PJ PALGO JOURNAL OF MEDICINE AND MEDICAL SCIENCES

ISSN 2476-8340
Volume 2 Issue 1, January 2015,Page 24 - 32
http://www.palgojournals.org/PJMMS/Index.htm
Authors Email:adanekss97@gmail.com

SYNTHESIS, CHARACTERIZATION AND ANTIMICROBIAL ACTIVITIES OF Ni(II)- AND Cu(II)-COMPLEXES OF N-PYRAZOLYLPROPANAMIDE

Adane Kassa

Debre Markos University, College of Natural and Computational Science, Department of Chemistry

Accepted 27 January,2015

The ligand N-pyrazolylpropanamide and two biologically active complexes of Ni^{+2} and Cu^{+2} with N-pyrazolylpropanamide have been synthesized and characterized. The synthesized ligand, metal salts and its complexes have been screened for their antimicrobial activities against two species of bacteria (*Bacillus subtillis* and *Shigella*) by agar diffusion method. The results showed that the metal complexes are more active than the ligand and metal salts alone against these bacterial species. In addition, the biological activity of the Cu^{+2} complex is more active than the Ni^{+2} complex against these same bacterial species.

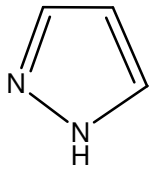
Key words: N-pyrazolylpropanamide, transition metal complexes, antimicrobialactivity, Bacillus subtillis, Shigella, agar

INTRODUCTION

Transition Metal Complexes Of Aromatic Heterocyclic Derivatives

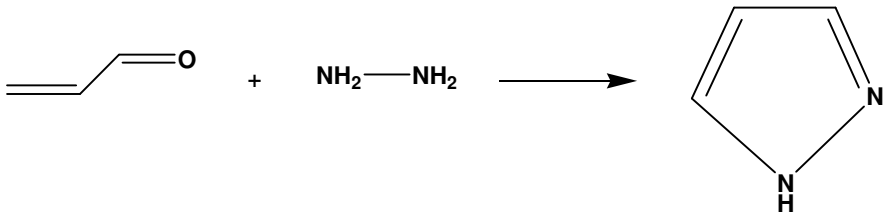
The heterocycles can be conveniently defined as cyclic organic compounds in which one or more of the ring carbon atoms have been replaced by another element such as N, O, or S. They may be either simple aromatic rings or non-aromatic rings. Some examples are Pyrazole ($C_3H_4N_2$), Pyridine (C_5H_5N), Pyrimidine ($C_4H_4N_2$) and dioxane ($C_4H_8O_2$). Heterocycles containing sulfur and/or nitrogen atoms are useful as components of functional materials since heteroatoms in their rings are helpful to stabilize ions or ion radical species, and extended π-conjugation decreases columbic repulsion. In addition intermolecular interactions caused by heteroatom contacts can be expected to form novel molecular assemblies (Yohannes, 2007; Joulef and Mills,2010).

Imidazole, Pyrazole and triazole for instance, are an abundant ligand in chemical and biological systems as it appears as such in proteins, and together with its derivatives, has been extensively employed for modeling in a wide range of inorganic subject areas, from biological applications to electronic devices and materials (Yohannes, 2007; Badea *et al*., 2008). Finding new species, with a wide spectrum of biological activity (antifungal, antimicrobial and antiviral) and also low cytotoxicity represents a very important aspect in the area of biomedical research. Important groups of organic compounds to be considered with this reference are those which form complexes with metal ions (Mahmud, 1994). The coordination chemistry of Pyrazoles and their derivatives is one area interest as these compounds show significant biological activities. Pyrazoles shown in scheme 1 refers to the class of simple aromatic ring organic compounds of the heterocyclic series. The un-substituted parent compound is known to be important intermediates for the preparation of agrochemicals and pharmaceutical compounds (http://www.britannica.com).



Scheme 1: Structure of Pyrazole

The simplest member of the pyrazole family is pyrazole itself, a compound with molecular formula $C_3H_4N_2$. It is capable of coordination to metal atoms through nitrogen. Pyrazole is usually prepared by the reaction of hydrazines with α,β-unsaturated aldehydes (scheme 2).



α,β-unsaturated aldehydes Hydrazine Pyrazole

Scheme 2. Pyrazole formation reaction

Pyrazole derivatives have played a remarkably diverse role as chelating ligands in transition metal chemistry (K. B. Girma et al., 2007). A series of new pyrazole derivatives and their complexes with many metals and their antimicrobial properties has been presented (Karamat Mahmud, 1994; K. B. Girma et al., 2007). It was observed that biological activity of these ligands increases on complexation with transition metal ions (Mahmud *et al.,* 2001).

Antimicrobial Properties Of Transition Metal Complexes

The activity of various metal ions, organic compounds and metal complexes in biological system has been explained on the basis of different complex species formed in the living organisms.

Metal ions are also found in several bacterial species and are reported to play an important role in different enzymatic and physiological reactions (Karamat Mahmud et al., 2001). Recently, the role of metal complexes has been researched in relation to antimicrobial activity and studies of transition metal complexes of heterocyclic ligands have gained importance because of their biological significance and interesting spectral and magnetic properties (Seth *et al*., 2005).

Certain drugs also play a vital role as bio-ligands in the biological systems. It has been found that the activity of the bio-metals is attained through the formation of complexes with different bio-ligands and the thermodynamic and kinetic properties of the complexes govern the mode of biological action. Sometimes, the permeability, i.e. lipophilicity of drugs is increased through the formation of chelates in vivo and drug action is significantly increased due to much more effective penetration of the drug into the site of action (Rauf, 2005).

It is worth-mentioning that many drugs substances, such as antimicrobial drugs and anticancer drugs exhibit the drug action through complexation with the available bio-metals in vivo. Interaction of various metal ions with antibiotics may enhance or suppress their antimicrobial activity but usually in many cases, the pharmacological activity of antibiotics, after complexation with metal, is enhanced as compared to that of the free ligands (Mahmud, 1994; Mahmud *et al*., 2001; Hindy *et al.,* 2006). Many of the well known antibiotics, penicillin, streptomycin, tetracycline, etc are chelating agents and their action is improved by the presence of small amount of metal ions (Hindy *et al.,* 2006).

The formation of the metal complex is dictated by its stability. This stability of the metal complex depends both on the metal ion and the ligand (Robert, 1996).

Copper (II) and Nickel (II) forms many complexes with nitrogen donors. The four- coordinate complexes are usually much more stable than the six –coordinate ones, although many complexes with six coordinates exist. The (+II) state is the most stable and important for copper.

Biologically, copper (II) will occupy unique place because of the relative stability of its complexes. Thus, the impact of biology on copper chemistry has been largely to stimulate more specific studies of area where adequate knowledge seemed to exist until the more specific questions relating to particular types of biological activity were posed.

Research Purpose And Scope

The objective of this research is to obtain complex species of biocations mainly with transition metals that possess a bioactive ligand of pyrazole derivative which might play an essential role as an antimicrobial agent. The introduction and the literature surveys on the transitional complexes of aromatic heterocyclic derivatives including the Pyrazoles, Imidazoles and triazoles revealed that metal complexes of such ligands are shown to show the significant antibacterial activities of heterocycles and their metal complexes. In addition, these complexes are shown also to play essential roles in demonstrating diverse chemical and biological processes. Hence the purposes of the present research work are:

i. To synthesize metallic complexes of the ligand, N-pyrazolylpropanamide with divalent transition metal ions including

26.Palgo J.Med.Medical Sci.

Ni^{+2} and Cu^{+2}.

ii. To characterize the synthesized ligand the metal complexes based on some physico-chemical methods.

iii. To investigate the potential of antimicrobial activities of the ligand, the Ni and Cu metal salts and their complexes towards *Bacillus subtillis* and *Shigella.*

In this study, Ni(II)-and Cu(II)- complexes of a pyrazole derivative N-pyrazolylpropanamide as a ligand are prepared and characterized. The ligand, the metal salts and their complexes are then screened against microorganisms *Bacillus subtillis* and *Shigella* for their antimicrobial activity.

EXPERIMENTAL SECTION

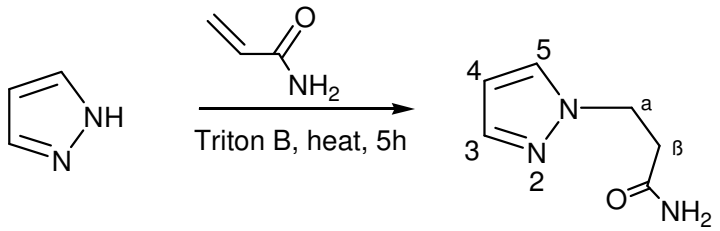
Reagents, Standards And Samples

All reagents and standard chemicals and solvents used were Analar Grade obtained commercially. Acrylamide, pyrazole, trimethylbenzylammonium hydroxide (Triton B, 40% in methanol), anhydrous copper (II) chloride, anhydrous nickel (II) chloride and absolute ethanol were used as received. Diethyl ether. The ligand N-pyrazolylpropanamide prepared form the staring materials were obtained from the institute of chemistry Otto-von Guericke University Magdeburg, Germany (Girma *et. al*., 2007). The Cu and Ni complexes of the ligand were synthesized by literature methods (Mahmud, 1994; Girma *et. al.,* 2007).

PROCEDURES: SYNTHESIS OF THE LIGAND AND COMPLEXES

Synthesis of N-pyrazolylpropanamide

The ligand was prepared on the literature (Girma *et. al*., 2007) by Michael addition of pyrazole to acrylamide in the presence of trimethylbenzylammonium hydroxide (Triton B, 40% in methanol) as a basic catalyst (Scheme 3). After about five hours of reflux in a boiling water bath the ligand was obtained in good yield (94%). Recently, de la Cruz et al have reported the synthesis and spectroscopic properties of several N-azolylpropanamides including N-pyrazolylpropanamide (Yield: 82%) obtained from acrylamide and the corresponding azoles using pyridine-sodium methoxide as a basic catalyst. Here trimethylbenzylammonium hydroxide has been used as a basic catalyst in a high-yield synthesis (Girma *et. al.,* 2007).



Scheme 3. The synthesis of N-pyrazolylpropanamide

Synthesis of the complex of Ni(II) chloride with N-pyrazolylpropanamide

The coordination compound $[NiL_2Cl_2]$ (L = N-pyrazolylpropanamide) was prepared by dissolving hexahydrated nickel (II) chloride in hot absolute ethanol. To this solution, a solution of the ligand N-pyrazolylpropanamide in absolute ethanol was added and stirred for about half an hour. The complex crystallized after the reaction mixture was allowed to stand at room temperature over fused calcium chloride for 48 hours.

Synthesis of the complex of Cu(II) chloride with N-pyrazolylpropanamide

The coordination compound $[CuL_2Cl_2]$ (L = N-pyrazolylpropanamide) was prepared by dissolving dihydrated copper (II) chloride in hot absolute ethanol. To this solution, a solution of the ligand N-pyrazolylpropanamide in absolute ethanol was added without stirring. The complex crystallized after the reaction mixture was allowed to stand at room temperature over fused calcium chloride for 48 hours.

PHYSICAL METHODS: ANALYTICAL METHODS AND PHYSICAL MEASUREMENTS

Microanalyses of the compounds described in here were performed using a Leco CHNS 923 apparatus. Melting points were determined with a Büchi B-450 digital melting point apparatus. The IR spectra of the complexes were recorded using KBr pellets on a *JASCO* FTIR 460 PLUS spectrometer system between 3500 cm^{-1} and 400 cm^{-1}.
Antimicrobial Studies was performed by using incubator and autoclave for sterilizing the material.

ANTIBACTERIAL STUDIES

Agar Diffusion Assay

The plate agar diffusion method with paper disks were used to study the microbial activities of the ligand, the metal salts and the complexes against the microorganisms investigated. This method can gave qualitative results about antimicrobial activities of antibiotic compounds.

The plate diffusion method with paper disks gave qualitative results about antimicrobial activity of the complexes. Inhibition zones larger than 6mm indicated antimicrobial activity (Pereira *et. al.,* 2007). Quantitative measurements were made by Chain and his colleagues in 1940 to monitor purification process in the isolation of penicillin (Mahmud, 1994).

The relationship between applied dose of antibiotic and the size of the resulting inhibition zone has remained for over thirty years as a basis for the comparison of samples of unknown potency with standard reference substance. The agar diffusion method is now used extensively in quality control laboratories throughout the world (Mahmud, 1994) it is a method potentially capable of yielding reliable potency estimates.

COLLECTION OF TEST ORGANISMS

The following bacteria were used for this research work: *Bacillus subtillis* and *Shigella*. They were all obtained from the Laboratory Unit, in Microbiology laboratory from Department of Biology, Bahir Dar University, Bahir Dar, Ethiopia.

CULTURE MEDIUM

Culture medium used for growth and testing of antibacterial activity was nutrient broth and nutrient agar. The composition of nutrient broth and nutrient agar is;

NUTRIENT BROTH

Sodium chloride 6g
Peptone 6g
Beef Extract 3g

These were dissolved in 1.0L distilled water and used after autoclaving the culture media at a temperature of 121^{o}c for 15 to 20 minutes. Nutrient agar was prepared in the same way by addition of only 1.5% fine agar powder along with other ingredients of nutrient broth (Karamat Mahmud, 1994; Karamat Mahmud *et.al*., 2001).

Preparation Of Petri Plates And Test Tube For Slants

Petri plates were washed with water carefully and rinsed with acetone. The rinsed Petri plates were heat sterilized in oven at 110^{o}C for 30 minutes. Test tubes for slants were plugged with cotton and sterilized in autoclave.

Pouring of Culture Medium

Culture medium was allowed to cool around 50^{o}C and then poured about 16-20ml, in each sterilized Petri plate. The slants were also prepared for making water suspension of bacterial species. The nutrient agar containing Petri plates and slants were kept for solidification of nutrient agar and stored at 4^{o}C.

Inoculation

One isolated bacterial colony was taken from such culture with the help of sterilized loop and streaked out on the slants containing nutrient agar medium. Loop was sterilized in the flame before each inculcation. Slants were labeled and placed in an incubator at 32-37^{o}C for 24 hours for growth. Bacteria grown slants were suspended in 10 ml sterilized

28.Palgo J.Med.Medical Sci.

distilled water.

Sample Ratio

The solid complexes, Metal salts and ligand (0.2gm) was dissolved in 1.0ml of acetone. Acetone was used as negative control and streptomycin sulphate was used as standard or positive control. The plates of filter paper were ranged on cool medium. After cooling, the samples were placed by micropipette on the filter paper. Then Petri plates were incubated at 32-37^{o}C for 24 hours in an incubator.

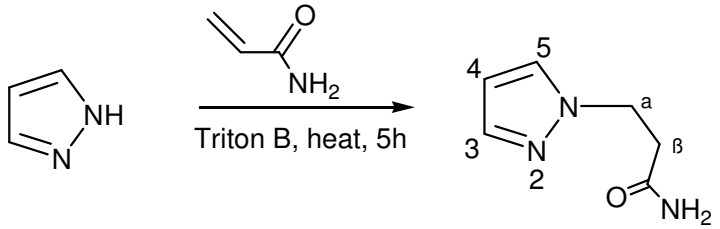
Calculations

The diameter of clear zones surrounding the disc was measured with a ruler on the under surface of the Petri plates (NCCLS, 2000). The end point was taken as complete inhibition of growth as determined by the naked eye.

RESULTS AND DISCUSSION

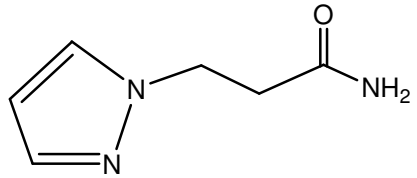
Synthesis and structure of N-pyrazolylpropanamide

A mixture of 6.8(0.1 mole) of pyrazole, 7.1g (0.1 mole) of acrylamide, and 2ml of trimethylbenzylammoniumhydroxide (Triton B, 40% in methanol) was heated for 5h in a boiling water bath. As it is indicated in scheme 4, the cooled reaction mixture was washed with diethyl ether and dried in vacuo to give of analytically pure yellowish N-pyrazolylpropanamide (Girma *et.al.,* 2007).



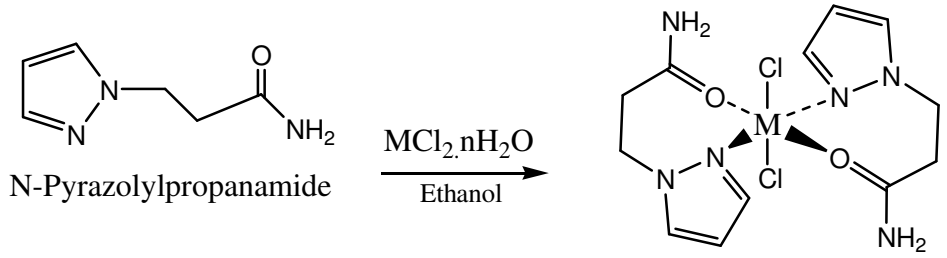
Scheme 4. The synthesis of N-pyrazolylpropanamide

IR spectrum (KBr, cm^{-1}): 3408 vs, 3349 vs, 3155 vs, 2798 vs, 2436 vs, 1670 vs, 1411 vs, 1055 vs, 971 vs, 768 vs, 668 vs, 596 vs, 577 vs, 566 vs, 525 vs, 490 vs, 475 vs, 437 (appendix A) this data were in good agreement with those reported in the literature (K. B. Girma et.al., 2007) that shows the structure of NNP (scheme 5).



Scheme 5. Structure of N-pyrazolylpropanamide

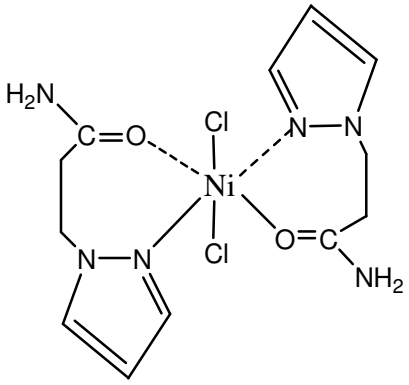
Synthesis and structure of Metal complex of N-pyrazolylpropanamide
The stoichiometric reaction between the metal (II) ion and synthesized ligands in molar ratio of M: L (1: 2) resulted in the formation of the metal complexes of type $[ML_2Cl_2]$ (Scheme 6) (where M = Cu(II), Ni(II)).



Scheme 6. Synthesis and proposed structure of metal complexes of NPP

Synthesis and structure of nickel complex of N-pyrazolylpropanamide

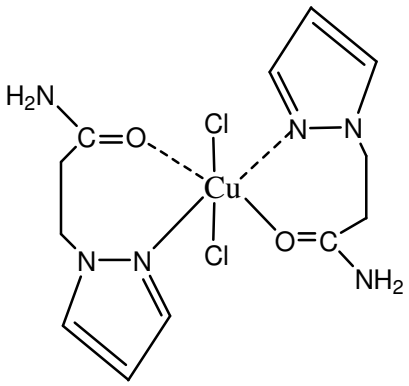
To an ethanol solution (10 ml) of Nickel (II) chloride hexahydrated (0.340 g, 0.002 mole) and an ethanol solution (10 ml) of N-pyrazolylpropanamide (1, 0.557 g 0.004 mole)) was added and stirred for about half an hour. Bright green solid substance was obtained after the solution was kept over fused calcium chloride for 48 hours which were filtered off and washed several times with ethanol. The product was dried in air to get the complex that represent on scheme 7.
IR spectrum (KBr, cm^{-1}): 3430 vs, 3372 vs, 3229 vs, 1646 vs, 1414 vs, 1296 vs, 759 vs, 660 vs, 579 vs, 511 vs, 482 vs, 452 vs (appendix C).



Scheme 7. The proposed structure of the Ni (II) complex with NPP

Synthesis and structure of copper complex of N-pyrazolylpropanamide

To an ethanol solution (10 ml) of copper (II) chloride dihydrate (0.340 g, 0.002 mole) and an ethanol solution (10 ml) of N-pyrazolylpropanamide (1, 0.557 g, 0.004 mole) was added carefully without stirring. Green crystals were obtained after the clear greenish yellow solution was kept over fused calcium chloride for 48 hours which were filtered off and washed several times with ethanol. The product was dried in air to get the complex (Scheme 8).
IR spectrum (KBr, cm^{-1}): 3447 vs, 3280 vs, 3164 vs, 3116 vs, 3011 vs, 2911 vs, 2762 vs, 2699 vs, 1662 vs, 1603 vs, 1408 vs, 1286 vs, 784 vs, 617 vs, 550 vs, 503 vs, 448vs (appendix B).



Scheme 8. The Structure of the Cu (II) complex with NPP

FTIR spectra analysis of KBr pellets showed some characteristic results for the metal complexes that were compared with the ligand spectrum to determine the changes that might have taken place during complexation. The important bands and assignments of ligands and their complexes are summarized in Table 1. The results indicate that the ligands are bidentate in nature (http://www.britannica.com).

Table 1. Important characteristic IR bands (cm^{-1}) of the Ligand and its Metal Complexes

Compound	v(NH_2)	v(C=O)
Ligand (NPP)	3408	1670

Continuation of **Table 1**

$[CuL_2Cl_2]$	3447	1662,1608
$[NiL_2Cl_2]$	3430	1646

The free ligand NPP exhibits strong bands at 1670.05 and 3408.57 cm^{-1} due to C=O and $-NH_2$ groups, respectively. In the IR spectra of the free ligand and the complex in (table 1) revealed a significant decrease to lower wave numbers at 1662cm^{-1} and 1646cm^{-1} in the CO absorptions and an increase in the positions of the NH_2 vibrations to higher wave numbers at 3447cm^{-1} and 3430cm^{-1}upon complexation. The shift of these bands in complexes suggests the coordination of nitrogen of pyrazole ring and oxygen atom of acrylamide of ligand to metal ions.

The bonding of metal ion to the ligands through N, N in NPP and N, O atoms in NPP was further supported by the presence of new bands in the region. These results are in agreement with the results of others (Girma *et.al.,* 2007) who indicate that this band shift is caused by the binding of the metal ion with the condensed ring of the NPP through N and O atoms of the ligand.

The coordination geometry of the metal centre in the complex involves an octahedral structure with two chelate ligands in the equatorial positions and two chlorides in the apical positions (Scheme 6). Each ligand is coordinated to the metal via the coordinating nitrogen of the pyrazole group and the carbonyl oxygen of the amide group in a seven-membered chelate ring.

Screening For Antimicrobial Activity

The antimicrobial activity of each of these complexes was compared with streptomycin used as positive control. 0.5 ml of each compound (0.2gm/1ml) was applied to 0.5 ml of streptomycin (0.2gm/1ml) individually. Acetone was used as a solvent for dissolving each of the compounds and it also acted as the negative control. The test organisms showed an inhibition zone of 22mm with streptomycin (positive control) (Mahmud, 1994) while, acetone has no antibacterial activity against each of the micro-organism as shown in (Table 2).

Table 2. Inhibition zone of the ligand, Metal salt and Complexes against two bacterial species from the biology laboratory a) Bacillus subtillis and b) Shigella.;

Positive Control = Streptomycin Negative Control = Acetone

Compound	**Bacillus subtillis**	**Shigella**
$CuCl_2.2H_2O$	12	13
$NiCl_2.6H_2O$	9	8
NPP	9	10
Cu(II) Complex	18	20
Ni(II) Complex	12	11
Streptomycin	22 mm	22 mm
Acetone	No Growth	No Growth

Evaluation of the antimicrobial activity of the the ligands, metal salts and metal complexes obtained from them were carried out by comparing the results of the metal complexes in percentages with the positive and negative controls. Ligands, metal salts and metal complexes were screened against the bacteria species: *Bacillus subtillis* and *Shigella*.

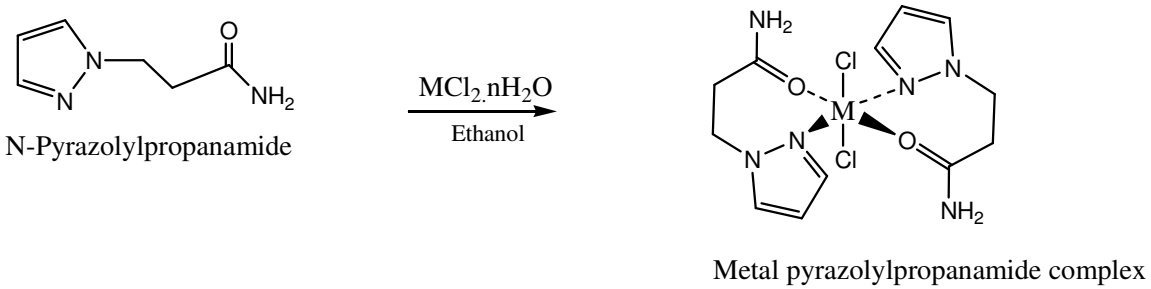
The results in the table 2 show that the free ligand, the metallic salts and their complexes were compared and found to have potent against the bacteria, the complexes have slightly more antibiotic activities than their parent ligands or metal salts against the tested microorganisms under identical experimental conditions. It is known that chelation tends to make ligands act as more powerful and potent bactericidal agents (Olagboye *et.al.,* 2013).

The values indicate that the metal complexes had higher bactericidal activity than the free ligand and metal salts. Such increased activity of the metal complexes can be explained on the basis of the overtone concept of the cell permeability, the lipid membrane that surrounds the cell favours the passage of only lipid soluble materials, due to which liposolubility is an important factor controlling the antimicrobial activity (Mishra and Singh, 1993). The bacteria screening results

reveal that the free ligand N-pyrazolylpropanamide and nickel salt showed minimum activity against bacteria species. The copper and nickel complexes showed potent against the microorganisms.
The antimicrobial activity of Cu(II) complexes show better activity than that of Ni(II) complexes. These effects may also be associated with the electronic distribution and stereochemistry of the ligand due to complexation.

Conclusion

N-pyrazolylpropanamide derived by condensation of acrylamide with pyrazole, act as a bidentate ligand towards metal ions (copper, and nickel) via the pyrazole-N and deprotonated-O of the acrylamide. The stoichiometric reaction between the metal (II) ion and synthesized ligands in molar ratio of M: L (1: 2) resulted in the formation of the metal complexes of type $[ML_2Cl_2]$ (where M = Cu(II), Ni(II)) (Scheme 9). The IR data suggested for the complexes to have an octahedral geometry around the central metal atom.
Based on infrared spectral data the following Synthesis and structures were proposed for the reaction between N-pyrazolylpropanamide and Metal salts.



N-Pyrazolylpropanamide

Metal pyrazolylpropanamide complex

Scheme 9. Synthesis and proposed structure of metal complexes of NPP

This study shows that the antimicrobial activity of N-pyrazolylpropanamide is modified in the presence of metal ions. The metal (II) complexes exhibit more antimicrobial properties against Bacillus subtillis and Shigella as compared to the uncomplexed ligand or metal salts.

Acknowledgement

I express my deepest gratitude and respect to my advisor Dr. Girma Kibatu for his guidance, advice, and comments on the work throughout the investigation.

References

Badea Mihaela, Olar Rodica, Marinescu Dana and Vasile Gina (2008).Thermal behavior of some new triazole derivative complexes,*Journal of thermal analysis and calorimetry*, Vol. 92, Page 209-214.
Girma K. B., Lorenz V., Blaurock S. and. Edelmann F. T. (2007). Synthesis and coordination compounds of N-pyrazolylpropanamide- a Versatile Acrylamide-derived Ligand; *Zeitschrift für anorganische und allgemeine Chemie*; Volume 634, Issue 2, Pages: 267-273.
Hindy A. M., Mohamed G.G., Omar M. M. and Turk J. (2006). Metal complexes of Schiff bases preparation, characterization, and Biological Activity; chem. 30, page, 1-22.
Joulef J.A. and Mills K. (2010). Heterocyclic Chemistry (4th edition). Wiley-Blackwell. page 1-60.
Mahmud Karamat (1994). Synthesis, Stability constant and Evaluation of Pyrazole complexes; PhD Thesis, submitted to University of the Punjab, Quaid-E-Azam campus Larhore; Pakistan.
Mahmud Karamat, Khan Misbahul A. and Iqbal Muhammad Z. (2001). Antibacterial property of some transition metal complexes of pyrazole derivatives; *Pakistan Journal of Biological Sciences* 4(8), page,1000-1001.
Mishra L. and Singh V. K. (1993). Synthesis and pharmacological evaluation of new benzimidazole derivatives. Indian J Chem. 32A: 446.
National Committee for Clinical Laboratory Standards (NCCLS) Approval Standard Document M2-A7 (2000). Vilanova, Pa, USA.
Olagboye S.A., Okoronkwo A.E. and Lajide L. (2013). Synthesis, Characterization and Antimicrobial Evaluation of Mixed Ligand Complexes of Ni (II) and Co (II) 1,2,3- Triazole with Thiocyanate. 2: 25- 31.
Pereira Regina M.S., et. al. (2007). Synthesis and Characterization of a Metal complex containing Naringin and Cu, and its Antioxidant, Antimicrobial, Antiinflammatory and Tumor cell cytotoxicity; molecules, 12, page 1352-1366.

32.Palgo J.Med.Medical Sci.

Pyrazole, http://www.britannica.com/EBchecked/topic/484811/pyrazole;19 Aug,2009.

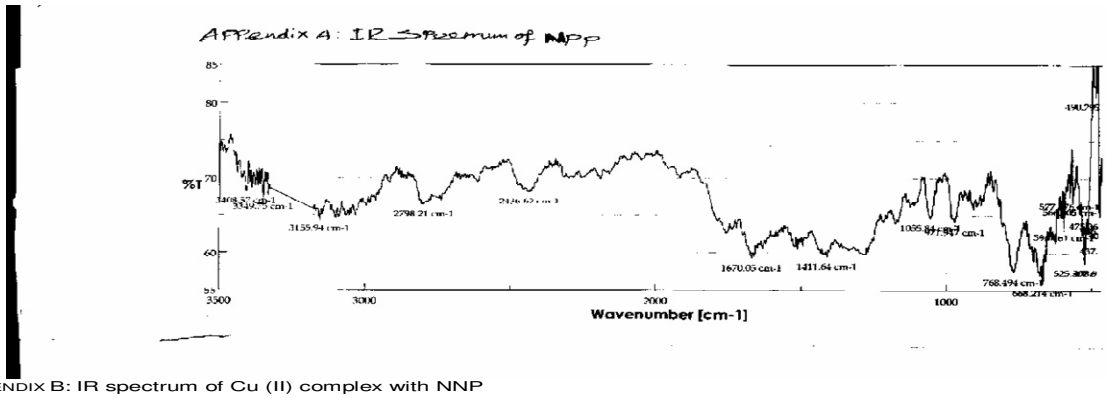
Rauf Abdul (2005). Synthesis and biological studies of some Schiff base compounds and their transition metal Complexes; PhD Thesis, submitted to Bahauddin Zakariya University.

Robert E.M.(1996). Modern Inorganic Chemistry, metal complexes in aqueous solution. New York, page 15-30.

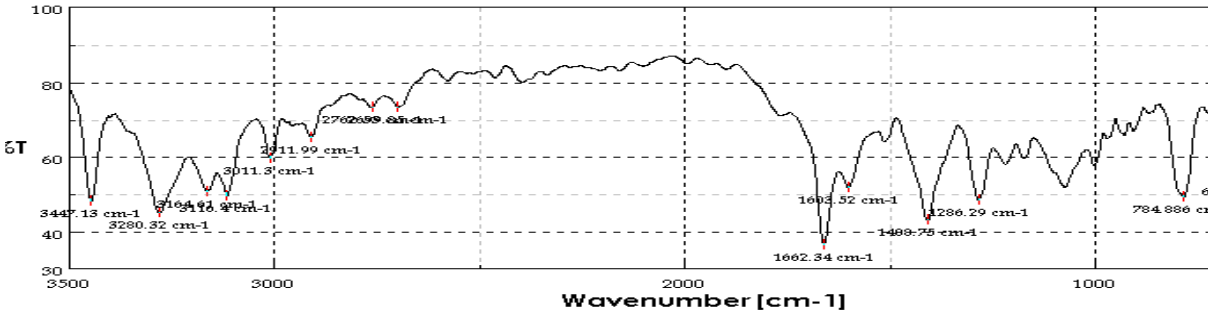
Seth Gita, Samota Mahesh K. and Mourya Nerendra K. (2005). Synthetic strategy, structural Investigations and Antifungal Studies of Cobalt (II) Complexes with Bidentate hetrocyclic Ligands; *Chemistry an Idian Journal*, 2(3-4), page 109-112.

Yohannes Misskire (2007). Synthesis and Characterization of Metal Complexes of a New Heterocyclic Ligand; Master Thesis, submitted to Addis Ababa University.

Appendices



APPENDIX B: IR spectrum of Cu (II) complex with NNP



APPENDIX C: IR spectrum of Ni (II) complex with NNP