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# Synthesis, Characterization and Antifungal Studies of Some Metronidazole Complexes

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**ABSTRACT**: Some transition metal complexes of metronidazole (met) have been prepared from the metal halide salt and the ligand at mole ratio 1:2. These complexes have been characterized by vibrational, electronics, HNMR and Atomic absorption spectroscopy. The antifungal activities of the ligand and its complexes are also described. The analysis of the spectroscopic data suggests that the ligand is bidentate in its mode of coordination and the structure of the complex is four coordinate. @ JASEM

**Keywords:** Metronidazole-complexes, Antifungal studies.

Metronidazole (MTN) is one of the group of clinically significant heterocyclic radiosensitizing drugs (Callaghan, 1983).

Several metal chelates are known to possess antibacterial, antifungicidal, antiviral and anticancer activity. In several cases, the metal chelates have been found to be more antimicrobial than the chalating agents themselves (Srivastava, 1981). Also it is known that some drugs act via chelation or by inhibitory metalloenzymes but for most of the drugs that act as potential ligands, a lot of studies are being carried out to ascertain how metal binding influences the activities of the drugs (Behren et al, 1986).

However, metal ions play an important role in bioinorganic chemistry and metals such as Fe, Co, Ni, Cu, Zn, and Cd may exist in trace amounts in biological systems. Structural studies of the complexes of these metals with biological compounds are extremely important (Canpolat et al, 2004).

Although a few paper have been published on the transition metal complexes of related ligands (Hwang and Wang, 1992; Mendiola and Mesaguer, 1992). There is relatively little work reported on the metal complexes of metronidazole, however, Callaghan, 1983 reported platinum and copper complexes of metronidazole and 2-methyl–5-nitrobenzimidazole. The two ligands form tetrahedral complexes of the type $ML_2X_2$.

It was therefore considered necessary to prepare transition metal complexes of metronidazole and to investigate their antifungal activities. It is hope that the novel complexes will have biological potency than the original metronidazole.

## EXPERIMENTAL

## MATERIALS AND METHODS

All materials (Metal salts and solvents) were of standard analytical grade and were used as supplied, without further purification.

The ligand, metronidazole was obtained from Sam Pharmaceutical limited, Ilorin, Nigeria.

The thin layer Chromatograph was run using a solvent mixture of ethanol and acetone in the 9:1 mole ratio for the metal complexes. The metal contents analyses were carried out using an SP9 Atomic Absorption spectrophotometer with PM8251 simple-pen recorder. The infrared spectra of the ligand, metronidazole (Met) and its complexes were recorded in solid state as KBr pellets from 4000 – 600$cm^{-1}$. The uv/visible spectral of the ligand, metronidazole (Met) and its complexes were recorded on SP8-400 uv/visible spectrophotometer, using methanol as solvent.

The proton Nmr spectra of the ligand, Metronidazole (Met) and its complexes were obtained on Brucer AC 200/300 spectrometer using deuterated chloroform as solvent.

## ANTIFIGUAL ACTIVITY TEST

The antifungal activities test of the compounds was assayed against three fungi: *Aspergillus niger Aspergillus flavus* and *Rhizopus species*. All media and fungal suspension were prepared using previous described methods (as reported by Turet, 2003). The antifungal activity was determined on the potato dextrose agar. The fungal cultures were incubated at $30^{O}$C for 48 hours. The agar was allowed to solidify. Three cups of 10mm in diameter and 5mm deep were then removed from each agar dish. Fresh fungal suspension was spread on each cup. Each of he cups was spotted three times with test solution at concentration of 0.1% and 1.0%.

The diameter experiment was conducted in triplicate. The percentage inhibition was calculated as follows.

Where B = average diameter of fungal growth of on the test sample

A = average diameter of fungal growth without test sample

## SYNTHESIS OF METAL COMPLEXES

The synthesis procedure was carried out using

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standard procedures as detail elsewhere (Nadira, 1987, Galvan–Tejada, 2002) in which ethanolic solutions of the metronidazole (Met) and metal salts were prepared separately. Metronidazole (3.424g, 0.02M) in ethanol (20ml) was added to the hydrated metal salt (0.01M) in ethanol (10ml) in a round-bottomed flask fitted with a condenser and refluxed for 2hours. It was then cooled and the resulting precipitate filtered. The precipitate formed was recrystallized from ethanol and dry in a dessicator.

**RESULT AND DISSCUSION**

The analytical data and other physical properties of the complexes are presented in Table 1: The formation of the complexes undergo the following reactions scheme.

$MX_2.nH_2O + 2Met \rightarrow M(Met)_2X_2 + nH_2O$ *[M = Co(II).Ni(II)]*

$MX_3.nH_2O + 2Met \rightarrow M(Met)_2X_3 + nH_2O$ *[M = Fe(III)]*

$Met = C_6H_9N_3O_3$

All the complexes synthesized are coloured, from creamy to black. The molar conductance data (Table 7) for metal complexes in methanol indicate non-electrolytic behaviour in this solvent.

The retention factor values calculated from the developed single spot for the complexes indicate the purity of the compounds.

The uv spectral assignment of Metronidazole (Met) and its complexes are also presented in table 1. The uv spectra bands of the ligand and its complexes have been assigned tentatively to charge transfer transition from the metals to the anti-bonding orbital of the ligand. The $\prod \rightarrow \prod^*$ transitions of the ligand are also observed (Orgel, 1985; Williams and Fleming, 1980). The ultraviolet spectrum of the free metronidazole shows four absorption bands at 314mn, 262nm, 227nm and 198nm. These bands undergo hypsochromic shifts in the metal complexes due to complexation.

The infrared spectra of the free metronidazole and its complexes are presented in Table 2. The spectra show similar bands, due to the presence of the same ligand in all the complexes. Though, the spectrum of the free metronidazole was compared with the spectra of its complexes. The strong absorption bands at 3780.1cm$^{-1}$ and 3707.1cm$^{-1}$ in the spectrum of the Metronidazole have been attributed to O –H stretchings (Williams kemp (1991). These bands have been shifted in the spectra of the complexes. The shifting of these (O-H) stretching vibration bands provided evidence that this group is one of the coordination site of Metronidazole. The spectrum of metronidazole also shows one absorption band at 1615cm$^{-1}$ attributed to aromatic C=C and C=N stretchings. This band still appears in the spectra of the metal complexes, with slight change in intensity.

A strong absorption peak at 1570cm$^{-1}$ and 1360cm$^{-1}$ in the spectrum of Metronidazole have been assigned to N=O stretchings. This band still appears in the spectra of the metal complexes with changes in intensity.

The medium broad bands at 1129cm$^{-1}$ and 958.9cm$^{-1}$ have been assigned to C – N stretching and C- N bending respectively (William and Fleming, 1980; William 1991).

The weak absorption bands observed at 674.4cm$^{-1}$ in $Co(Met)_2Cl_2$ and 748.7cm-1 in $Ni(Met)_2Cl_2$ which could not be traced metronidazole, have been tentatively assigned to M-OH bands of these metal complexes (Lever, 1984).

The antifungal activities of the metronidazole and it complexes are presented in Table 4. The results show that metronidazole complexes are active against all the fungi species. Thus the antifungal effect varies with the type of organism. Generally all the complexes have greater inhibitory activity on the organism compared to its free metronidazole (Ali, 1992).

The free metal salts ($NiCl_2.6H_2O$, $CoCl_2.6H_2O$ and $FeCl_3.6H_2O$) have been reported to posses absolutely no inhibitory activity on the selected fungal species (Obaleye, 1999).

By taking into consideration all the above analytical data, spectroscopic data and physical properties. The following structure is tentatively proposed for the metal complex.

M = Co(II), Ni(II), n = 2
M = Fe(III), n = 3

Fig 1: The proposed structure for the Metal Complexes.

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Table 1: Physical properties and analytical data of the metronidazole (met) and its complexes

| Metronidazole ($cm^{-1}$) | $Co(Met)_2Cl_2$ ($cm^{-1}$) | $Ni(Met)_2Cl_2$ ($cm^{-1}$) | $Fe(Met)_2Cl_3$ ($cm^{-1}$) | Tentative assignment |
|---|---|---|---|---|
| 3780.1s,b<br>3707.0v.s | 3779.1s,b<br>3713.0v,s | 3788.6s,b<br>3703.4v,s | 3791.0s,b<br>3688.8v,s | O – H |
| 1615.6s,b | 1679.3s<br>1661.Os,b | 1666.6w,b<br>1496.3w,b | 1661.2w | C = C &<br>C = N |
| 1570.0s<br>1300.0s | 1560.0m<br>1360.3m | 1581.6s,b<br>1374.7w,b | 1564.8s<br>1522.7w,b | N = O str. |
| 1238.9m,b | 1263.9s,b | 1271.4v,s | 1203.82 | C = C |
| 1129.2m | 1191.8m,b | 1192.3v,s | - | C – N str. |
| 1074.6w,s | 1077.6s,b | 1083.1s | 1077.6s,b | C – O str |
| 958.9w,b | 969.2s | 973.6s<br>918.7w,b | 957.22s | C – N bending |
| | 674.4w,b | 748.7w,b | 842.8s,b | M – OH |

Table 2: IR Spectra Assignment of Metronidazole and its metal complexes

| Compound | M.P ($^OC$) | Colour | % Yield | %M | □m | Retention Factor | Electronic Transition (nm) |
|---|---|---|---|---|---|---|---|
| Met | 158 - 160 | White creamy | - | - | $2.1 \times 10^{-7}$ | 0.80 | 314,262,277,198 |
| $Co(Met)_2Cl_2$ | 150 - 152 | Creamy | 67.8 | 11.76 (11.20) | $2.32 \times 1^{-7}$ | 0.76 | 301,258,222,194 |
| $Ni(Met)_2Cl_2$ | 152 - 154 | Light green | 59.0 | 12.75 (12.48) | $1.64 \times 10^{-6}$ | 0.82 | 302,258,222,194 |
| $Fe(Met)_2Cl_3$ | 148 - 150 | Black | 50.1 | 9.84 (9.58) | $0.93 \times 10^{-6}$ | 0.78 | 306,257,218 |

Table 3: Hnmr spetral Assignment of Metronidazole and its metal complexes

| Metronidazole ppm | $Co(Met)_2Cl_2$ | $Ni(Met)_2Cl_2$ | $Fe(Met)_2Cl_3$ | Assignment |
|---|---|---|---|---|
| 7.91 (1H,S) | 8.01 (1H,S) | 7.92 (1H,S) | 7.95 (1H,S) | E |
| 7.24 | 7.23 | 7.24 | 7.24 | Solvent peak |
| 4.48-4.45 (2H,t) | 4.48-4.45 (2H,t) | 4.48-4.45 (2H,t) | 4.47-4.45(2H,t) | C |
| 4.01-3,96 (2H,t) | 4.00-3,95 (2H.t) | 4.00- 3.95 (2H,t) | 3.98 (2H,t) | B |
| 2.52 (3H,S) | 2.50 (3H,S) | 2.52 (3H,S) | 2.53 (3H,S) | A |
| 2.08-2.05 (1H,t) | 2.07-2.03 (1H,t) | 1.97 – 1.94 (1H,t) | 1.69 (1H,t) | D |
| 1.57 | 1.57 | 1.56 | 1.55 | Solvent peak |

Table 4: Result of antifungal test for Metronidazole and its metal complexes at different concentration

| **Concentration** | **Compound** | ***A. Niger*** | ***A. flavus*** | ***Rhizopus*** |
|---|---|---|---|---|
| 0.1% | Met | 0 0 0 | 0 0 0 | 0 0 0 |
| | $Co(Met)_2Cl_2$ | 28 30 31 | 22 21 21 | 20 18 18 |
| | $Ni(Met)_2Cl_2$ | 14 14 15 | 35 34 35 | 32 33 32 |
| | $Fe(Met)_2Cl_3$ | 18 17 17 | 38 38 36 | 40 37 38 |
| 1.0% | Met | 24 26 23 | 47 48 48 | 45 45 46 |
| | $Co(Met)_2Cl_2$ | 60 65 69 | 80 85 85 | 95 95 98 |
| | $Ni(Met)_2Cl_2$ | 40 42 44 | 98 92 90 | 78 78 80 |
| | $Fe(Met)_2Cl_3$ | 42 40 40 | 80 81 81 | 81 82 88 |

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## REFERENCES

Ali, M A; Harron, C M; Nazimuddim, M; Mahbud-ul-Hazue, SM; Tarafeler, SM.; Khair, MA, (1992) Transition Met. Chem., 1, 133 – 136.

Behrens, N B; Diaz, GM; Goodgame DML; (1986) Metal complexes of the antibiotics Nalidixic acid. Inorg. Chim. Acta, 125, 21-26.

Callaghan, V; David, M.J; Goodgame L; Robert, R T; (1983) Platinum and other metal complexes of 2-methyl-5-nitrobenzimidazole. Inorg. Chim. Acta, 78, L1-L4.

Canpolat, E; Kaya, M; Gur, S; (2004) Synthesis, Characterization of Some Co(III) Complexes with Vic-Dioxime ligands and their antimicrobial properties. Turk. J. Chem. 28, 235 – 242.

Galvan–Tejada, N; Bernes, S; Castillo-Blum, S E; Noth, IT; Vicente, R. and Barba-Behrens, N

*Corresponding author: LAWAL, A

(2002) supramolecular structure of metronidazole and its copper(II) coordination compound J. Inorg. Biochem, 25, 91(1), 399-480.

Hwang, CC; Su and Wang, OYP (1992) Transition Met. Chem. 17, 91.

Issa, A; (2002) M.Sc. Thesis, Dept. of Chemistry, University of Ilorin, Ilorin, Nigeria, P. 46-64.

Lever, ABP (1984) Inorganic Electronic Spetroscopy, 2nd edn, Elsevier, Amsterdam.

Mendiola, MA; Masaguer, JR; Molloida, C (1992) Iron (II) Chloride Complexes with some 2-amino benzimidazole ligands. Synth. React. Inorg. Met. Org. 22 (77) 955-969.

Nadira, W; Singh, H B (1987) Sythesis of metal complexes of Antimalarial drugs and in vitro Evaluation of their activity against plasmodium falciparum, Inorg. Chim. Acta, 136, 133-137.

Obaleye, J A; Balogun, E A; Adeyemi, O G; (1999) Synthesis and in vivo effects of some metal – drug complexes on malaria parasites, Biokemistri, Vol. 9, pp. 23-27.

Orgel, K E (1985) An introduction to transition metal chemistry ligand filed theory, John Wiley & Sons, New York, pp. 18 -22.

Srivastava, R S; (1981) Synthesis, Characterization and fungitoxicity of Bidenntate High-spin six coordinate 3d metal complexes with N-(5-phenyl-3-4-thiadiazol – 2-yl) acetabenzamidine. Inorg. Chim. Acta, 55 L71–L74.

Turel, I; Golobiga, A; Klavzar, A; Phihlar, B; Buglyo, P; Tohse, E; Rehder, D; Sepele, K (2003) interaction of oxovanadium (IV) and the quinolone family member–ciprofloxacin, J. Inorg. Biochem, vol 1, pp1

Williams, H D; Fleming, I. (1980) Spectroscopic methods in Organic Chemistry, 4th Ed, McGraw-Hill Book Company Limited, London, pp. 41-45.

William, K, (1991) Organic Spectroscopy, 3rd Ed., Macmillan Education Ltd., London, pp. 49-54, 60-75, 121-137.

*Corresponding author: LAWAL, A