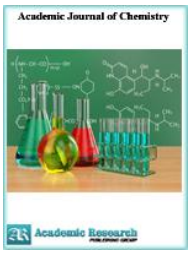

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Effect of Mn(II), Co(II), Ni(II), Cu(II), Zn(II) and Fe(III) Mixed Ligand Complexes on some Bacteria Causing Eyes Infections (BCEI)

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Abstract: The Mn(II), Co(II), Ni(II), Cu(II), Zn(II) and Fe(III) mixed ligand complexes were prepared by using 1,2-dihydroxybenzene (Catechol) as main ligand (HL1) and 2-hydroxybenzaldehyde (Salicyladehyde) as secondary-ligand (HL2). All the prepared mixed ligand complexes were characterized by using several physiochemical tools. Both electronic spectra and magnetic moment measurements exhibited the existence of an octahedral structure for Mn(II), Co(II) and Fe(III) complexes, square planar geometry for Ni(II) and Cu(II) complexes and tetrahedral structure for Zn(II) complex. The effect of the free ligands, metal salts and their mixed ligand complexes were examined on some bacteria species causing eyes infections (BCEI) such as Escherichia coli, Klebsiella pneumouiae and Staphylococcus aureus. Catechol has more effect than salicyldehyde on all tested bacteria, this is due to the existence of phenolic groups which are electron-releasing so increased the activity, meanwhile, the ligand and their mixed ligand complexes exhibited varying degrees of inhibitory effects on the growth of the bacteria strains.

Keywords: Mixed ligand complexes; Salts; Ligands; Pathogenic Bacteria (E. coli, K. pneumouiae and S. aureus).

1. Introduction

The pathogenic microorganism cause infections to eyes due to their strength and host's reduced resistance from many factors, for example; Personal hygiene, nutrition, age and fever [1]. In recent years, the mixed ligand complexes with several transition metal ions are well known to play a good role in biological applications [2, 3]. The Ce(III) complexes of some amino acids with 2, 3-dimethyl-1-phenyl-4-salicylidene-3-pyrazolin-5-one have been prepared and investigated based on some physical techniques. The free ligands and their complexes were tested for their antimicrobial activities and exhibited the potent biological activities against *S. aureus, Corynebacterium diphtheriae, P. aeruginosa and E. coli* species [4]. Some mixed ligand complexes of 2-hydroxynapthaldehyde/substituted salicylaldehyde and heterocyclic nitrogen base 8-hydroxyquinoline with isatin monohydrazone were prepared and characterized by several physiochemical tools; Based on the obtained data, an octahedral structure was suggested for all the complexes. The metal salts, free ligands and mixed ligand complexes were examined for their antibacterial activity against antimicrobial activity, two types of bacteria which are Gram negative and Gram positive and fungi strains. The results showed that the mixed ligand complexes have enhanced activity as compared to free ligands. Cu(II) mixed ligand complex was found to be most potent antimicrobial agent [5].

The eye infections is mainly caused by these pathogenic bacteria *(E. coli, K. pneumouiae* and *S. aureus*), this study aims to examine the activity of the used metal salts, ligands, and mixed ligand complexes on the pathogenic bacteria which causing eyes infections.

2. Experimental

2.1. Preparation of the Mixed Ligand Complexes

The mixed ligand complexes of catechol and salicylaldehyde with Mn(II), Co(II), Ni(II), Cu(II), Zn(II) and Fe(III) ions have been reported [6].

2.2. Isolation of Some Bacterial Infections of Eyes Patients

Eighteen samples were collected from patients attending to Al-Naher hospital of eyes diseases. The specimens were collected from patients by cotton swaps were processed as follows; a small portion of the most purulent of the

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samples, transferred with a platinum wire loop and placed at one of the extremities of the slide. The small portion of pus was smeared to the other extremity of the slide. The sample portion remaining on the slide was distended and fixed for staining.

2.3. Gram Stain Technique

The Gram stain reaction was used to help in the identification of pathogens in specimens and cultured by their Gram reaction (Gram positive or Gram negative).The morphology sample can also be identified in Gram smears.

2.4. Bacterial Cultures

Plate cultures of nutrient agar (OXID) medium were used for culture of bacteria. The medium was prepared by dissolving 28 g of powder in 1 liter of sterile distilled water, then the medium was sterilized by autoclaving at 121 °C for 15 minutes. The bacteria were cultured and incubated at 37 °C for 24 hrs.

2.5. Bacterial Cultures on Prepared Media

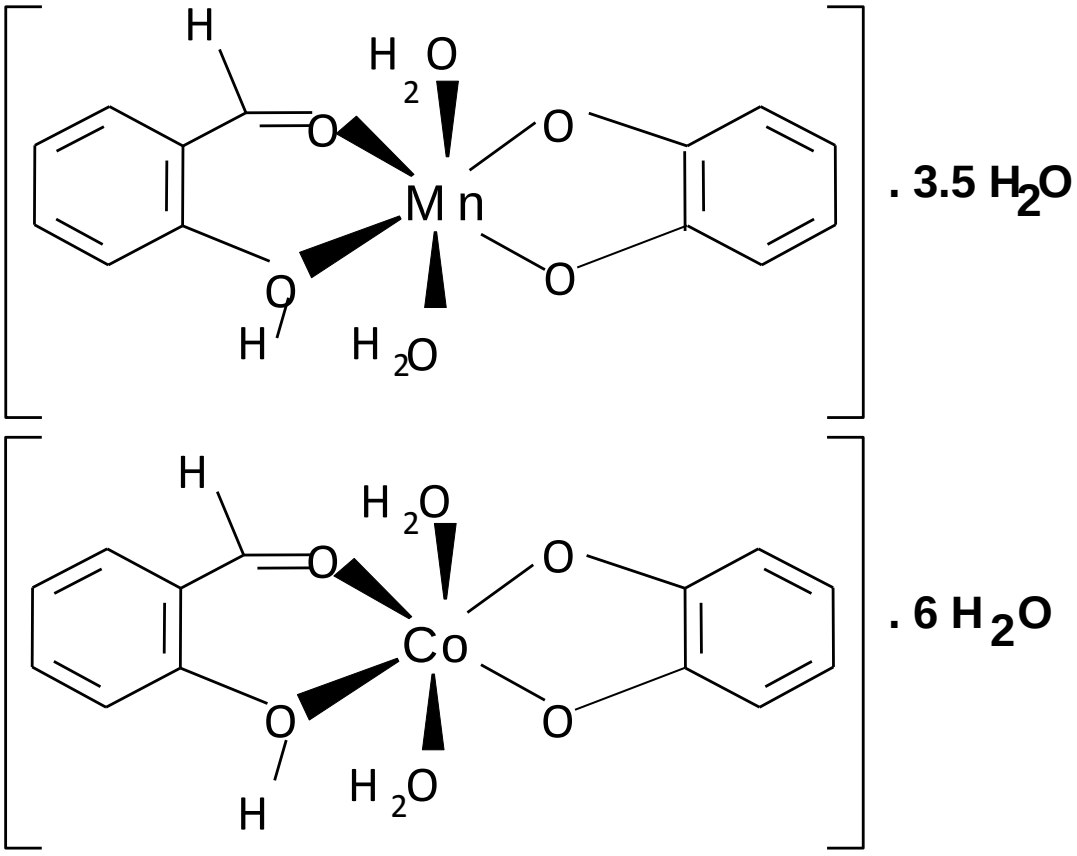
After examined by microscopic examination, microbial culture which confirmed by special media (Hi media) for all strains of bacteria. Use a new type of prepared plate cultures to confirm the obtained results of cultures where, identify the pathogenic bacteria according to special color of each bacteria. The bacteria were cultured and incubated at 37 ^{o}C for 24 hours.

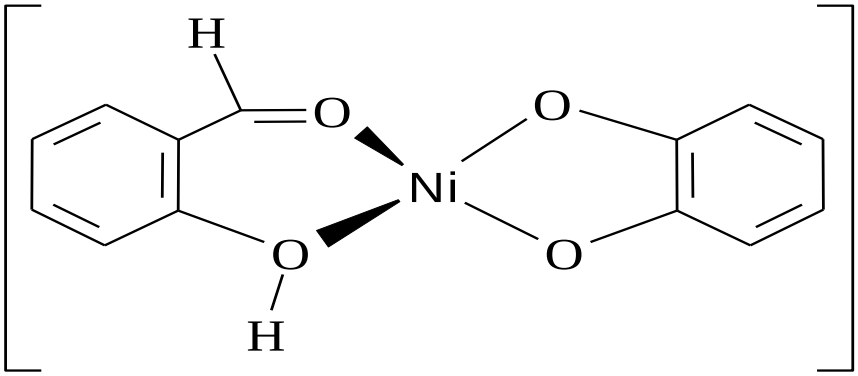
2.6. Antibacterial Assay

The antibacterial tests were assayed according to the diffusion method. The strains of bacteria used were *E. coli*, *K. pneumouiae* and *S. aureus*. The identity of all the strains was confirmed. A bacterial suspension was prepared and added to the sterilized medium before solidification. The medium with bacteria was poured into sterilized Petri dishes under aseptic condition. Different weights of metal salts, ligands and mixed ligand complexes (5 mg, 10 mg and 20 mg) were placed on the surface of the culture and incubated at 37 °C for 24 hrs. After incubation, the average of inhibition zones was recorded by (mm). The antibacterial activity was indicated by the presence of clear inhibition zones around the samples.

3. Results and Discussion

The reaction of the ligands with the metal ions gives three types of the geometrical structures shown below:





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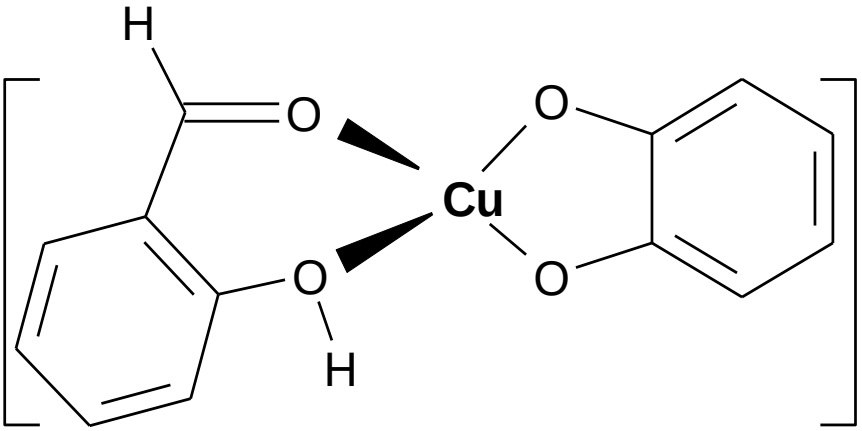

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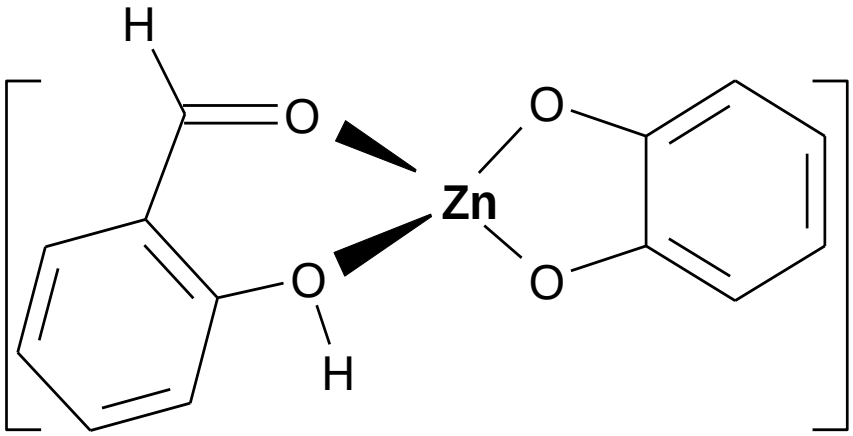

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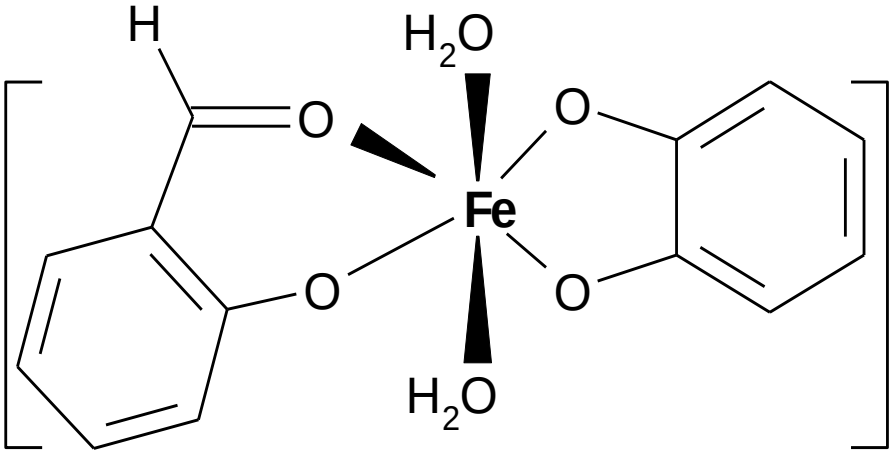

. 2 H_2O

3.1. Antibacterial Activities

In the present study, table (1) reveals the antibacterial activity results of metal salts, ligands and mixed ligand complexes which were tested at different concentrations of (5, 10 and 20 mg) against some pathogenic bacteria strains which cause eyes infections, whereas, some of tested bacteria strains were resistant to conventional antibiotics. *K. pneumoniae* has become highly resistant to antibiotics. When *K. pneumoniae* produces an enzyme known as a carbapenemase (referred to as KPC-producing organisms), then the class of antibiotics called carbapenems will not work to kill the bacteria and treat the infection. Infections caused by KPC-producing bacteria can be difficult to treat because fewer antibiotics are effective against them [7]. CRE, which stands for carbapenem-resistant *Enterobacteriaceae*, are a family of germs that are difficult to treat because they have high levels of resistance to antibiotics. *K. pneumoniae* and *E. coli* are examples of *Enterobacteriaceae*, a normal part of the human gut bacteria, that can become carbapenem-resistant. Few antimicrobial therapy options exist for infections caused by CRE [8]. In such cases, a microbiology laboratory must run tests to determine which antibiotics will treat the infection. However, few studies have analyzed the trends and prevalence of in vitro *K. pneumoniae* antimicrobial drug resistance in the United States during the late 1990s [9]. Furthermore, few investigations have examined antimicrobial drug resistance with regard to specimen source or cross-resistance patterns among CRE [10]. Thus, in this study also all used metal salts have varying antibacterial influence on bacterial species, whereas, the Co (II) salt has antibacterial activity of greater effect than all other compounds against all bacteria even resistant bacteria such as; *K. pneumonia* [11]. All the ligands and their mixed ligand complexes individually exhibit varying degrees of inhibitory effects on the growth of the tested bacterial species. Meanwhile, the free ligands; catechol has more antibacterial activity than salicylaldehyde that is may be due to presence of a phenolic (-OH) groups which are electron - releasing so increased activity [12-14]. The obtained results evidently show that the activity of the free ligand compounds becomes more pronounced when coordination to the metal ions [15]. However, the moderate effect was observed with Cu (II) complex against *S. aures* and no activity was observed against *E. coli* and *K. pneumonia* with all concentrations. Mn (II) complex was more effective against *K. pneumonia* and revealed no effect against *E. coli* and *S. aures*. Compartively, Zn (II) complex shows an active effect against *S. aures* with all concentrations. However, the same complex has resistant bacteria at high concentration against *K. pneumonia*. In contrast, no effect was observed against *E. coli*. but both Ni (II) and Fe (III) complexes were found to be inactive against all bacteria used. Whereas, Co (II) complex possess the best results and presented antibacterial activity over both types of bacterial strains (Gram positive and Gram negative) compared to other complexes and it is definitive that metal salts do play a significant role in enhancing the antibacterial activity on complexation. It is suggested that in the complex, the positive charge of the metal ion is partially shared with the donor atoms and there is π - electron delocalization over the whole complex ring. This increases in the lipophilic character of the metal complex and favors its permeation through lipoid layers of the bacterial membranes. It is also suspected that factors such as solubility, dipole moment and cell permeability mechanisms are also influenced by the metal ions [16]. The results reveals the decrease of the complexes weight placed on the bacterial culture.

Table -1. Effect of the metal salts, ligands and mixed ligand complexes on bacteria growth which causing eyes infections(BCEI)

Samples	*E. coli*			*K. pneumouiae*			*S. aures*		
	5mg	**10mg**	**20mg**	**5mg**	**10mg**	**20mg**	**5mg**	**10mg**	**20mg**
$MnCl_2.4H_2O$	18	22	30	19	25	34	8	9	16
$CoCl_2.6H_2O$	29	32	34	34	37	44	20	25	30
$NiCl_2.6H_2O$	20	25	30	20	30	35	19	25	27
$CuCl_2.2H_2O$	15	24	30	19	25	30	20	24	29
$ZnSO_4.7H_2O$	12	15	19	10	15	20	13	17	19
$FeCl_3.6H_2O$	14	19	25	15	24	20	10	17	18
Catechol	16	21	25	14	18	20	15	18	21
Salicyaldehyde	6	10	12	20	25	29	7	8	19
Mn(II) complex	-	-	-	-	14	34	-	-	-
Co(II) complex	16	21	26	10	12	15	14	19	21
Ni(II) complex	-	-	-	-	-	-	-	-	-
Cu(II) complex	-	-	-	-	-	-	12	15	20
Zn(II) complex	-	-	-	-	-	21	15	20	23
Fe(III) complex	-	-	-	-	-	-	-	-	-

* Mean of inhibition zone by (mm)

4. Conclusion

From this study, it was observed that all the salts and ligands have an effect on all types of bacteria which caused infections to eyes, whereas, Ni(II) and Fe(III) mixed ligand complexes have no effect on the used pathogenic bacteria, but the Cu(II) mixed ligand complex has an effect on *S. aures* bacteria and Zn(II) mixed ligand complex has effect on *K. pneumouiae* and *S. aures* strains.

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