate or monodentate nature of the dithiocarbamato group**[7, 9]**. The singlet in the infrared spectrum of dithiocarbamato ligand confirms the equivalence of the S-atoms, but the splitting in the infrared spectrum indicates non-equivalence of the S-atoms in these compounds. In the present work, the bands appeared in this region are splitting refers to ansio bidentate binding of dithiocarbamato group **[10]**. The band of νC-S which seen in 995cm^{-1} for free ligand was suffered to negative shift about (-20) cm^{-1} in both complexes. Also we noticed lower shifted in complexes at about (21-27) cm^{-1} in νC=S band which appeared at 1041cm^{-1} in free ligand , this results indicate to contribution of dithiocarbamato moiety in complexation from S-atom as M-S bond which appeared at the positions (435 and 439) cm^{-1} in complexes**[11]**.

Table (1): Some physical and analytical data for the ligands and their metal complexes

Compdound formula Colour	% Yield	M. p. °C	M. Wt g.mol $^{-1}$	% Elemental and metal analysis / Found (Calc.)				
				C	*H*	*N*	*S*	*M*
TRZ.DTC (L_1) Pale Orange	88.5	66-68	394.4	45.84 (45.63)	3.05 (2.78)	21.99 (21.29)	16.80 (16.22)	----
phen (L_2) white	----	100-102	198	----	----	----	----	----
[Mn(L_1)(L_2)Cl H_2O].H_2O Dark orange	85	163-165	695.8	45.87 (46.56)	3.38 (3.59)	15.98 (16.09)	9.54 (9.19)	7.25 (7.89)
[Fe(L_1)(L_2)Cl_2].2H_2O Reddish orange	89	293d	732.2	44.02 (44.25)	3.79 (3.41)	15.67 (15.29)	8.96 (8.74)	7.05 (7.62)

Where: d = decomposition degree

A ν$C{=}{=}N$ band at (1438- 1442) cm^{-1} region was observed, which matches well with the literature values **[12]**, while the band at (1496) cm^{-1} of ν N-N mode remained without change in both complexes.

The ligand show stretching and bending bands of NH(1,2) at 3380 and 1652 cm^{-1} respectively, which undergoes a slightly shift to higher frequency in Fe complex about (+5) cm^{-1} while not change in Mn complex, which is mean that these groups did not support the coordination ligand with central metal ion.

Also the presence of peaks that attributed to the aromatic C=C and C=N stretching of L_2 around (1616-1419 cm^{-1})**[13]**,so the spectra of the complexes show also characteristic bands which shifting and assigned to the stretching vibration of ν(C=C+C=N) of co-ligand,besides it shows bands at (270 and 265 cm^{-1}) has also been observed in the complexes that refer to coordination of co-ligand nitrogen atoms with metal ions as (M-N)**[13]**. In addition to a bands which assigned to lattice and coordinated water, also the bands of M-O and M-Cl in both complexes are illustrated in Table (2).

Table (2): Most diagnostic FTIR bands of L_1 and L_2 and their metal complexes in (cm^{-1}).

Compounds	TRZ.DTC (L_1)	Phen (L_2)	[Mn L_1 L_2 Cl H_2O].H_2O	[Fe L_1 L_2 Cl_2].2H_2O
ν$NH_{(1,2)}$	3380,3380	-----	3380,3380	3385,3385
δ$NH_{(1,2)}$	1652,1652	-----	1650,1650	1654,1654
νC=S	1041	-----	1020	1014
νC-S	995	-----	975	983
νN-N	1496	-----	1496	1496
νC-N	1438	-----	1432	1440
ν(C=C+C=N) Phen	-----	1616,1589,1558, 1504,1446,1419	1620,1585,1512, 1454,1432	1620,1577,1562, 1440,1384
νCH. arom.	-----	3059	3093	3082
νM-O	-----	-----	525	----
νM-S	-----	-----	439	435
νM-N	-----	-----	270	265
Others	-----	-----	νOH=3414 δH_2O =821 νMnCl =303	νOH=3421 νFeCl =317

Electronic spectral analysis: The electronic spectra of compounds were recorded as solution in DMF solvent. The electronic spectrum of free ligand (L_1) exhibited three main bands, when the first absorption band appeared at 266 nm (37593.98) cm^{-1} refers to interaligand (π→π*) transition located on the $N{=}{=}C{=}{=}S$ group. The second absorption band located at 312 nm (32051.28) cm^{-1} also back to (π→π*) transition, but within the $N{=}{=}C{=}{=}S$ group. The third absorption band assigned as (n→π*) and arise to sulfur atoms that appeared at 353 nm (28328.61) cm^{-1}**[10]**. The electronic spectrum of co-ligand L_2 shows a very strong absorption band in the ultraviolet region at 227 nm (44052 cm^{-1}) assinged to

the transition (π→π*) for the intera-ligand aromatic system C=C, while the other second and third absorption appeared at 270 nm (37037 cm^{-1}) and 344 nm (29069 cm^{-1}), respectively due to the transition (n→π*) of imine group C=N**[13]**.

The electronic spectrum of the orange Mn(II) complex shows four absorption bands, the first appeared at 18181 cm^{-1} due to the electronic transition $^6A_1g \rightarrow {}^4T_2g_{(G)}$ which represent ν_2. Also the second absorption at 20833cm^{-1} refers to $^6A_1g \rightarrow {}^4A_1g + {}^4Eg_{(G)}$, in addition to charge transfer bands CT that emerged at 28901 and 36101cm^{-1} respectively. These frequinces agreement with previous published for Mn(II) complexes that have octaheadral geometry**[14]**.The megnatic moment of prepared complexes shows paramagnetic properities and has the value 6.4 B.M. The electronic spectrum of reddnish orange Fe(III) complex demonstrated similar assignment of Mn(II) complex, where four absorption bands that appeared.The first band represent ν_2 = 19455 cm^{-1} arise to the electronic transition $^6A_1g \rightarrow {}^4T_2g_{(G)}$. The second absorption band at 25974cm^{-1} which corresponding to $^6A_1g \rightarrow {}^4A_1g + {}^4Eg_{(G)}$. The bands at 28571 and 36496 cm^{-1} refers to charge transfer bands CT, these values agreement with literatures of Oh geometry **[15]**. The megnatic moment value is equal to 5.24 B.M.

The diagrams of Tanabe–Sugano used to calculate the value of ν_n in the complexes of Mn(II) and Fe(III) ions. In addition to estimate the values; 10Dq and other ligand field parametre β, B′ and 15B′ as tabulated in Tables (3). The conductance measurements shows that the non-electrical behavior for both complexes.All results of electronic spectra, magnetic moment and electricity showed in Table (3).

Table (3): Electronic spectra, Conductance and magnetic moment for the ligands and their metal complexes.

Compounds	**TRZ.DTC (L_1)**	**Phen. (L_2)**	**[Mn L_1 L_2 Cl H_2O].H_2O**	**[Fe L_1 L_2 Cl_2].2H_2O**
Absorption Bands(cm^{-1})	28328 32051 37593	29069 37037 44052	9834(calc.) 1818 20833 28901 36101	11188(calc.) 19455 25974 28571 36496
Assignments	n→π* π→π* π→π*	n→π* n →π* π→π*	$^6A_1g \rightarrow {}^4T_1g_{(G)}$ $^6A_1g \rightarrow {}^4T_2g_{(G)}$ $^6A_1g \rightarrow {}^4A_1g + {}^4Eg_{(G)}$ CT CT	$^6A_1g \rightarrow {}^4T_1g_{(G)}$ $^6A_1g \rightarrow {}^4T_2g_{(G)}$ $^6A_1g \rightarrow {}^4A_1g + {}^4Eg_{(G)}$ CT CT
Bº	----	----	866	1300
B′	----	----	634	792.9
β			0.737	0.608
Dq/B′	----	----	1.65	1.75
10Dq	----	----	10461	13842
15B′	----	----	9510	11865
μ_{eff} (B.M.)	----	----	6.4	5.24
μs.cm^{-1}	----	----	27.99	35.52
Suggested geometry	----	----	Distorted Oh	Distorted Oh

CT = Charge Transfer

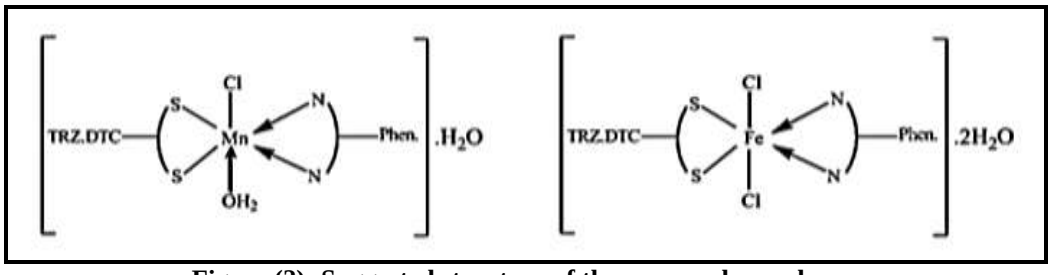

Figure (2): Suggested structure of the prepared complexes.

According to the results obtained from the spectral analysis, magnetic moment, the conductivity measurement and elemental analysis, so an octaheadral structure was suggested for prepared complexes as shown in Figure (2).

Cytotoxicity effect: The results of the *in vitro* cytotoxicity test of mixed ligands and their metal complexes were given in Table (4). The data showed that most of these prepared compounds were may be potent as anticancer activities against human cancer cell line HepG2 in time of exposure 72 hrs comparable with standard drug cis-Pt as control positive. The mixed ligand exhibited cytotoxic activity against selected cell line and in inhibition rates fairly close to the standard drug in low concentration more than high concentration and this effect may be due to presence of CS_2 group of dithiocarbamato as well as the chelating nitrogen atoms of 1,10-phenanthroline.

Most complexes of dithiocarbamato showed a clear preference for S-donor ligands, such as glutathione and cysteine with only limited reactivity against nucleosides and their bases **[16]**. The antitumor reactivity of several dithiocarbamato complexes of $[Pd(S_2CNEt_2)$ (1,10-phen.)$]NO_3$ aginst leukemia cells has been established **[17,18]**. Generally mixed ligands show more cytotoxicity compared with the prepared complexes. The results show that the Fe (III) complex has more influence toward selected cell line than the other Mn(II) complex , which not appears any activity in the concentration of 375 µg/ml and low rates of inhibition in other concentration comparable with Fe(III) and mixed ligands.

Both Mn(II) and Fe(III) is isoelectronic as a d^5 configuration in six coordination Oh system. The lowest inhibition for Mn(II) comparable to iron complex may be becuase the charge, size of metal and presence one chloride group makes unstable complex and doesn't have cytotoxicity effect on cell line and this may be the explanation of sudden downward inhibition rate on HepG2 cell line in low concentration of Mn(II) complex, while the Fe(III) complex exhibited highest inhibition rate rather than Mn(II) complex. This may be due to high oxidation state in Oh system and presence of two coavelant bonds chloride which is good leaving group to obtained another system and occurrece Red-Ox reaction.

Table (4): Inhibition rates of mixed ligands and thier metal complex comparable with control positive cis-Pt on Hep-2 cell line.

Conc. (µg/ml)	Cis-Pt	Mixed ligand	[Mn L_1 L_2 Cl H_2O].H_2O	[Fe L_1 L_2 Cl_2].$2H_2O$
3000	45.52	23.16	14.67	35.90
1500	55.31	45.55	26.64	33.20
750	62.05	51.35	27.41	54.44
375	70.99	68.33	Negative	54.44

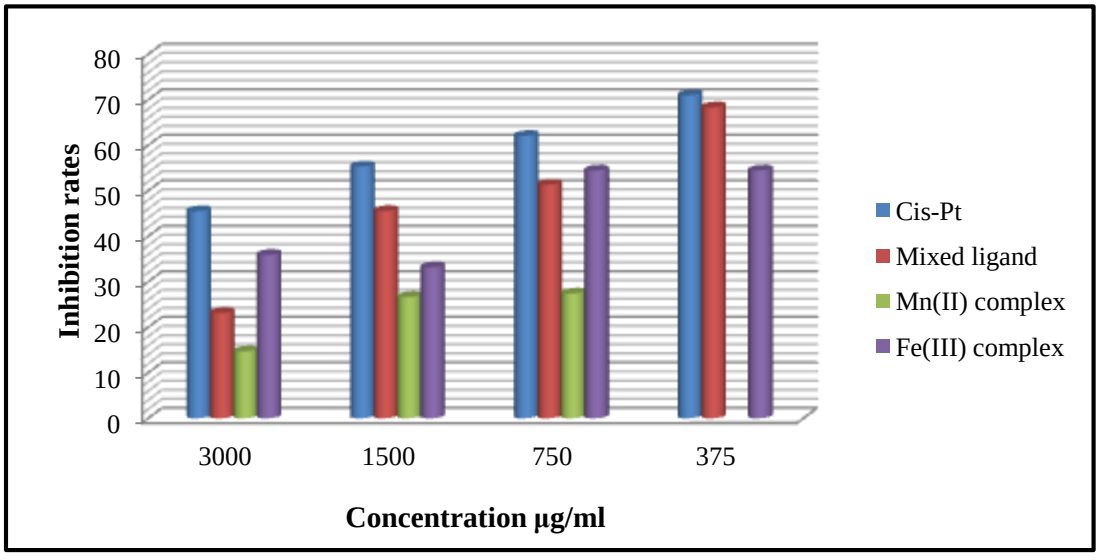

Figure (3): Shows the percentage inhibition rate on Hpe-2 cell lines after exposure to Mixed ligands and their metal complexes.

REFERENCES

1. Dawood Z. F. , Mohammed T. J. and Sharif M. R, IJVS,2009; 23(Supplement II) :135-141.
2. Balaramesh P., Abdul Jabbar A. and Venkatesh P, ijspr, 2014; 4(5):1-7.
3. Balaramesh P., Venkatesh P., Abdul Jabbar A, IJIRSET,2014; 3(8):15301-15309.
4. Valentina U. , Mihaela B., Victoria A. , Liviu Ch. and Rodica O. J Therm Anal Calorim , 2013;111:1177-1182 .
5. Mirela C. , Daniela M. and Gabriela P. , Rev. Roum. Chim. 2011; 56(3):231-237.
6. Sasikala K. and Arunachalam S. ,Chem Sci Trans., 2013;2(1):157- 166.
7. Carolin, S. H. and Alias M., Baghdad Sci. J. 2012; 9(4) : 668-679.
8. Gao X. , Xu X., Janakiraman N. and Gautam S., Nutv., 2002;132, 2076- 2081.
9. Bonati F. and Ugo R., J. Organomet. Chem. 1967; 10, 257–268.
10. Alias M., Rawnaq B. J., Farooq I. M. and Carolin Sh. H., Baghdad Sci. J. , 2015; 12(1):127- 139.
11. Siddiqi K. S.,Nami S. A., Lutfullaha and Chebude Y., J. Braz. Chem. Soc., 2006; 17 (1), 107–112.
12. Rayag, I., Baba, I., Yamin, B. M., Malaysia J. Anal. Sci., 2006;10 (1), 93-98.
13. Alias M., Farah S. J. and Amal S. S.,Int J Pharm ,2015; 5(2): 451-458.
14. Rahangdale M. , Pethe G. , Yaul A. and Aswar A., Res. J. Pharm. Biolog. Chem. Sci. , 2011; 2(3),: 341-348.
15. Deshmukh P. S., Yaul A. R., Bhojane J. N., Aswar A.,World J. Chem., 2010;5(1):57-61.
16. Alias M. and Maysoon M. A., Baghdad Sci. J., 2015; 12(4): 740- 752.
17. Marzano C., Fregona D., Baccichetti F., Trevisan A., Giovagnini L. and Bordin F. ,Chem. Biol. Interact, 2002;140: 215-229.
18. Chen-Shang C., Yip-Foo W., Sie-Tiong H. and Yousif E. , A. J. Chem., 2014 ; 26 (10) : 2949- 2953.