Determination of Antioxidant Activity for Some Metal Ions Complexes Derived from Mefanamic acid and Benzidine

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الملخص

تحضير وتشخيص بعض معقدات ايونات الفلزات من تفاعل حامض الميفانامك والبنزدين و بعض ايونات الفلزات ، ثم تحديد الفعالية المضادة للاكسدة ضد ١,١ داي فنيل ٢- بيكريل هيدرازيل (DPPH) الجذر الحر المستقر . معقدات الفلزات [Zn(II) , Cu(II) , Ni (II) (Mef-B)- Mn(II)] شخصت بواسطة التحليل الدقيق للعناصر ، محتوى الفلز ,محتوى الكلور، التوصيلية ، طيف الاشعة تحت الحمراء , طيف الاشعة فوق البنفسجية المرئية و الحساسية المغناطيسية . يتم دراسة الفعالية المضادة للاكسدة للمركبات المحضرة وثم توضع لمقارنتها مع حامض الغاليك القياسي الذي استخدم كمرجع فينولي لعمل دراسة الفعالية المضادة للاكسدة . تظهر الطريقة التغير في قدرة الكسح الحرة للمركبات المحضرة في تثبيط الجذور الحرة.

الكلمات المفتاحية : البنزدين ، الفعالية المضادة للاكسدة، الجذر الحر.

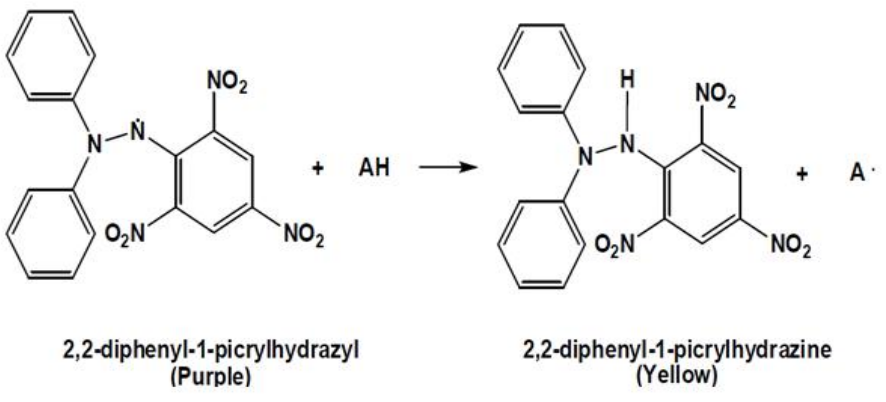
Abstract

The synthesis and characterization of the complexes of some metal ions from the reaction mefanamic acid with benzidine and some metal ions ,then the evaluation of its antioxidant activities against 1.1-Diphenyl-2-picrylhydrazyl (DPPH). The (Mef-B) metal complexes for [Mn(II), Ni(II) , Cu(II) and Zn(II)] characterization by elemental analysis, metal content, chloride containing, molar conductance, FT-IR spectra , UV-Vis spectra and magnetic susceptibility. Antioxidant efficiencies from compounds had been estimated, then subject of comparison with Gallic acid that employed as phenolic reference in order to make a study of the antioxidant activity. The outcomes illustrate that the compounds display exceptional root scavenging efficacies.

***KEYWORDS*:** Benzidine, antioxidant activities, free-radical

1. INTRODUCTION

Antioxidants represent a “free-radical-scavengers” and prevent or slow the damage done by these free radicals. Antioxidants act substances that neutralize the harmful free radicals in our bodies [1]. An antioxidant is a molecule that inhibits the oxidation of other molecules; oxidation is a chemical reaction that transfers electrons or hydrogen from a substance to an oxidizing agent, their activity redox properties [2-6]. Free-radicals are responsible for induction of several types of diseases such as Parkinson disease, cancer, heart diseases, arthritis, al zheimer diseases and diabets mellitus when it is produced in excess amount [7]. The different analytical methods for determining of the antioxidant capacity fall into four categories: (a) Spectrometry techniques. (b) Electrochemical techniques. (c) Chromatography techniques. (d) Fluorescence techniques. Spectrometry techniques consist of the DPPH (2, 2-dipheyl-l- picrylhydrazyl) method [8, 9]. Blois has developed the DPPH (2, 2-diphenyl-l- picrylhydrazyl) method in 1958 from a stable free-radical α, α-diphenyl-β-picrylhydrazy (M_{wt} =394.3 formula DPPH= $C_{18}H_{12}N_5O_6$) to evaluate the antioxidant activity. The assay is used for the measurement of the scavenging capacity of antioxidants towards it. The single electrons of the nitrogen atoms in DPPH are reduced by receiving a hydrogen atom from antioxidants to hydrazine. Electron delocalization also produces dark purple; which is characterized by an absorption band in an ethanol solution centered at about (517 nm) , when the DPPH radical solution is mixed with the antioxidant, the color of the respective hydrazine changes from violet to yellow[10,11], (Scheme -1).



Scheme [1]: DPPH-radical's chemical structure and its reaction with a scavenger indicated by AH

2. EXPERIMENTAL

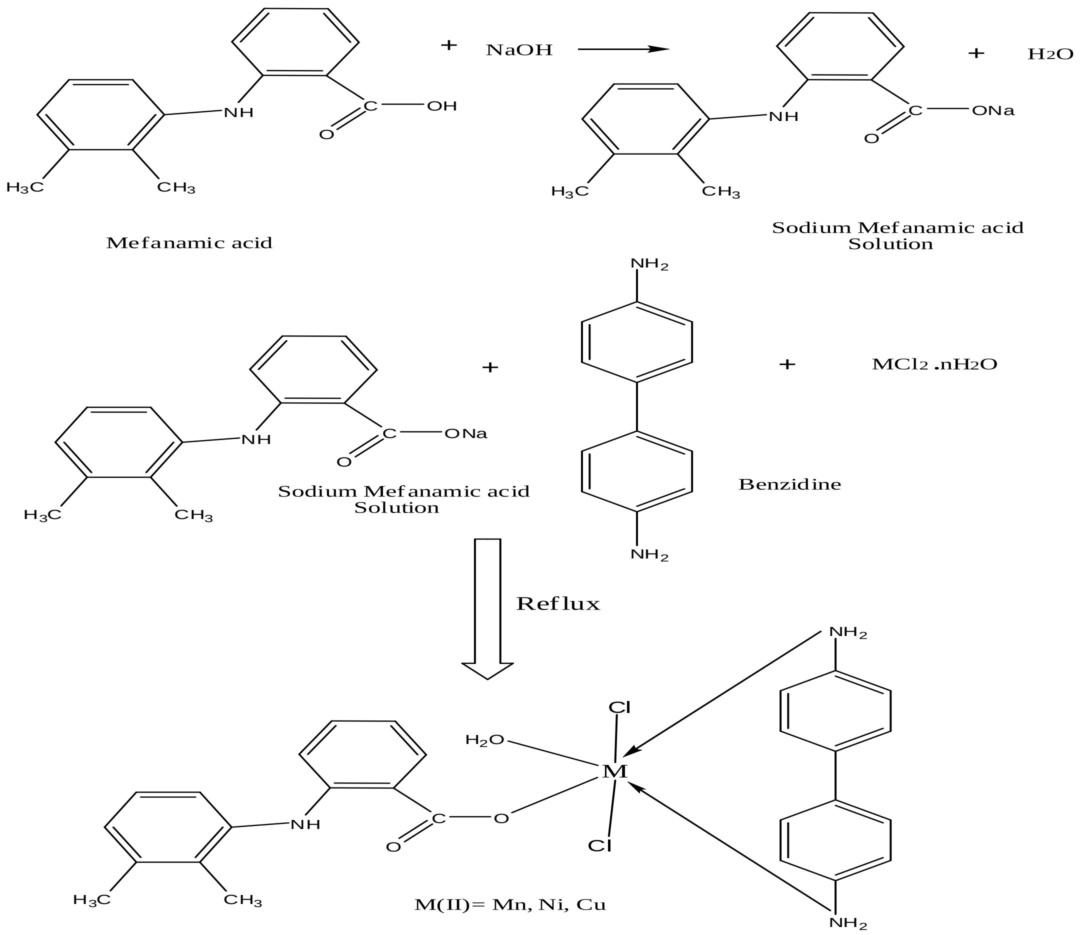
2.1 Materials and measurements

All the chemicals and reagents have been acquired from the commercial sources (Sigma-Aldrich, Merck, Fluka, Analar) and used without further purification.Elemental microanalysis (C.H.N) were performed on Eurovector EA 3000A. The content of metal ions was calculated gravimetrically as metal oxides. Molar conductance measurements of the (Mef-B) metal complexes were obtained using Conductometer WTW,at 25˚C with concentration of $1x10^{-3}$M. Magnetic measurements were examined by Balance of Johnson mattey catalytic system division at 25 °C, England. The UV vis spectra were examined using UV.-Vis. spectrophotometer, UV-1800 Shimadzu by cell quartz 1.0cm from of wavelength (200-1100 nm), using 1 cm quartz cell. Fourier Transform Infrared (FT-IR) spectra were obtained using SHIMADZU FT-IR 8400S Fourier transforms, within the wavenumber region between 4000 and 200 cm-1 using KBr disc and CsI disc.

2.2 General procedure for the synthesis of (Mef-B) metal complexes

The (Mef-B) metal complexes were prepared according in (Scheme-2), through the addition of the: First : Sodium mefanamic acid solution (10 mmol) in 30 ml of ethyl alchol with adding (5 mmol) of sodium hydroxide with distilled water

Second : benzidine solution (10 mmol), in 20 ml of ethyl alchol . All two ligands have been added with stirring to aqueous solution of $MnCl_2.4H_2O$, $NiCl_2.6H_2O$, $CuCl_2.2H_2O$ and $ZnCl_2$. The resulted homogeneous solution were refluxed for 2hr with stirring, the formation precipitate, then they were filtered, washed with ethyl alchol and recrystallized in ethanol and diethyl ether after that dried in vacuo anhydrous $CaCl_2$.

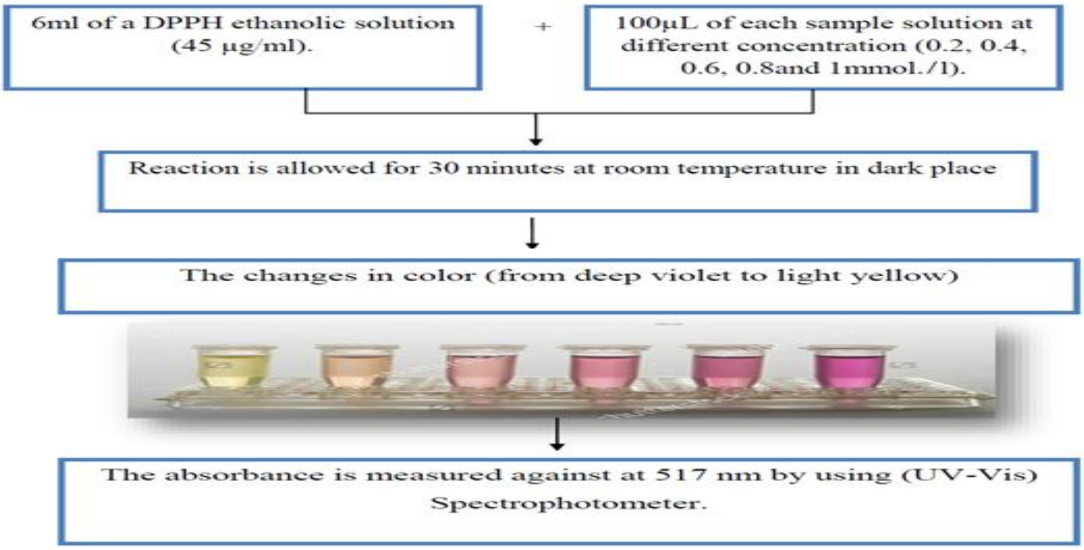


Scheme [2]: Proposed structures of (Mef-B) metal complexes

2.3 Study for antioxidant efficacy through means from DPPH manner

To study antioxidant efficacy for (Mef-B) metal complexes , Gallic acid has been utilized like phenolic reference. A chain from standards for gallic acid were prepared five normal solutions for various condensation (0.2, 0.4, 0.6, 0.8, as well 1 mmol1^{-1}were prepared of a 10 mmol l^{-1} solution from gallic acid, by the means of ethanol as diluents. 6 ml from 45 µg ml^{-1} DPPH solution has been added into 100 µL of each from normal solutions for gallic acid. Mixture has been brood on room temperature of 30 minutes at darkness, and thereafter, then absorbance from reacted mixture has been read on 517 nm. New (Mef-B) metal complexes have been dissolved on DMSO as well ethanol into get condensation from(10 mmol l^{-1}). Stock solutions had been diluted after that into (0.2, 0.4, 0.6, 0.8 as well 1 mmol.l^{-1}). (Scheme -3) Method of assaying free radical scavenging activity. Whereas gallic acid like standard, the percentage from DPPH root scavenger has been calculated utilizing Equation [10].

$$\text{DPPH scavenging ability (\%)} = \frac{\textit{Abs control} - \textit{Abs sample}}{\textit{Abs control}} \times 100$$



6ml of a DPPH ethanolic solution (45 μg/ml).

+

100μL of each sample solution at different concentration (0.2, 0.4, 0.6, 0.8and 1mmol./l).

Reaction is allowed for 30 minutes at room temperature in dark place

The changes in color (from deep violet to light yellow)

The absorbance is measured against at 517 nm by using (UV-Vis) Spectrophotometer.

Scheme [3]: Method of assaying free radical scavenging activity

3. RESULTS AND DISCUSSION

The physical properties and elemental results obtained from C.H.N. analyses, chloride contents and metal contents of the prepared compounds are described in (Table -1).

Table -1:-Elemental microanalysis results and some physical properties of (Mef-B) metal complexes

Compound	Formula	M.$_{Wt}$	Elemental Analysis Cal (Found)%				
			C	H	N	M	Cl
Mn-(Mef-B) $Cl_2.H_2O$	$C_{27}H_{26}N_3O_2Mn$ $Cl_2.H_2O$	567.7	(56.72) 57.07	(5.32) 4.93	(7.22) 7.39	(9.12) 9.67	(11.24) 12.47
Ni-(Mef-B) $Cl_2.H_2O$	$C_{27}H_{26}N_3O_2Ni$ $Cl_2.H_2O$	571.4	(57.12) 56.70	(5.05) 4.90	(6.93) 7.35	(9.88) 10.25	(12.02) 12.39

Cu-(Mef-B) $Cl_2.H_2O$	$C_{27}H_{26}N_3O_2Cu$ $Cl_2.H_2O$	576.3	(57.15) 56.22	(5.11) 4.85	(7.05) 7.28	(10.86) 11.01	(12.07) 12.28
Zn-(Mef-B) $Cl_2.H_2O$	$C_{27}H_{26}N_3O_2Zn$ $Cl_2.H_2O$	578.1	(56.50) 56.04	(4.30) 4.84	(7.09) 7.26	(10.92) 11.29	(11.87) 12.24

3.1 Infrared analysis

The important characteristics absorption bands of the (Mef-B) metal complexes shown in figures (1-4) and describes in (Table -2)[12,13].

Table -2 :- Infrared spectral data of the (Mef-B) metal complexes

Compound	v(NH)	v(NH_2)	v(C=O)	v(C=C) Arom.	v (M-N)	v (M-Cl)	v (H_2O)
Mn-(Mef-B) $Cl_2.H_2O$	٣٤٥٦.١٢	٣٣٤٧.٤٢	1734.21	١٤٣٥.٧٣	٥٣٣.١٢	٤٦٥.٢١	3540.33
Ni-(Mef-B) $Cl_2.H_2O$	3451.32	3341.28	١٧٢٣.٤٣	1448.98	542.77	478.23	٣٥٤٧.١٣
Cu-(Mef-B) $Cl_2.H_2O$	3457.43	3354.49	١٧٠٣.٦٧	1475.67	537.41	443.67	٣٥٣٣.٤٥
Zn-(Mef-B) $Cl_2.H_2O$	3449.25	3350.72	١٧٤٢.١٢	1427.52	٥٤١.٣٥	٤٧٨.٥٣	٣٥٣٧.٩٦

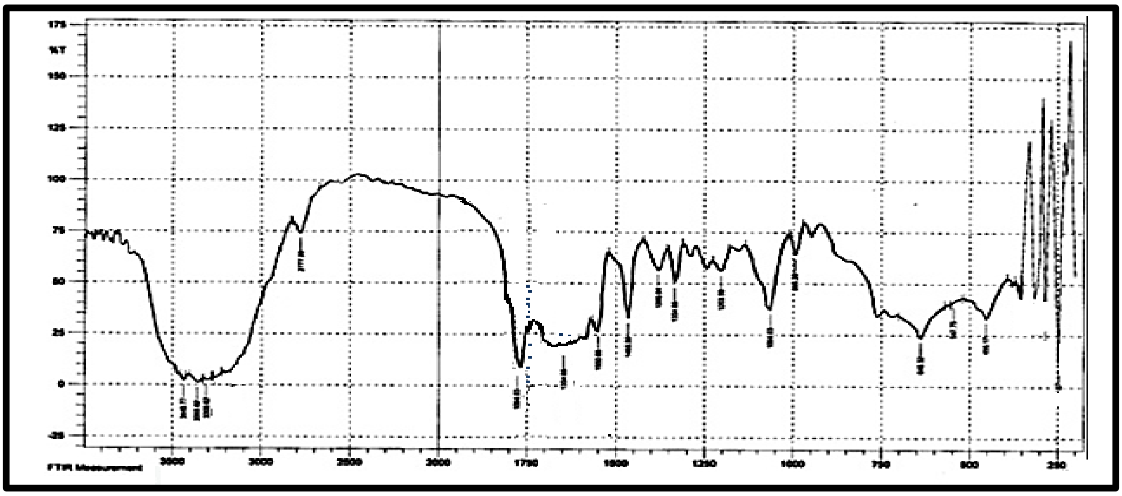


Figure -1: FTIR spectrum of Mn-(Mef-B) $Cl_2.H_2O$ complex

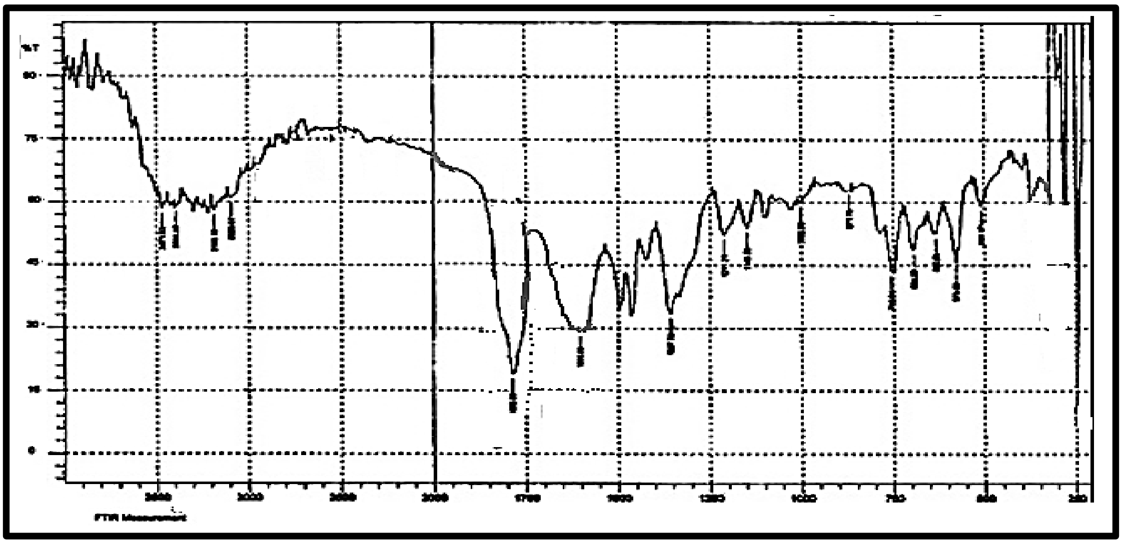


Figure -2: FTIR spectrum of Ni-(Mef-B) $Cl_2.H_2O$ complex

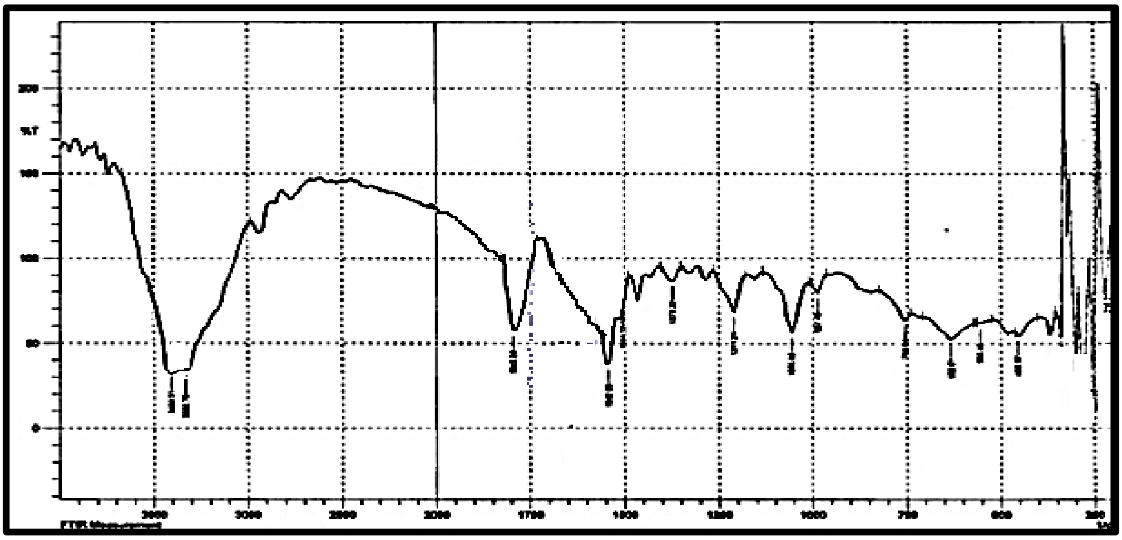


Figure -3: FTIR spectrum of Cu-(Mef-B) $Cl_2.H_2O$ complex

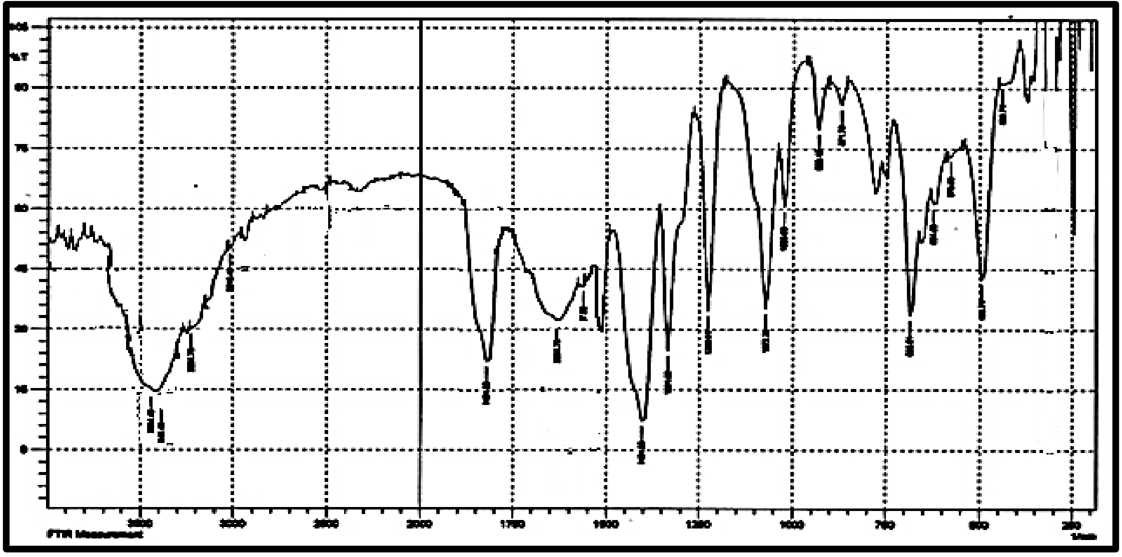


Figure -4: FTIR spectrum of Zn-(Mef-B) $Cl_2.H_2O$ complex

3.2 Electronic spectra and magnetic moments studies

3.2.1. Mn-(Mef-B) $Cl_2.H_2O$ complex

The Mn(II) complex shows a magnetic moment of 5.06 BM at room temperature corresponding to the five unpaired electrons.The electronic spectrum exhibited four absorption peaks. The two peaks at (265, 375) nm attributed to the ligand field and (C. T), The last two peaks at (534,615) nm attributed due to (d-d)electronic transitions , which are a good evidence for Octahedral geometry.

3.2.2. Ni-(Mef-B) $Cl_2.H_2O$ complex

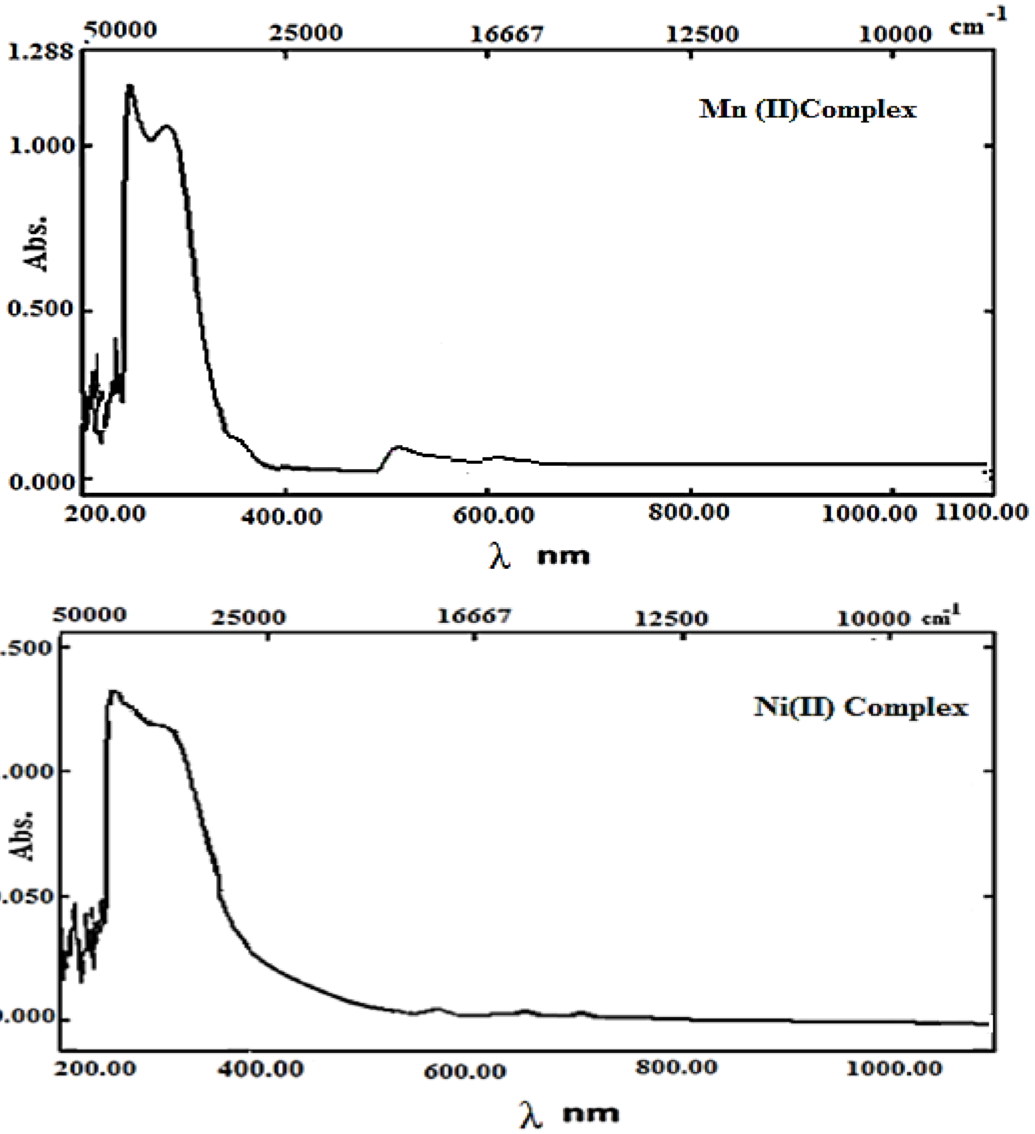
The absorption spectra of Ni(II) complex display five absorption peaks, The two peaks at (272, 372) nm attributed to the ligand field and (C. T), the last three d–d transition bands correspond to $^3A_2g(F)\rightarrow{}^3T_1g(P_)$, $^3A_2g(F)\rightarrow{}^3T_1g(F)$ and $^3A_2g(F)\rightarrow{}^3T_2g(F)$. The magnetic moments of Ni (II) complexes were found to be 2.45 BM supporting the d^8 high spin distorted octahedral structure. The absorption spectra shown in figure (5).

3.2.3. Cu-(Mef-B) $Cl_2.H_2O$ complex

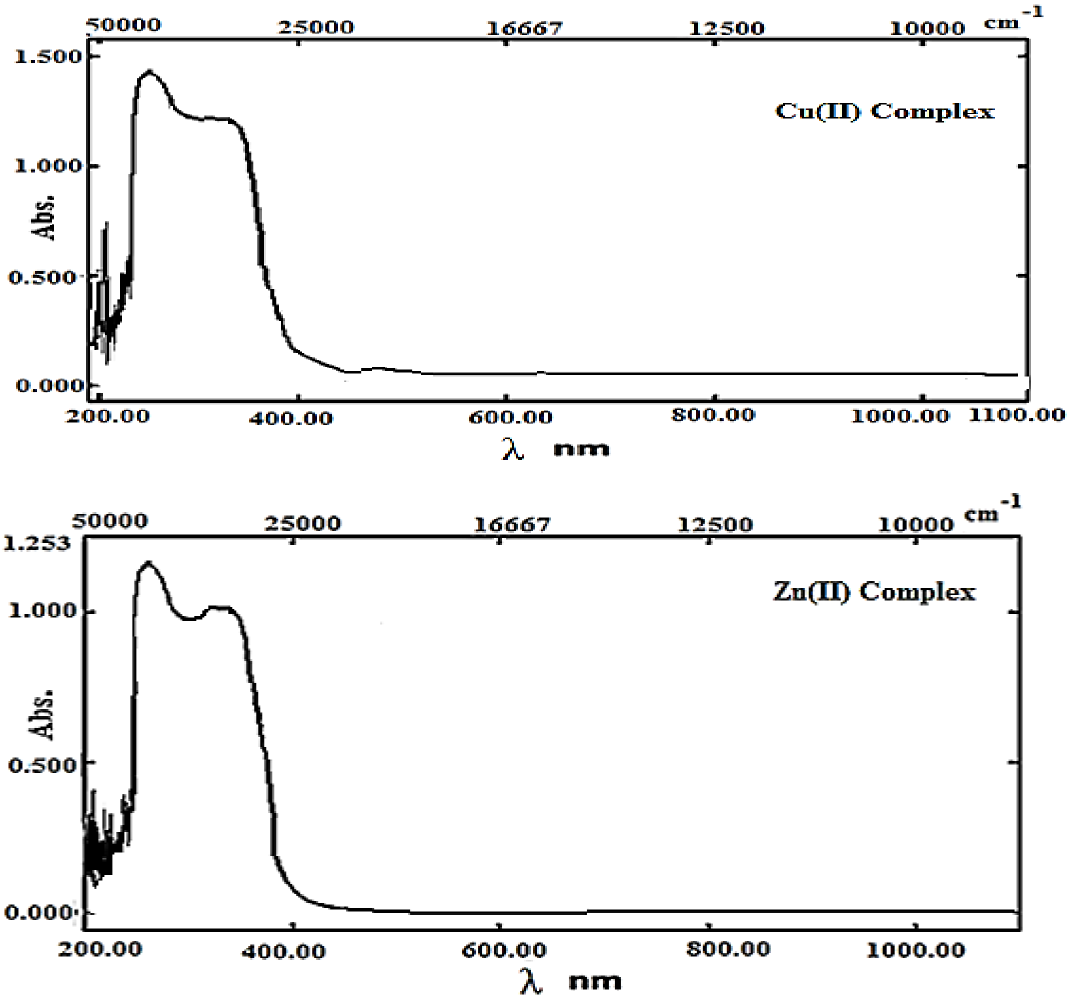
The absorption spectra of Cu(II) complex display three absorption peaks, The two peaks at (285, 378) nm attibuted to the ligand field and (C. T), the last peak attributed to d–d transition $^2Eg\rightarrow{}^2T_2g$. The magnetic moments of Cu(II) complex were found to be 1.82 BM also indicating octahedral geometry. The absorption spectra shown in figure (6).

3.2.4. Zn-(Mef-B) $Cl_2.H_2O$ complex

The absorption spectra of Zn(II) complex display two absorption peaks at (290, 383) nm attibuted to the ligand field and (C. T), the Zn(II) complex is diamagnetic. While the low conductivity values of the (Mef-B) metal complexes (13.04, 11.30, 16.05 and 18.20) respectively suggested that the complexes are non-electrolytes and that the chloride ion is still coordinated to the metal ion complex [14-16].



Figures - 5 : Electronic spectrum of Mn and Ni (Mef-B) complexes



Figures - 6 : Electronic spectrum of Cu and Zn (Mef-B) complexes

3.3 Antioxidant efficacy for (Mef-B) metal complexes by DPPH manner

The DPPH reacts with (Mef-B) metal complexes change in color from purple to yellow, after the reduction which can be measurement by their absorbance at wavelength (517) nm using UV–Vis spectrophotometer [17-27] , which shown in (Figures (7-10)). The following order of antioxidant activity: (Zn-(Mef-B) complex > Cu-(Mef-B) complex > Mn-(Mef-B) complex > Ni-(Mef-B) complex) at 30 minute, the Zn-(Mef-B) complex show more antioxidant activity at 30 minute, which shown in (Figures (11-14)). And (Figure -15 a,b : Antioxidant activity of the (Mef-B) metal complexes).

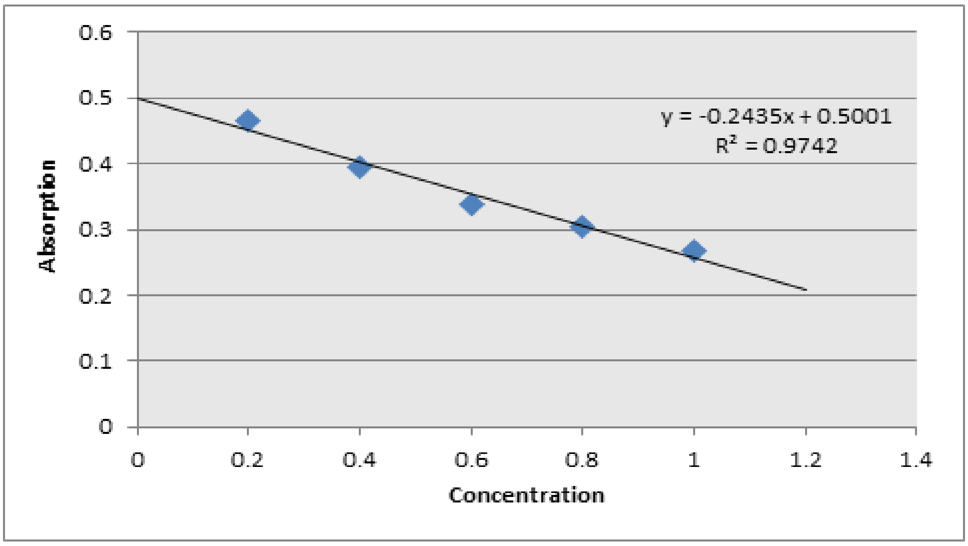


Figure - 7: Standard curve of Mn-(Mef-B) $Cl_2.H_2O$ complex

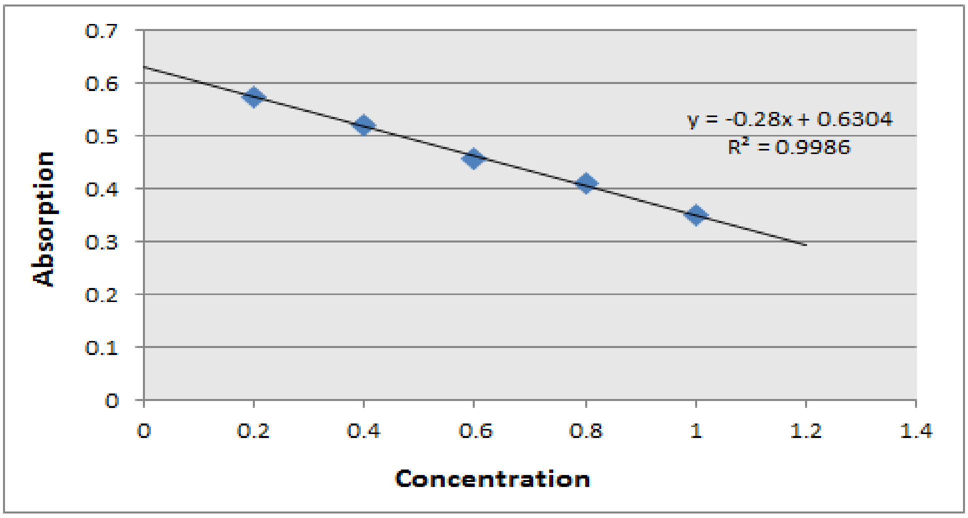


Figure - 8: Standard curve of Ni-(Mef-B) $Cl_2.H_2O$ complex

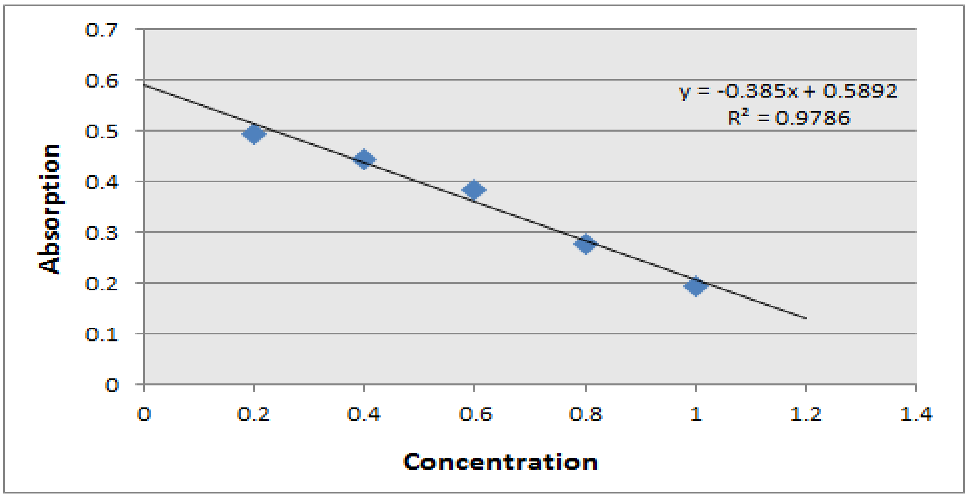


Figure -9: Standard curve of Cu-(Mef-B) $Cl_2.H_2O$ complex

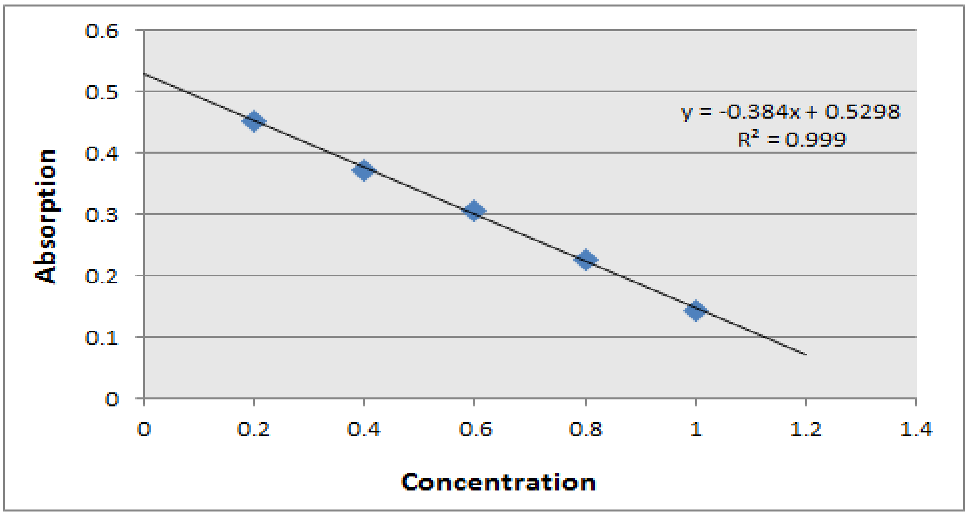
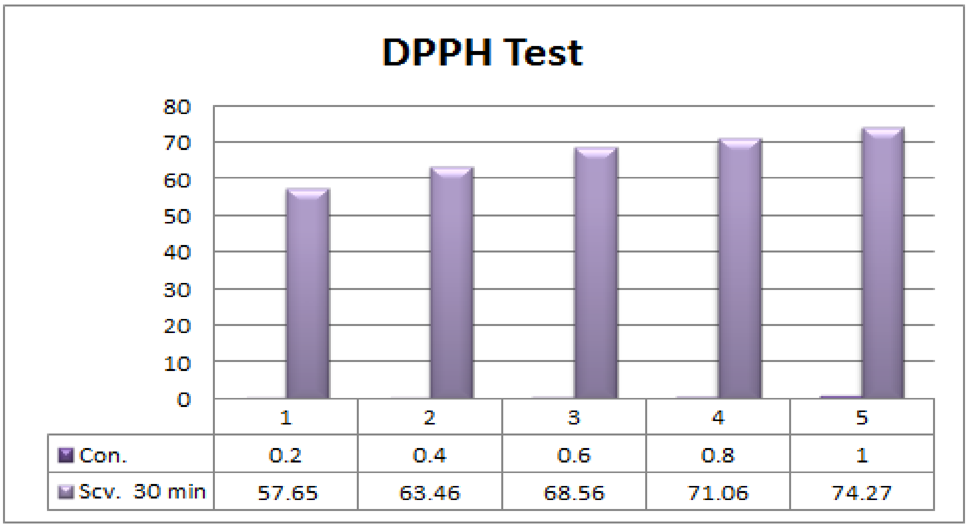


Figure -10: Standard curve of Zn-(Mef-B) $Cl_2.H_2O$ complex



	1	2	3	4	5
Con.	0.2	0.4	0.6	0.8	1
Scv. 30 min	57.65	63.46	68.56	71.06	74.27

Figure -11: DPPH Test of Mn-(Mef-B) $Cl_2.H_2O$ complex

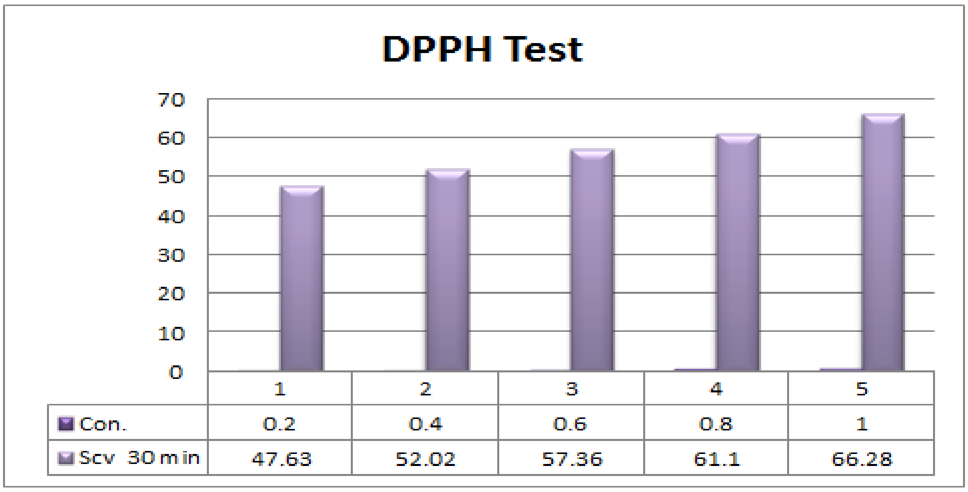


Figure - 12: DPPH Test of Ni-(Mef-B) $Cl_2.H_2O$ complex

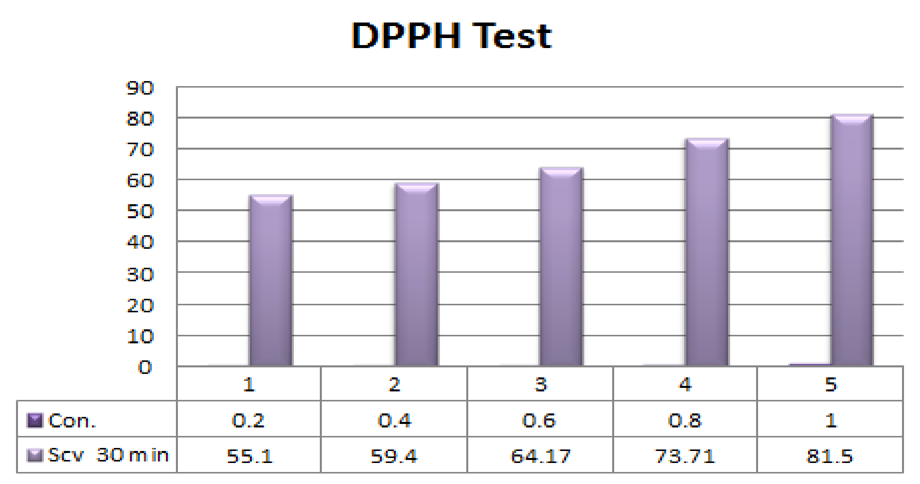
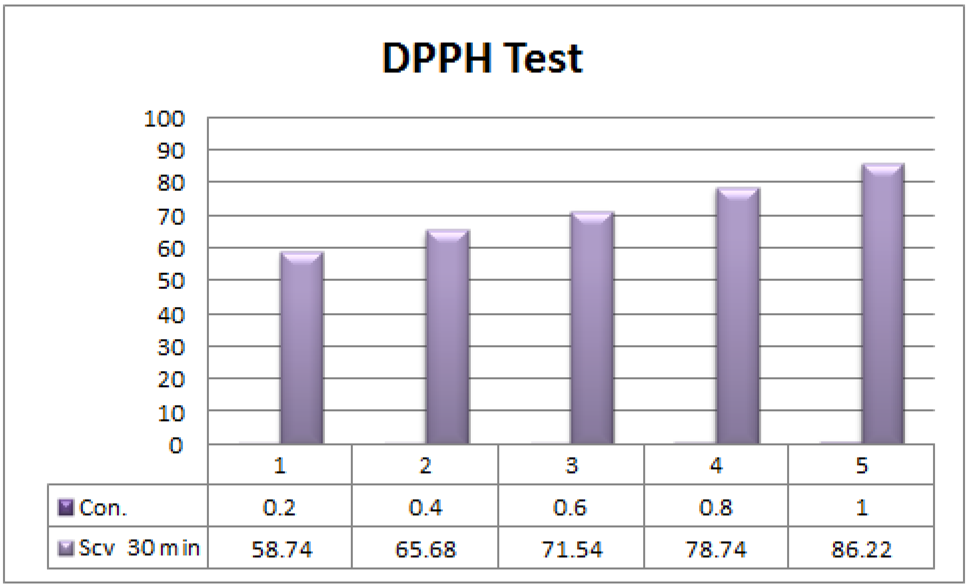
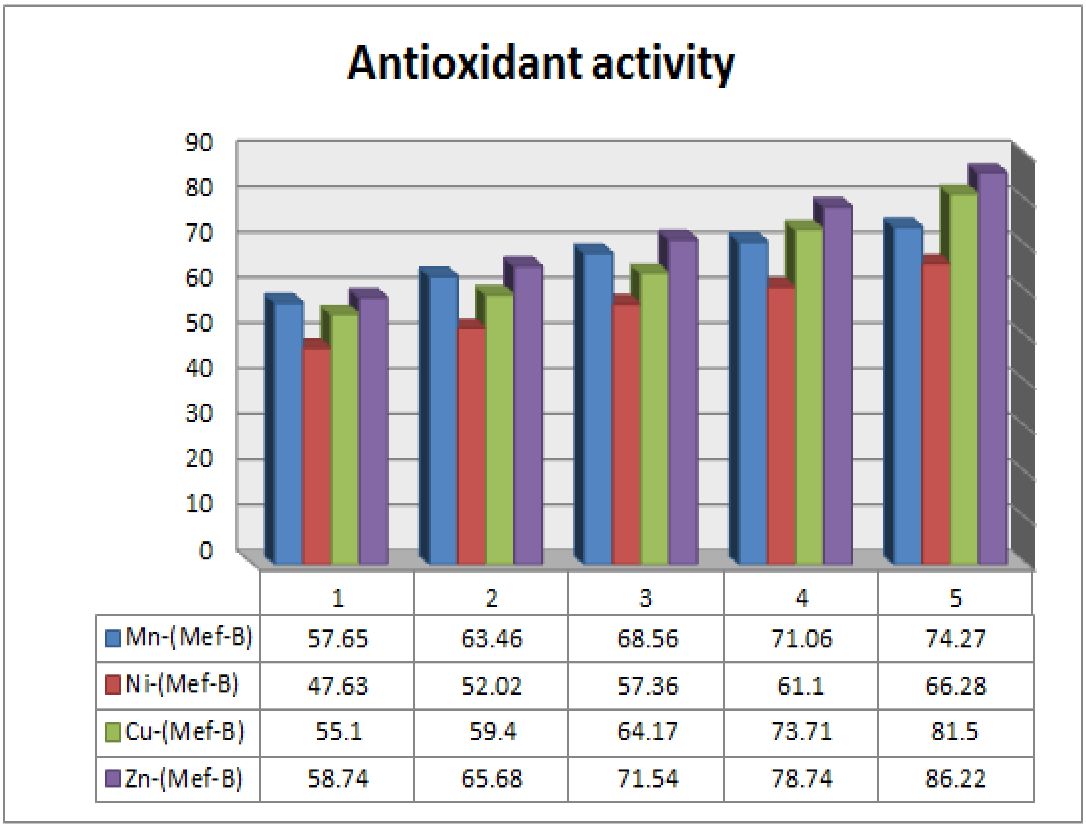


Figure - 13: DPPH Test of Cu-(Mef-B) $Cl_2.H_2O$ complex



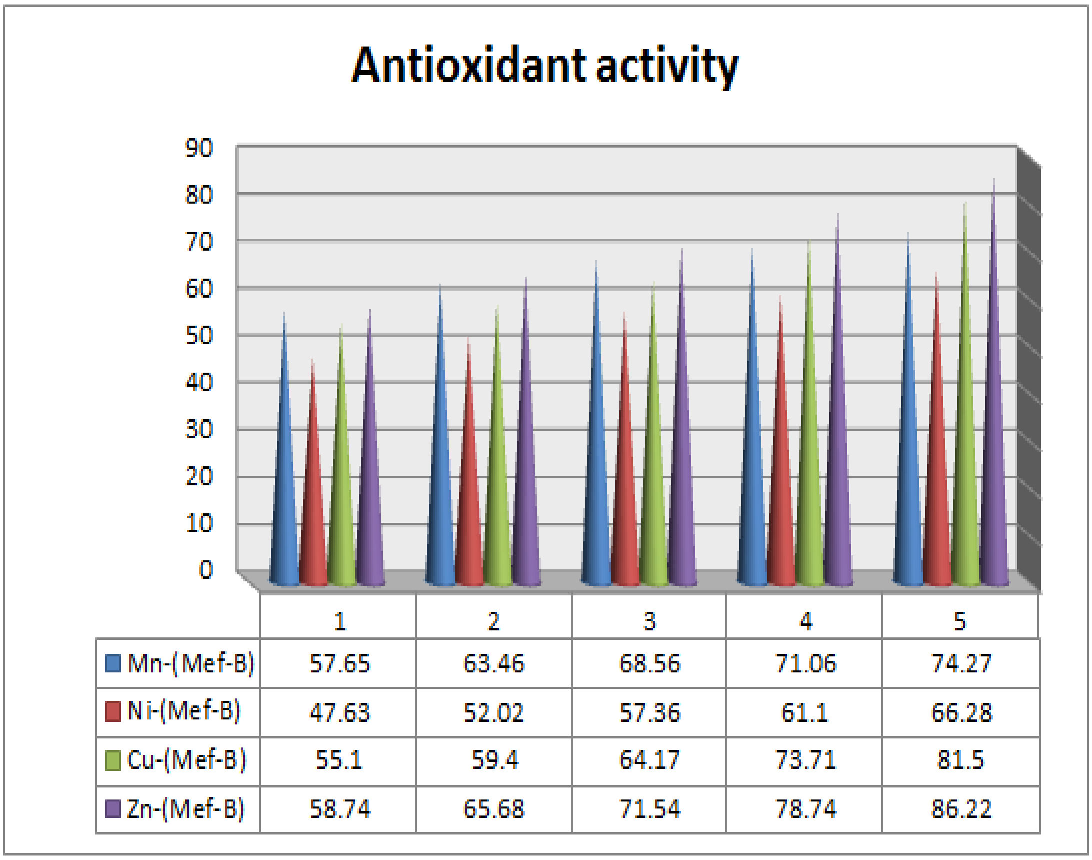
	1	2	3	4	5
Con.	0.2	0.4	0.6	0.8	1
Scv 30 min	58.74	65.68	71.54	78.74	86.22

Figure - 14: DPPH Test of Zn-(Mef-B) $Cl_2.H_2O$ complex



	1	2	3	4	5
🟦 Mn-(Mef-B)	57.65	63.46	68.56	71.06	74.27
🟥 Ni-(Mef-B)	47.63	52.02	57.36	61.1	66.28
🟩 Cu-(Mef-B)	55.1	59.4	64.17	73.71	81.5
🟪 Zn-(Mef-B)	58.74	65.68	71.54	78.74	86.22

Figure -15 a : Antioxidant activity of (Mef-B) metal complexes



	1	2	3	4	5
🟦 Mn-(Mef-B)	57.65	63.46	68.56	71.06	74.27
🟥 Ni-(Mef-B)	47.63	52.02	57.36	61.1	66.28
🟩 Cu-(Mef-B)	55.1	59.4	64.17	73.71	81.5
🟪 Zn-(Mef-B)	58.74	65.68	71.54	78.74	86.22

Figure -15 b: Antioxidant activity of (Mef-B) metal complexes

4. CONCLUSION

In this study, new (Mef-B) metal complexes for [Mn(II), Ni(II) , Cu(II) and Zn(II)] were prepared. The mode of bonding and overall structure of the complexes were determined through physico-chemical and spectroscopic methods. The structure of the complexes based on Uv-vis, FT-IR spectra were proposed that the geometry of all metal complexes is octahedral geometry. The Zn-(Mef-B) complex show more antioxidant activity at 30 minute.

References:

1. Lugemwa FN, Snyder AL, Shaikh K. Determination of radical scavenging activity and total phenols of wine and spices: A randomized study. Antioxidants. 2013 Sep;2(3):110-121.
2. Verma DK, Maurya SK. Antioxidant activity of Rajatachandrodaya rasa. International Journal of Green Pharmacy. 2018; 5(11):69-71.

3. Balasubramanian K, Ragunathan R. Study of antioxidant and anticancer activity of natural sources. J. Nat. Prod. Plant Resour. 2012;2(1):192-197.

4. Kumar S. Free radicals and antioxidants: human and food system. Adv Appl Sci Res. 2011 Feb;2(1):129-135.

5. Medini F, Fellah H, Ksouri R, Abdelly C. Total phenolic, flavonoid and tannin contents and antioxidant and antimicrobial activities of organic extracts of shoots of the plant Limonium delicatulum. Journal of Taibah University for science. 2014 Jul 1;8(3):216-224.

6. Shebis Y, Iluz D, Kinel-Tahan Y, Dubinsky Z, Yehoshua Y. Natural antioxidants: function and sources. Food and Nutrition Sciences, 2013; 4(1):643-49.

7. Arman MS, Al Mahmud A, Mahmud HR, Reza AA. Free radical, oxidative stress and diabetes mellitus: A mini review. Discovery Phytomedicine. 2019;6(3):99-101.

8. Moharram HA, Youssef MM. Methods for determining the antioxidant activity: a review. Alex. J. Fd. Sci. & Technol. 2014;11(1):31-42.

9. Dehariya R, Dixit AK. A review on different methods of determination of antioxidant activity assay of herbai plants. Life science informatics publications, 2018.4 (6);708.

10. Kedare SB, Singh RP. Genesis and development of DPPH method of antioxidant assay. Journal of food science and technology. 2011 Aug 1;48(4):412-422.

11. Alam MN, Bristi NJ, Rafiquzzaman M. Review on in vivo and in vitro methods evaluation of antioxidant activity. Saudi pharmaceutical journal. 2013 Apr 1;21(2):143-52.

12. Silverstein R. M , Webster E. X, Kiley D. J . Spectrometric identification of organic compounds 7th ed , John Wilye and SonsS. INC. 2005, 1-550.

13. Figgis BN. Ligand field theory. Comprehensive Coordination Chemistry. 1987;1:213-79.

14. Lever AB. Inorganic Electronic Spectroscopy .Elsevier Publishing Company, Amsterdam, Londen. 1968.

15. Lever AB. Electronic spectra of some transition metal complexes. Journal of Chemical Education. 1968 Nov;45(11):711.

16. Figgis BN. Introduction to ligand fields. Inter science publishers; 1966.

17. Kumara P, Sunil K, Arun Kumar B. Determination of DPPH Free Radical Scavenging Activity by RP-HPLC, Rapid Sensitive Method for the Screening of Berry Fruit Juice Freeze Dried Extract. Nat Prod Chem Res. 2018;6:341.

18. Velho-Pereira S, Parvatkar P, Furtado IJ. Evaluation of antioxidant producing potential of halophilic bacterial bionts from marine invertebrates. Indian Journal of Pharmaceutical Sciences. 2015 Mar;77(2):183.

19. Feghhi-Najafabadi S, Safaeian L, Zolfaghari B. In vitro antioxidant effects of different extracts obtained from the leaves and seeds of Allium ampeloprasum subsp. persicum. Journal of Herbmed Pharmacology. 2019 May 9;8(3):256-60.

20. Horozić E, Suljagić J, Suljkanovic M. Synthesis, characterization, antioxidant and antimicrobial activity of Copper (II) complex with Schiff base derived from 2, 2-dihydroxyindane-1, 3-dione and Tryptophan. American Journal of Organic Chemistry. 2019;9(1):9-13.

21. Fidrianny I, Octaviani GD, Kusmardiyani S. Study of Antioxidant Profile and Phytochemical Content of Different Organs Extracts of Morinda citrifolia L. Journal of Pharmaceutical Sciences and Research. 2018 Aug 1;10(8):2102-5.

22. Aadesariya MK, Ram VR, Dave PN. Evaluation of antioxidant activities by use of various extracts from Abutilon pannosum and Grewia tenax in the Kachchh Region. International Journal of Research in Engineering and Innovation. 2017;1(3):97-112.

23. Saha S, Islam Z, Islam S, Hassan MF, Hossain M, Islam SM. Determination of Antioxidant Properties and the Bioactive Compounds in Wheat (Triticum aestivum L.). Jordan Journal of Biological Sciences. 2018 Sep 1;11(3).

24. Jimoh MO, Afolayan AJ, Lewu FB. Antioxidant and phytochemical activities of Amaranthus caudatus L. harvested from different soils at various growth stages. Scientific Reports. 2019 Sep 10;9(1):1-4.

25. Djenidi H, Khennouf S, Bouaziz A. Antioxidant activity and phenolic content of commonly consumed fruits and vegetables in Algeria. Progress in Nutrition. 2020 Mar 1;22(1):224-35.

26. Jirjees VY, Suleman VT, Ahmed SD, AL-Hamdani AA. Determination of Antioxidant Activity for Metal Ions Complexes. Journal of Duhok University. 2020 Jun 7; 23(1):41-50.

27. Aly SA, Fathalla SK. Preparation, characterization of some transition metal complexes of hydrazone derivatives and their antibacterial and antioxidant activities. Arabian Journal of Chemistry. 2020 Feb 1;13(2):3735-50.