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# Synthesis and Characterization of Schiff Base of 2-Acetyl-5-Methylfuran Glyoxime Hydrazine and its Metal (II) Complexes and Their Evalution for Antibacterial and Antifungal Activities

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**Abstract:** Two hydrazone Schiff base was synthesized by the interaction of ethanolic solution of antiglyoxime hydrazine with 2-acetyl-5-methylfuran to obtain the Schiff base. The complexes of Mn(II), Fe(II), Ni(II) and Cu(II) and their Schiff base were synthesized and characterized based on solubility, percentage yield, elemental analysis, melting point and decomposition temperature, infra-red spectra (FT-IR), conductivity measurement, magnetic susceptibility and water of crystallisation. The ligand and the complexes were coloured and stable at room temperature. They were soluble in acetone, DMSO, ethanol, methanol, hexane, chloroform, carbontetrachloride, benzene and DMF but insoluble in distilled water. The melting point temperature of the ligand was 118$^{o}$C while the decomposition temperatures for the complexes were found to be between 295-312$^{o}$C. The molar magnetic susceptibility of the complexes were in the range of 3.55 x $10^{-3}$ – 1.42 x $10^{-2}$erg.G$^{-2}$mol$^{-1}$. Molar conductance values of 2.17 – 2.87$\Omega^{-1}$cm$^{2}$mol$^{-1}$ show non electrolytic nature of the complexes. The spectral data of the ligand showed bands in the range 1581–1607cm$^{-1}$ revealing the formation of the azomethine group. A band between 760-795cm$^{-1}$ indicates the metal nitrogen bonds. The elemental analysis of the complexes showed the metal ligand ratio of 1:2 (M:2L). The antibacterial and antifungal studies showed that the complexes exhibited higher antibacterial and antifungal activities than the ligand but lower than the reference drugs.

**Keywords:** Substityted furan, glyoxime metal(II) complexes and microbial activity.

**Research Paper**

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## INTRODUCTION

Schiff bases are condensation products of primary amines with aldehydes or ketones they were first reported by Hugo Schiff in 1864. The most common structural feature of these compounds is the azomethine group (C=N) with the general formula RHC=N-R' where R and R' are alkyl, aryl, cyclo or heterocyclic groups which may be variously substituted, these compounds are also known as anils, imines, or azomethines [1, 2]. Schiff base metal complexes have been widely studied because they have industrial, antibacterial, antifungal, antiviral, anticancer, anticonvulsant and herbicidal applications [3, 4]. Some complexes containing nitrogen and oxygen donor atoms in their structures are effective as stereospecific catalysts for oxidation, reduction, hydrolysis, biological activity and other transformation of organic and inorganic chemistry [5-8]. Schiff base metal complexes exhibit a large number of biological activities which include antibacterial, antifungal, antimalarial, antiproliferative, antiinflamatory, antiviral and antipyretic activities, they also have industrial applications [9, 10].

Hydrazones are also a class of Schiff base containing C=NNH$_2$, as their functional group, they are formed by the condensation of substituted hydrazine with aldehyde or ketone [11, 12]. Hydrazone Schiff bases contain two connected nitrogen atoms of different nature and a C=N double bond that in conjugated with alone electron pair of the terminal nitrogen atom. These structural fragments are principally responsible for the physical and chemical properties of hydrazone Schiff bases [13, 14]. Hydrazones also constitute an important class of biologically active drugs molecules which has attracted attention of medical chemists due to their antimicrobial, antihypertensive, analgesic, anti-inflammatory, anti-tuberculosis, antiproliferative and antimalarial activities [15, 16].

Hydrazone is a versatile moiety that exhibits a wide variety of biological activities [17, 18]. Hydrazone nucleus exhibited immense pharmacological activities.

Hydrazone-based coupling methods are used in medical biotechnology as anticonvulsant, antidepressant, analgesic, anti-inflammatory, antiplatelet, antimalarial, antimicrobial, anticancer, vasodilator, antiviral, anti-HIV, antidiabetic and trypanocidal activities [19].

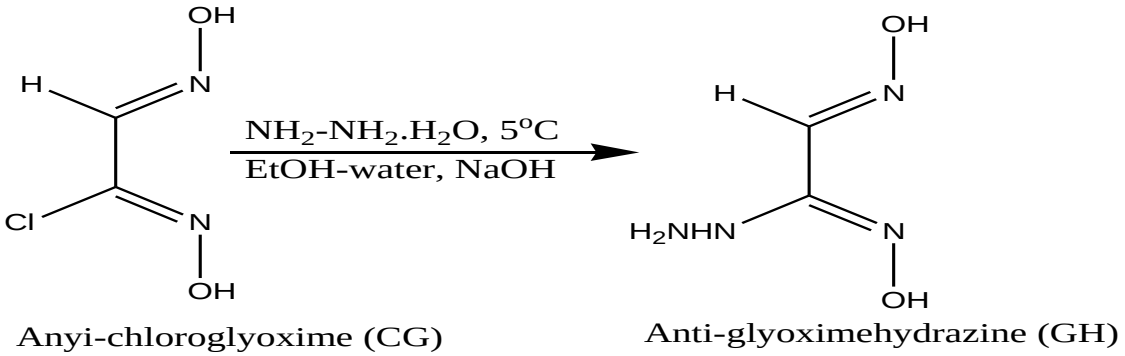
## MATERIALS AND METHODS

**Chemicals, Reagents and Apparatus**

All glass wares used in this work were properly washed with detergent, rinsed several times with tap water. They were then soaked in a concentrated solution of nitric acid for about 2 – 3 hrs after which they were rinsed 3 – 4 times with distilled water and dried in an oven maintained at 110$^{o}$C. All reagents and solvents used were of Analar grade and were used as supplied without further purification. All weighing were carried out on College B154 Metler Toledo electric balance. Melting point and decomposition temperatures were carried out on Stuart SMP 10 melting point apparatus. Determination of water of hydration was done on drying oven model DHO – 10053A. FT-IR spectra measurements were recorded using Agilent Technologies FT-IR spectrophotometer Carry 630, in the region of 400–4000cm$^{-1}$. The metal content of each complex was determined using Atomic Absorption Spectrophotometer (AAS), Bulk Scientific VGP 210. The elemental analysis (CHNS) was carried out at OEA labs., Callington, UK using a CE instruments (thermo) EA1110 Elemental analyser. The electrical conductivity measurements were also carried out using conductivity meter DDS-307, Jenway. The magnetic susceptibility measurements were carried out using magnetic measurement balance Sherwood scientific MK 01 model at room temperature. Three bacterial isolates were used in the test of microbial activity viz: *Salmonella typhimurium Proteus aureginosa* and *Staphylococcus aureus*, and two fungal isolates *viz: Candida utilis and Saccharomyces cerevisiae.*

**Synthesis of intermediate Glyoximehydrazine**

A solution of NaOH (0.4g, 10mmol) in 1cm$^3$ of distilled water was mixed with 10cm$^3$ of ethanol and 0.6cm$^3$ of hydrazinium hydroxide and cooled to 5$^{o}$C. A solution of chloroglyoxime (1.225g, 10mmol) in 5cm$^3$ of ethanol was added drop wise with stirring into the prepared mixture maintained at 5$^{o}$C. Stirring was continued for 15min at the same temperature to complete the reaction. The precipitate that formed was filtered, washed with cold ethanol and dried in a desiccator over phosphorus pentaoxide.

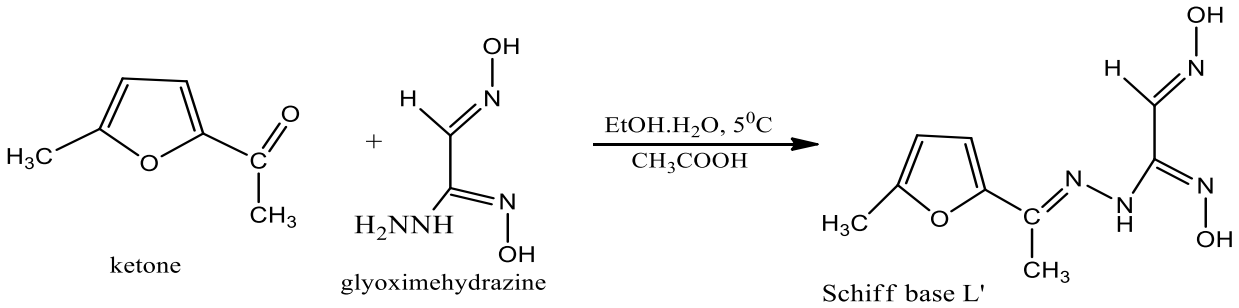



**Scheme 2.1: Preparation of glyoximehydrazine (GH)**

**Preparation of 2-acetyl-5-methylfuran glyoxime hydrazone**

A cooled solution 5$^{0}$C of 2-acetyl-5-methylfuran (1mmol) in ethanol was added drop wise into a cooled solution 5$^{0}$C containing 1mmol of glyoximehydrazine and 3 to 5 drops of acetic acid with constant stirring. After the addition of the substituted furan was completed, the solution was stirred for 4 to 6 hours at room temperature. The resulting precipitate (L′), was separated, washed with water, ethanol and dried over phosphorus pentaoxide in a desiccator.

The equation for the reaction is presented below:




**Scheme 2.2: Preparation of the Schiff base L**

***Synthesis of Metal(II) Complexes***

A solution of Mn(II), Fe(II), Ni(II) and Cu(II) chlorides (1mmol) in 20cm$^3$ of distilled water were added to 2mmol of the ligand in 15cm$^3$ of ethanol with constant stirring. An initial sharp decrease of pH of the solution from 5.5 to 3.5 was observed. After raising the pH to 5.5 using aqueous NaOH solution, the reaction mixture was kept in a hot water bath (60$^o$C) for 2 to 3 hrs to complete the precipitation. Then the precipitated complexes were filtered, washed with water and dried in a desiccator over phosphorus pentaoxide.

***Antibacterial Activity Test***

Antibacterial activity of the Schiff base and its complexes were tested using disc diffusion method on Mueller Hinton agar (MHA) and potato dextrose agar (PDA) sterile plates. The ligand and its complexes were dispensed into separately labelled wells using sterile 1mL insulin syringe. The preparations were incubated at 37$^O$C for 24 hrs. Mean zones of inhibitions were measured using metre rule and recorded in millimetre [18]. Three bacterial isolates of which three viz: *Salmonella typhimurium Proteus aureginosa* and *Staphylococcus aureus*.

***Antifungal Activity Test***

Two fungal isolates *viz: Candida utilis and Saccharomyces cerevisiae*were were used to evaluate the antifungal activities of the Schiff base and its complexes.

**Table 1: Physical Properties of Schiff Base and its Metal(II) complexes**

| Compound | Colour | Molecular weight(g/mol) | % yield | Melting Temp. ($^o$C) | D. Tem. ($^o$C) |
|---|---|---|---|---|---|
| L ($C_9H_{12}N_4O_3$) | Orange | 224.2188 | 65 | 118 | - |
| $[Mn(C_9H_{11}N_4O_3)_2].2H_2O$ | Brown | 537.3902 | 59 | - | 295 |
| $[Fe(C_9H_{11}N_4O_3)_2].2H_2O$ | Black | 538.2992 | 65 | - | 312 |
| $[Ni(C_9H_{11}N_4O_3)_2].4H_2O$ | Green | 577.1726 | 65 | - | 308 |
| $[Cu(C_9H_{11}N_4O_3)_2].2H_2O$ | Green | 545.9982 | 60 | - | 300 |

Key: L′ = 2-acetyl-5-methylfuran glyoxime hydrazone.
Decomp. Temp. = Decomposition Temperature.

**Table 2: Results for the Determination of the Metal Concentration in the Metal (II) Complexes**

| Compound | Absorbance (y) | Concentration (x)(mg/l) | Metal percentage (%) |
|---|---|---|---|
| $[MnL_2].2H_2O$ | 0.220 | 47.00 | 9.40 |
| $[FeL_2].2H_2O$ | 0.046 | 51.11 | 10.22 |
| $[NiL_2].4H_2O$ | 0.070 | 51.47 | 10.29 |
| $[CuL_2].2H_2O$ | 0.990 | 51.32 | 10.26 |

L′ = 2-acetyl-5-methylfuran glyoximehydrazone'

**Table 3: IR Spectral Data (cm$^{-1}$) for the Schiff base and its Metal(II) Complexes.**

| Compound | ν(N-H) | ν(O-H) | ν(C=N)ox | ν(C=N)hyd | ν(C-H) | ν(M –N) |
|---|---|---|---|---|---|---|
| L ($C_9H_{12}N_4O_3$) | 3156 | 3085 | 1584 | 1640 | 2947-2918 | - |
| $[MnL_2].2H_2O$ | 3156 | 3085 | 1614 | 1640 | 2947-2918 | 791 |
| $[FeL_2].2H_2O$ | 3085 | 2970 | 1607 | 1641 | 2970-2940 | 791 |
| $[NiL_2].4H_2O$ | 3156 | 3115 | 1640 | 1640 | 2947-2918 | 791 |
| $[CuL_2].2H_2O$ | 3353 | 3095 | 1640 | 1640 | 2947-2880 | 791 |

Key: L = 2-acetyl-5-methylfuran glyoximehydrazone. ox = oxime. And hyd = hydrazone.

**Table 4: Microanalysis Data for the Schiff base and its Metal(II) Complexes**

| Compound | M. Wt (g/mol) | Elemental %Found (Calculated) | | | |
|---|---|---|---|---|---|
| | | C | H | N | M |
| L ($C_9H_{12}N_4O_3$) | 224.2188 | 48.67(48.21) | 6.72(5.39) | 22.43(24.99) | - |
| $[MnL_2].2H_2O$ | 537.3902 | 40.07(40.23) | 4.70(4.13) | 20.81(20.85) | 9.40(10.22) |
| $[FeL_2].2H_2O$ | 538.2992 | 40.09(40.16) | 4.31(4.12) | 20.95(20.82) | 10.22(10.37) |
| $[NiL_2].4H_2O$ | 577.1726 | 37.84(37.46) | 3.93(3.84) | 19.32(19.41) | 10.29(10.17) |
| $[CuL_2].2H_2O$ | 545.9982 | 37.10(39.60) | 4.85(4.06) | 20.40(20.52) | 10.26(11.64) |

L = 2-acetyl-5-methylfuran glyoximehydrazone.

**Table 5: Molar Conductance Measurement for metal(II) complexes**

| Compound | Molecular Mass (g/mol) | Specific Conductance ($\Omega^{-1}cm^{-1}$) | Molar Conductance ($\Omega^{-1}cm^2mol^{-1}$) |
|---|---|---|---|
| $[MnL_2].2H_2O$ | 537.3902 | $2.85x\ 10^{-6}$ | 2.85 |
| $[FeL_2].2H_2O$ | 538.2992 | $2.87x\ 10^{-6}$ | 2.87 |
| $[NiL_2].4H_2O$ | 577.1726 | $2.17x\ 10^{-6}$ | 2.17 |
| $[CuL_2].2H_2O$ | 545.9982 | $2.43x\ 10^{-6}$ | 2.43 |

L = 2-acetyl-5-methylfuran glyoximehydrazone.

**Table 6: Magnetic Susceptibility Data for Metal(II) Complexes**

| Compound | Gram magnetic susceptibility $X_g(erg.G^{-2}g^{-1})$ | Molar magnetic Susceptibility $X_m(erg.G^{-2}mol^{-1})$ | Number of unpaired Electron(s) | Geometry |
|---|---|---|---|---|
| $[MnL_2].2H_2O$ | $2.66 \times 10^{-5}$ | $1.42 \times 10^{-2}$ | 5 | Tetrahedral |
| $[FeL_2].2H_2O$ | $2.13 \times 10^{-5}$ | $1.14 \times 10^{-2}$ | 4 | Tetrahedral |
| $[NiL_2].4H_2O$ | $6.16 \times 10^{-6}$ | $3.55 \times 10^{-3}$ | 2 | Tetrahedral |
| $[CuL_2].2H_2O$ | $3.85 \times 10^{-6}$ | $2.10 \times 10^{-3}$ | 1 | Tetrahedral |

L′ = 2-acetyl-5-methylfuran glyoximehydrazone.

**Table 7: Antibacterial Sensitivity Test for the Schiff base and its Metal(II) Complexes**

| | Concentration (µg/ml) | *Escherichia coli* | *Salmonella typhimurium* | *Staphylococcus epidermidis* | *Streptococcus pneumoniae* |
|---|---|---|---|---|---|
| **L** | 600 | 10 | 08 | 10 | 06 |
| | 300 | 08 | 06 | 06 | 06 |
| | 150 | 06 | 06 | 06 | 06 |
| **$[MnL_2].2H_2O$** | 600 | 10 | 10 | 06 | 06 |
| | 300 | 08 | 08 | 06 | 06 |
| | 150 | 06 | 06 | 06 | 06 |
| **$[FeL_2].2H_2O$** | 600 | 10 | 10 | 06 | 06 |
| | 300 | 08 | 08 | 06 | 06 |
| | 150 | 06 | 06 | 06 | 06 |
| **$[NiL_2].4H_2O$** | 600 | 11 | 10 | 06 | 08 |
| | 300 | 10 | 06 | 06 | 06 |
| | 150 | 06 | 06 | 06 | 06 |
| **$[CuL_2].2H_2O$** | 600 | 10 | 11 | 08 | 08 |
| | 300 | 10 | 10 | 06 | 06 |
| | 150 | 08 | 06 | 06 | 06 |
| ***Amphicillin (control)*** | 600 | 20 | 18 | 18 | 17 |
| | 300 | 16 | 16 | 14 | 14 |
| | 150 | 14 | 12 | 10 | 10 |

L′ = 2-acetyl-5-methylfuran glyoximehydrazone

**Table 8: Antifungal Sensitivity Test on the Schiff base and its Metal(II) Complexes**

| Compound | Concentration(µg/ml) | Fungal zone of Inhibition (mm) | | |
|---|---|---|---|---|
| | | *Candida albicans* | *Candida utilis* | *Saccharomyces cerevisiae* |
| L | 600 | 06 | 06 | 06 |
| | 300 | 06 | 06 | 06 |
| | 150 | 06 | 06 | 06 |
| $[MnL_2].2H_2O$ | 600 | 10 | 06 | 08 |
| | 300 | 08 | 06 | 06 |
| | 150 | 06 | 06 | 06 |
| $[FeL_2].2H_2O$ | 600 | 08 | 08 | 08 |
| | 300 | 06 | 06 | 06 |
| | 150 | 06 | 06 | 06 |
| $[NiL_2].4H_2O$ | 600 | 08 | 08 | 08 |
| | 300 | 08 | 06 | 06 |
| | 150 | 06 | 06 | 06 |
| $[CuL_2].2H_2O$ | 600 | 10 | 08 | 12 |
| | 300 | 08 | 06 | 10 |
| | 150 | 06 | 06 | 08 |
| *Nystatin(control)* | 600 | 20 | 22 | 20 |
| | 300 | 15 | 18 | 18 |
| | 150 | 10 | 15 | 16 |

L′ = 2-acetyl-5-methylfuran glyoximehydrazone

## DISCUSSION

The reaction between the Schiff base hydrazone in the molar ratio of 2:1 and aqueous solution of Fe(II), Mn(II), Ni(II) and Cu(II) chlorides in ethanol produced the respective metal(II) complexes having various colours. The colour of transition metal complexes are as

a result of d-d transitions of electrons between energy levels. All the metal(II) complex compounds produced were stable, non-hygroscopic crystals. They remained dry even when exposed to air, showing that they are not deliquescent.

The metal concentration in the complex compounds were determined using Atomic Absorption Spectrophotometer (AAS), used to calculate the percentage of the metal(II) ion in the complexes. The result obtained agreed with the theoretical values (Hassan *et al.*, 2013).

The Schiff bases were in accord with the previously reported hydrazones (Sarikavakli and Irez, 2005, Abou-Melha, 2008 and Babahan *et al.*, 2013). The IR spectra of the Schiff bases showed strong bands at 1581 – 1607$cm^{-1}$ assigned to ν(C=N) stretching frequencies, suggesting the formation of Schiff base. This bands shifted to higher frequency of 1607 – 1684$cm^{-1}$ in the metal(II) complexes revealing that coordination has taken place between the metal(II) ions and the respective Schiff bases through the (C=N) of the oxime portion while the of (C=N) portion of the hydrazones were constant (Chandraleka and Chanramahon, 2014; Canpolat and Kaya, 2004).

The IR spectra of Schiff bases show a sharp and strong intensity bands around 3022-3461 $cm^{-1}$and 1584 – 1680$cm^{-1}$ assigned to ν(N-H) and ν(C=N) respectively. The strong band due to azomethine function and amide carbonyl ν(C=O) appeared around 1650 - 1640 and 1675-1660 $cm^{-1}$respectively. The IR spectra of the Schiff bases show a broad medium intensity bands in the region of 3450-3430 $cm^{-1}$, and the band due to ν(C-O) oxime was located in the region of 1530-1520 $cm^{-1}$ due to oxime-OH. All the Schiff bases as well as the corresponding complexes show broad medium intensity bands in the region of 3310-3200 $cm^{-1}$ which were assigned to ν(N-H). The band around 1650-1640 $cm^{-1}$ in the spectra of the Schiff bases assigned to ν(C=O) was found to appear around 1630-1605$cm^{-1}$ in the complexes which indicated a significant shift. Similarly, the bands occurring around 1625-1610 $cm^{-1}$ in the spectra of the Schiff bases may be assigned to ν(C=N) which was shifted on complexation and appeared around 1585 $cm^{-1}$ in the complexes. The absence of ν(O-H) bands in all the complexes suggested involvement of oxygen of the oxime group in hydrogen bonds formation. In addition, another weak bands were also observed in the range of 760 – 790 $cm^{-1}$which may be attributed to ν(M-N).

Molar conductance measurements were carried out in $10^{-3}$ $moldm^{-3}$ DMSO at 25 $^{o}$C (at room temperature). Molar conductivity values of the complexes were given in Table 4.7a - 4.7d. The molar conductance of the synthesized metal(II) complexes of Mn(II), Fe(II), Ni(II) and Cu(II) were in the range of 2.00 – 3.02 $ohm^{-1}cm^{2}mol^{-1}$, these low values agrees with those obtained revealed the non-electrolytic nature of the complexes (Jones and Fleming, 2010).

The magnetic susceptibility results of the complexes taken under room temperature. The values for the magnetic moments for all the metal(II) complexes conformed to a tetrahedral geometry. The complex compounds have high magnetic moments values suggesting high spin complexes.

The antibacterial assay was carried out on two Gram positive: *Escherichia coli* and *Salmonella typhimurium*, and two Gram negative: *Staphylococcus epidermidis* and *Streptococcus pneumonia* bacteria using ampicillin capsule purchased from Magrib Pharmaceutical Store as control during the experiment. Zones of inhibition based upon size around each of the discs were measured in millimetre and recorded. The ligands and metal(II) complexes affect the bacterial isolates differently; the ligands showed lower activity compared to the metal(II) complexes. A comparable study of the ligands and their metal(II) complexes indicated that the metal(II) complexes exhibited higher anti-bacterial activity than the free ligands, but lower anti-bacterial activity compared to the control drugs, as recorded.

The antifungal results of the Schiff base hydrazone shown against the three fungal isolates indicated no activity at any concentrations, with the zone of inhibition were only within the disc i:e 6 mm. However the results of complexes $[MnL_2].2H_2O$ and $[CuL_2].2H_2O$ showed a higher activity of 10 mm at 600 µg/ml while complexes $[FeL_2].2H_2O$ and $[NiL_2].4H_2O$ showed lower activity of 8 mm at the same concentration when *Candida albicans* were used. Moderate zones of inhibition were observed for the remaining two fungal isolates at the remaining concentrations.

## CONCLUSION

The synthesized metal(II) complexes and newly hydrazone Schiff base successively characterized and the complexes were found to be non – electrolytic thermally stable, paramagnetic with tetrahedral geometry. They all showed good percentage yields, relatively effective to the bacterial and fungal isolates tested.

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