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Synthesis, analysis and biological studies of transition metal complexes of cefixime

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Abstract

Cephalosporin is extensively used broad-spectrum antibiotic, containing β-lactam ring. However, due to overuse of this drug many bacteriological pathogens are now resistant. The study aim to prepare new derivatives of above drug, which will enhance biological activity against pathogens. In current study, cefixime was reacted with essential trace elements to synthesize respective metal complexes. It comprises formation of mixed ligand complexes of cefixime as primary ligand and glycine as secondary ligand with transition metals. The novel compounds were characterized physically like shape, color, melting point, solubility in different polar and non-polar solvents, pH and Rf value. Spectroscopic characterization like electronic spectra IR spectroscopy and antimicrobial screening against gram positive and gram negative bacterial strain. Biological screening of novel compounds was accomplished against diverse group of pathogens. Some of our novel compounds showed high efficacy against the pathogens as compare to parent composites.

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Introduction

Cephalosporins are diverse group of beta-lactam broad spectrum antibiotics that were first isolated from cultures of *Cephalosporiumacremonium* in 1948 by Italian scientist Giuseppe Brotzu. They interfere with the synthesis of bacterial cell wall by disrupting the cross linking of bacterial peptidoglycans (Fountain and Russell, 1969). Four generations are available in market with different antibacterial spectrum (O'Callaghan, 1975). All cephalosporins are synthesized from cephalosporins C.Cefixime is a third generation semisynthetic cephalosporin antibiotics broad spectrum more active against gram negative strains than 1st and second generations and less active against gram positive strains. Chemically it is 7-2(2-(amino-4-thiazolyl)-2 (carboxymethoxyimino) acetamido)-3-vinyl-cephem-4-carboxylic acid (Ali, 2003). Construction of molecular models indicates that its structures is suitable for chelate formation and acts as polydentate ligand (Anacona and Estacio, 2006;Arayne *et al.*, 2002;Pillai and Latha, 2012).

Glycine is one of the smallest of 20 amino acid found in protein formation. (Ingber, 1998). There is substantial experimental evidence that free glycine may have a role in protecting tissue, against diseases such as ischemia hypoxia and reperfusion. In glycine structure following functional groups must be consider as site for metal attachments the terminal amino group, the terminal carboxylate group the peptide oxygen atom and the peptide nitrogen atom. Glycine complexes with transition metals have been reported earlier(Li *et al.*, 1957) these complexes have promising antibacterial properties. Glycine act as bidentate ligand through both carboxylate and amino groups (El-Said *et al.*, 2009) . Glycine is also used as Secondary ligand in many complexation reactions (Aiyelabola *et al.*, 2012;Mahmoud *et al.*, 2015;Sanoja *et al.*, 2014;Soliman *et al.*, 2014).

Metal complexes of many drugs possess modified pharmacological and toxicological properties revealed from many studies done on drugs metal complexes from past three decades. Many antibiotics require a metal atom for its functioning or antimicrobial action like bleomycin, streptogirin and bacitracin. Metal complexes of antibiotics possess unique bioactive properties as they can interact with biological molecules like DNA, RNA, lipids and protein receptors (Sugiura *et al.*, 1985, Vaidyanathan and Nair, 2003).Platinum metal complex cisplatin is used as chemotherapeutic agent (Cleare *et al.*, 1980). silver(I) complexes commonly used as anti-microbial agents, bismuth(III) complexes for anti-ulcer treatment, gold(I) complexes as anti-arthritic agents, gadolinium(III), manganese(II) and iron(III) complexes as magnetic resonance imaging (MRI) contrast agents, tech-netium (99Tc) and scandium (47Sc) as radiopharmaceutical agents (Hayes, 1978, Li *et al.*, 2012, Reilly, 2007).

Present study comprises of formation of mixed ligand complexes of cefixime as primary ligand and glycine as secondary ligand with transition metals. And its physical characterization like physical appearance Solubilities and melting points, spectroscopic characterization like electronic spectra IR spectroscopy and antimicrobial screening against gram positive and gram negative bacterial strains.

Materials and methods

The cefixime (ligand 1) used were of pharmaceutical grade gifted by Ameer Adnan pharmaceutical sundar industrial area Lahore. All the solvents were of commercial grade and other chemicals were of analytical grade; they were used without further purification. All the manipulations were performed under aerobic conditions.

Synthesis of complexes

For the synthesis of complexes tow techniques were used i.e. Reflux condensation and Magnetic stirring. Reflux involves heating the chemical reaction for a specific amount of time, while continually cooling the vapour produced back into liquid form, using a condenser. The vapours produced above the reaction continually undergo condensation, returning to the flask as a condensate.While using this method the reaction mixture was heated using reflux condenser

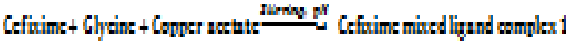
on a hot plate with continuous stirring for 3 to 5 hours. The heat and stirring provide enough energy to metal for binding to ligand.

Magnetic stirring involves stirring the reaction mixture with the help of magnetic stirring on a hot plate or a stirring device where the stirring rate can be controlled. While using this technique the reaction mixture was stirred for 8 hours or overnight. The stirring provide enough energy for the metal to bind with the ligand.

Synthesis of Complex 1

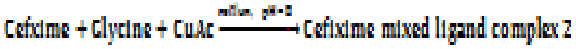
A mixed ligand complex was prepared with 1:2 ratio. Copper acetate was used as metal salt and cefixime and glycine in 1:1 as mixed ligand. Where cefixime act as ligand and glycine as co-ligand. 30mg (0.165 mmol) of copper acetate was weighed and transferred to a 50 ml beaker to which 5 ml of methanol was added and a clear light green sol was formed. 12 mg (0.1598 mmol) glycine sol was prepared in 5 ml of water. 74.82mg (0.165mmol) of cefixime was weighed and its sol was prepared in 10 ml of methanol in 50ml round bottom flask. Salt sol was heated on water bath to which glycine sol was added. The color change from greenish yellow to pure blue. The mixture was transparent and this was added dropwise to cefixime sol in round bottom flask with continuous magnetic stirring. The colour changes from blue to greenish yellow and turbid. The stirring was continued. PH of the mixture was 4. Mixture PH was adjusted to 8 (Anacona and Estacio, 2006, Pillai and Latha, 2012) by using 5% NaOHdropwise with continuous stirring. The colour of the mixture changes from greenish yellow to brownish yellow and less turbid than before. After maintaining the PH to 8 stirring was continued for 8 hours (El-Said *et al.*, 2009). TLC of the reaction mixture was also done and retention factor was determined and compared with the pure ligand four time during the reaction. After 8 hours the flask contents were filtered and poured into a 50 ml beaker. The beaker was covered with Aluminium foil and the foil was punctured at various places. The beaker was placed at room temp for the evaporation of solvent. After 7 days the product was dried and it was scratched and recrystallized twice. Once with methanol water co-solvent in 50:50 and then with methanol. Chocolate brown brick shaped tinny shiny crystals were obtained. These were dried in desiccator for 24 hours over activated silica gel. Physical characteristics of the product was measured as solubility, color, shape and melting point. The product was transferred to Eppendorf and labelled and send for spectroscopic measurements like FTIR UV-visible and also preserved for antimicrobial assessment.

The supposed reaction equation was as following



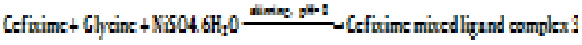
Mixed ligand complex of cefixime as primary ligand and glycine as secondary ligand with copper were synthesized in ratio of 1:1:1. Distil water and methanol in ratio of 50:50 were used as co-solvent. 30mg (0.165mmol) of copper acetate was added to a 50 ml beaker. To this 5 ml of co-solvent was added and stirred. 12mg (0.16 mmol) of glycine sol in 5ml of co-solvent was added to the copper acetate sol with continuous stirring. The sol color changes from light green to light blue.74mg (0.165mmol) of cefixime was added to 100ml round bottom flask properly labelled to which 10 ml of co-solvent was added. Copper acetate and glycine mixture was added dropwise to cefixime sol with continuous stirring. The mixture color changed to yellowish green and turbid. The PH of the mixture was 3. The PH of the mixture was maintained to basic at 8 by the addition of 5% NaOH dropwise (Anacona and Estacio, 2006, Pillai and Latha, 2012) with continuous stirring. At PH 8 the mixture colour become dark brown and transparent. This reaction mixture was refluxed for 5 hours(KAUSHAL *et al.*). During the refluxing process the RF values were checked by using TLC card to confirm the usage of ligand in reaction. After 5 hour reflux the flask was removed and the contents were filtered and transferred to a 50ml beaker. The beaker was covered with aluminum foil. The foil was punctured at various places. The beaker was placed at room temperature for evaporation of solvent. After two weeks the contents were dried to crystalline

powder. The contents were recrystallized twice with water and methanol. And dried again at room temp first and then in a desiccator using activated silica gel for 24 hours. After proper drying further characterization was performed. Physical characteristics of the product was measured as solubility, color, shape and melting point. The product was transferred to eppendorf and labelled and send for spectroscopic measurements like FTIR, UV-visible and also preserved for antimicrobial assessment. The reaction equation for the complex 2 is.



Synthesis of Complex 3

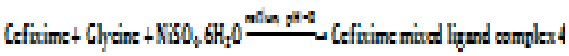
Cefixime and glycine mixed ligand complex were synthesized with Nickle sulphate hexahydrate metal. The ratio used for the reaction was 1:2 (M:L). 60mg (0.227mmol) of Nickle sulphate hexahydrate was weighed and dissolved in 5ml of methanol in a 50 ml beaker light green transparent color sol formed. 17.14mg (0.228mmol) of glycine was weighed and dissolved in 5 ml of distil water and a colorless transparent sol was formed. Glycine sol was added to salt sol with continuous stirring. 103.547mg (0.228mmol) of cefixime was weighed and dissolved in 10 ml of methanol and a transparent colorless sol was formed. Cefixime sol than added to the salt and glycine mixture slowly with continuous stirring. The mixture colour become light yellowish green turbid. pH of the mixture was acidic which was maintained to basic pH using 5% NaOHdropwise(Patil *et al.*, 2012, Anacona and Estacio, 2006) with continuous stirring. PH is maintained for deprotonation of cefixime so it could chelate with the metal ion. It was properly labelled and stirring was extended for 8 hours(El-Said *et al.*, 2009). TLC of the reaction mixture was done during the reaction processing with the help of TLC card. And RF values were determined for the reaction mixture and compared with the ligand sol to determine the ligand is being consumed during the reaction. After 8 hours there reaction was stopped and the mixture was filtered into a 50 ml beaker. The pH was 7.8. The beaker was covered with aluminum foil. The foil was puncture with the help of pin at various places for the evaporation of solvent. The beaker was kept at room temp for drying and solvent evaporation. After 2 weeks the product was dried. The product was recrystallized twice, once with water and methanol co-solvent and then with methanol. The product was dried in desiccator above activated silica gel. The product was stored in a tightly closed eppendorf with proper labelling for characterization. Physical measurements such as solubility melting point color and shape of product was performed. Spectroscopic measurements such FTIR UV-visible and antimicrobial screening was also performed to confirm the complexation. Following was the reaction equation for this complex.



Synthesis of Complex 4

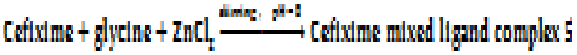
The complex 4 was formed with same ratio as used for complex 3. Distil water and methanol in 50:50 ratio was used as solvent for reaction. 60mg (0.227mmol) of Nickle sulphate hexahydrate was weighed and dissolved in reaction solvent and light blue sol was formed. 17.14mg (0.228mmol) was weighed and dissolved in the reaction solvent. Glycine and salt sol were mixed with continuous stirring. 103.547mg (0.228mmol) of cefixime was weighed and transferred to a round bottom flask. 10 ml of co-solvent was added and a transparent colorless sol was formed. Glycine and sat sol mixture was added to the cefixime sol dropwise by continuous stirring. The mixture colour was light yellow green with acidic pH. The pH of the mixture was maintained to basic at 8 by using 5% NaOH dropwise(Anacona and Estacio, 2006) with continuous stirring for deprotonation. The color changes from light green yellow to light brown transparent. Flask was properly labeled and the mixture was refluxed for 3 hours. During reflux TLC was done for determining the consumption of the ligand in the reaction. After 3 hours reflux the reaction was stopped its pH was 7.7. The mixture was filtered into a 50 ml beaker .The beaker was covered with aluminum foil. The foil was puncture with the help of pin at various places for the evaporation of solvent. The beaker was kept at room temp for drying and solvent evaporation. After 2 weeks the product

was dried. The product was recrystallized twice, once with water and methanol co-solvent and then with methanol. The product was dried in desiccator above activated silica gel. The product was stored in a tightly closed apendrof with proper labelling for characterization. Physical measurements such as solubility melting point color and shape of product was performed. Spectroscopic measurements such FTIR UV-visible and antimicrobial screening was also performed to confirm the complexation. Following was the reaction equation for this complex.



Synthesis of Complex 5

Cefixime and glycine mixed ligand complex was synthesized with zinc chloride. The ratio used for the reaction was 1:2 (M:L). 30mg (0.220mmol) of zinc chloride was weighed and dissolved in 5ml of methanol in a 50 ml beaker white transparent sol formed. 16.50mg (0.219mmol) of glycine was weighed and dissolved in 5 ml of distil water and a colorless transparent sol was formed. Glycine sol was added to salt sol with continuous stirring. 99.787mg (0.220mmol) of cefixime was weighed and dissolved in 10 ml of methanol and a transparent colorless sol was formed. Cefixime sol than added to the salt and glycine mixture slowly with continuous stirring. The mixture colour become light yellowish. pH of the mixture was maintained to basic pH using 5% NaOH dropwise with continuous stirring. PH is maintained for deprotonation of cefixime so it could chelate with the metal ion. It was properly labelled and stirring was extended for 8 hours. TLC of the reaction mixture was done during the reaction processing with the help of TLC card. And RF values were determined for the reaction mixture and compared with the ligand sol to determine the ligand is being consumed during the reaction. After 8 hours there reaction was stopped and the mixture was filtered into a 50 ml beaker. The pH was 7.8. The beaker was covered with aluminum foil. The foil was puncture with the help of pin at various places for the evaporation of solvent. The beaker was kept at room temp for drying and solvent evaporation. After 2 weeks the product was dried. The product was recrystallized twice, once with water and methanol co-solvent and then with methanol. The product was dried in desiccator above activated silica gel. The product was stored in a tightly closed eppendorf with proper labelling for characterization. Physical measurements such as solubility melting point color and shape of product was performed. Spectroscopic measurements such FTIR UV-visible and antimicrobial screening was also performed to confirm the complexation. Following was the reaction equation for this complex.

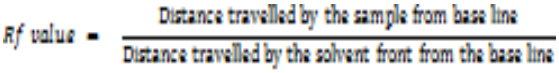


Analysis of complexes

Complexes were analyzed by various physical and spectroscopic measurements.

Physical measurements: Colour, shape and pH of the ligand sol and the complexes sol were measured. To check the solubility, the complexes were tried to dissolve in a series of solvents including distilled water, methanol, ethanol, n-hexane DMF () and acetonitrile and the results were compared with the solubility of ligand in theses solvents. Melting points of pure ligand and the complexes were measured by digital melting point apparatus (Bamstead/ Electrothermal 9100 series). The changes in the any of the physical property showed a change in the nature of compound.

Thin layer chromatography was performed during the reaction procedure to observe the consumption of ligand in the reaction mixture. The Rf values of the ligand and the compound were calculated by using the following formula;



Difference in the Rf values of the ligand and the compound gives evidence of complexation.

Spectroscopic measurements-FT-IR (Fourier transformed Infrared) spectroscopy:

The samples was completely dried over activated silica gel in a vacuum desiccator and placed in air tight eppendorf. The samples were analyzed for IR

spectrum using Kbr disc. The IR spectrum were compared and peaks were isolated interpreted and tabulated.

Antibacterial screening of ligands and samples: Antimicrobial activities of the cefixime and its complexes were measured by disk diffusion method (Arayne *et al.*, 2002, Anacona and Estacio, 2006, Auda *et al.*, 2008).

Results and discussion

Complex 1

Physical characteristics (shape, colour and solubility in polar and non-polar solvents) of the complexes 1,2,3,4 and 5 are given in Table 1 and 2. The melting point (decomposition) of product measured was 280˚C while the melting point (decomposition) of ligand was 220˚C. It also showed there is a change in the compound or complexation.

The pH of the ligand sol was 4 while the pH of complex 1 was 7.8. Ligand and product separate sol were prepared and their RF values were calculated as 0.7 and 0.65 respectively using methanol as mobile phase.

It shows that the product has less affinity for methanol which gives an evidence in the change in the nature of compound or formation of complex.

Table 1. Physical appearance of complexes.

Sr No	Samples	Color	Shape
1	Cefixime	creamy white	amorphous powder
2	Glycine	pure white	granular powder
3	complex 1	dark brown	shiny crystalline powder
4	complex 2	chocolate brown	shiny crystalline powder
5	complex 3	light brown	shiny crystalline powder
6	complex 4	orange brown	shiny crystalline powder
7	complex 5	light yellow	shiny crystalline powder

Table 2. Solubility of complexes in different solvents.

Sr no	Complexes	solubility in different solvents				
		n-hexane	DMF	water	methanol	ethanol
1	Ligand 1	insoluble	soluble	insoluble	very soluble	soluble
2	ligand 2	insoluble	soluble	very soluble	insoluble	soluble
3	complex 1	insoluble	less soluble	soluble	less soluble	soluble
4	complex 2	insoluble	less soluble	soluble	less soluble	Soluble
5	complex 3	insoluble	less soluble	soluble	less soluble	Soluble
6	complex 4	insoluble	less soluble	soluble	less soluble	Soluble
7	complex 5	insoluble	less soluble	soluble	less soluble	Soluble

The IR spectrum of the free drug was compared with those of the metal complexes in order to ascertain the binding mode of the drug to metal ion in the complexes. While comparing the FT-IR spectra of the complex 1 with the FT-IR spectra of cefixime and glycine; fruitful results were obtained regarding complexation of the metal with ligands.

The peak position of carbonyl stretching vibrations of the complex 1 occurs at 1761 cm^{-1} has not a big difference from cefixime which appear at 1755 cm^{-1}. Which showed that this position is intact and no coordination occurred at this position (Pillai and Latha, 2012). The stretching vibrations of b-lactam amide carbonyl bond appeared at 1659 cm^{-1} which are different free ligand. This showed coordination occurred at this position (Pillai and Latha, 2012). Overlapped stretching vibrations also occurred at 1551-1530 cm^{-1} due to coordination.

The glycine carboxylate band appeared at 1585 cm^{-1} which is different from carboxylate band in free glycine. This showed that the glycine coordinated through carboxylic group with metal (El-Said *et al.*, 2009). The glycine is attached through the carboxylic group to the metal atom. Complex 1 was employed for antibacterial screening.

Complex 2

The melting point (decomposition) of the product was 277˚C while the melting point (decomposition) of ligand was 220˚C. The pH of the ligand sol was 4 while the pH of complex 2 was 7.7.

Ligand and product separate sol were prepared and their RF values were calculated as 0.7 and 0.67 respectively using methanol as mobile phase. It shows that the product has less affinity for methanol which gives an evidence in the change in the nature of compound or formation of complex.

Table 3. Comparison of FT_IR spectra of major IR peaks of Ligands and complexes.

Sr no	complex	Ligand 1 (cefixime)				
		str C=O (b-lactam)	Str C=O (amide)	COO(asym)	COO(sym)	Str N-C
1	Cefixime	1755	1684	1520	1350	1391
2	complex 1	1761	1659	1551-1530	1339	
3	complex 2	1769	1646	1558-1541	1318	1361
4	complex 3	1771	1653	1558-1507	1338	1395
5	complex 4	1749		1540	1317	
6	complex 5	1769	1666	1539		1361
Sr no	complex	Ligand 2(glycine)				
		COO(asym)	COO(sym)		Str N-H2	
1	Glycine	1573	1386		2861	
2	complex 1	1585	1384		2953	
3	complex 2	1576	1387			
4	complex 3	1592	1386			
5	complex 4	1575	1386		2917	
6	complex 5	1598	1392			

While comparing the FT-IR spectra of the complex 1 with the FT-IR spectra cefixime and glycine Fruitful results were obtained regarding complexation of the metal with the ligand.

The peak position of b-lactam carbonyl stretching vibrations of the complex 1 occur at 1769 cm^{-1} with a short limb at 1716 cm^{-1} has remarkable difference from free cefixime which appear at 1755 cm^{-1}.

Which showed some changes had been occurred at this position(Anacona and Estacio, 2006).

Table 4. Comparison of pure and complexes against different stains.

Sr No	Samples	zones of clearance against different strains in mm				
		Bacillus anthrax	bacillus sabtilis	E.coli	Pseudomonas aeruginosa	controlled (no strain)
1	Ligand(Cefixime)	9	8	16	15	0
2	complex 1	8	0	18	17	0
3	complex 2	0	0	20	11	0
4	complex 3	0	11	12	14	0
5	complex 4	0	0	14	16	0
6	complex 5	7	0	9	10	0
7	No sample applied	0	0	0	0	0

The carbonyl oxygen has full chances for coordination to metal ion at this position. The stretching vibrations of amide carbonyl bond also appeared at 1646 cm^{-1} position which is different from free ligand. This showed coordination occurred at this position (Anacona and Estacio, 2006;Pillai and Latha, 2012). Overlapped stretching vibrations of carboxylate also occurred at 1558-1541 cm^{-1} due to coordination. The glycine carboxylate band appeared at 1576 cm^{-1} which is different from carboxylate band in free glycine. This showed that the glycine coordinated through carboxylic group with metal(El-Said *et al.*, 2009).The comparison of FT-IR spectra of the complex with free cefixime showed that cefixime act as meltdentate ligand while the secondary ligand glycine act as Monodentate ligand.

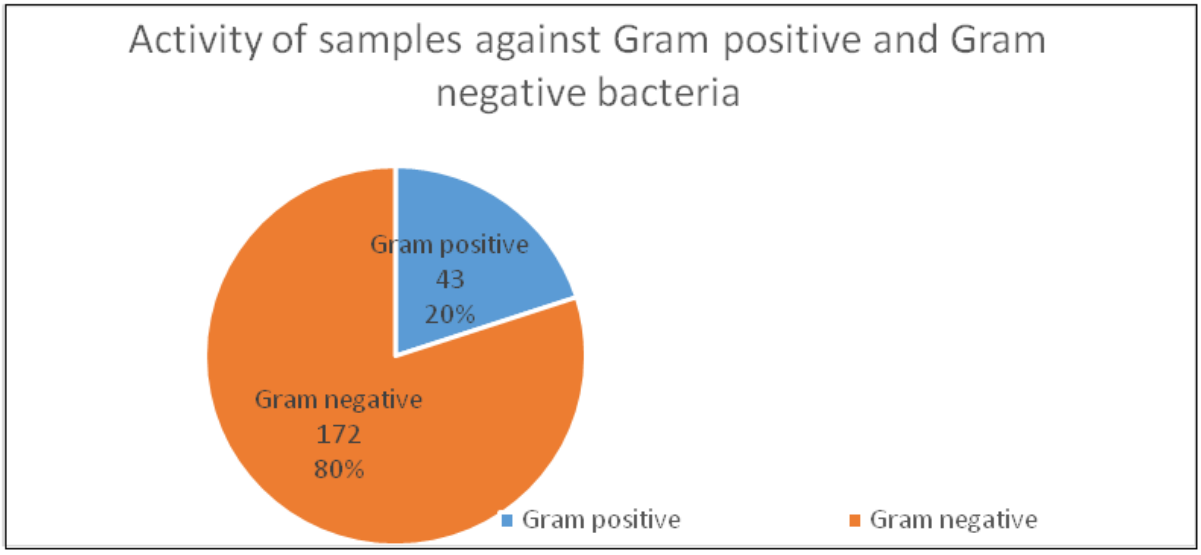


Fig. 1. Overall trend of antibacterial activity of ligand and complexes against Gram positive and Gram negative bacterial strains and Gram negative bacterial strains.

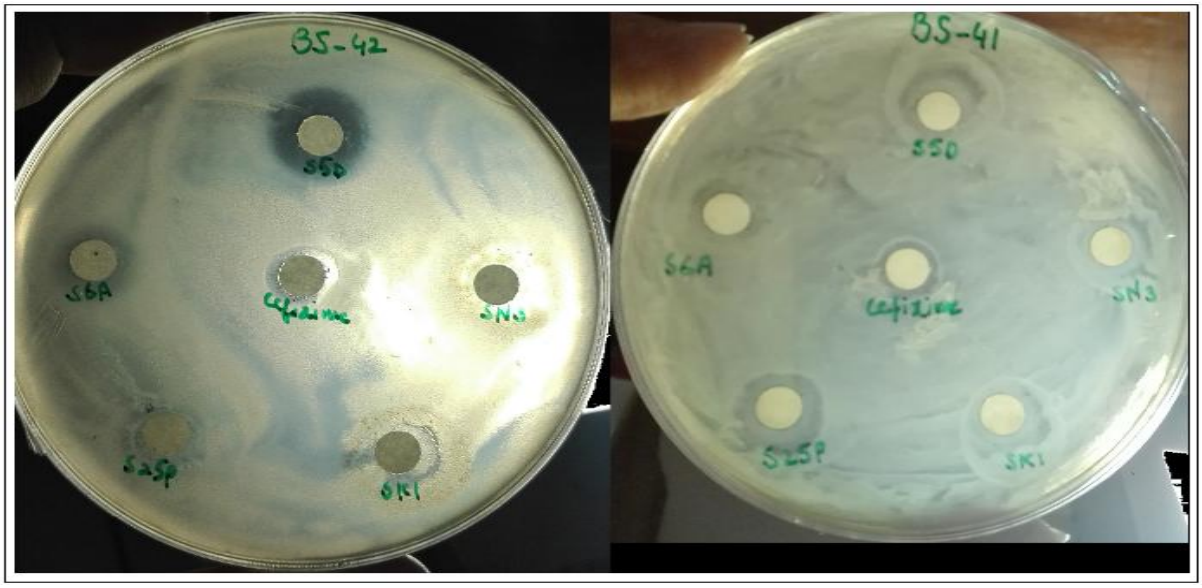


Fig. 2. Petri dishes showing zones of inhibition.

Complex 3

The melting point (decomposition) of product measured was 265˚C while the melting point (decomposition) of ligand was 220˚C.At 265 ˚C the colour of the complex start changing and became darker while there is no change in the product appearance until 400 ˚C reached. The pH of the ligand sol was 4 while the pH of complex 3 was 7.5.

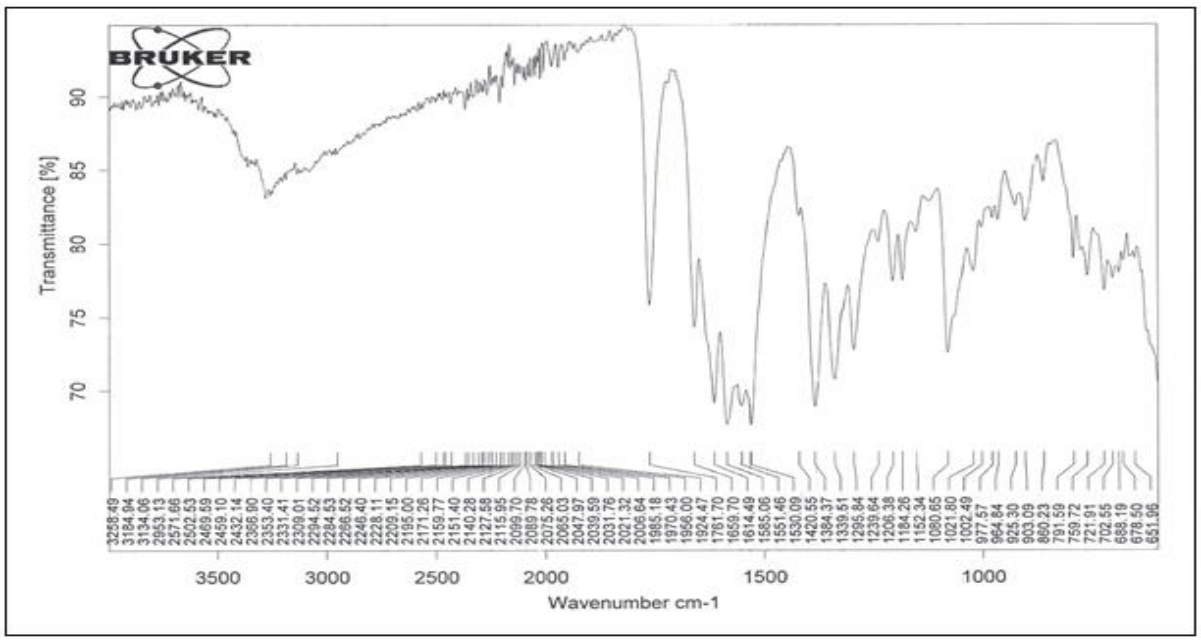


Fig. 3. FTIR spectrum of Complex 1.

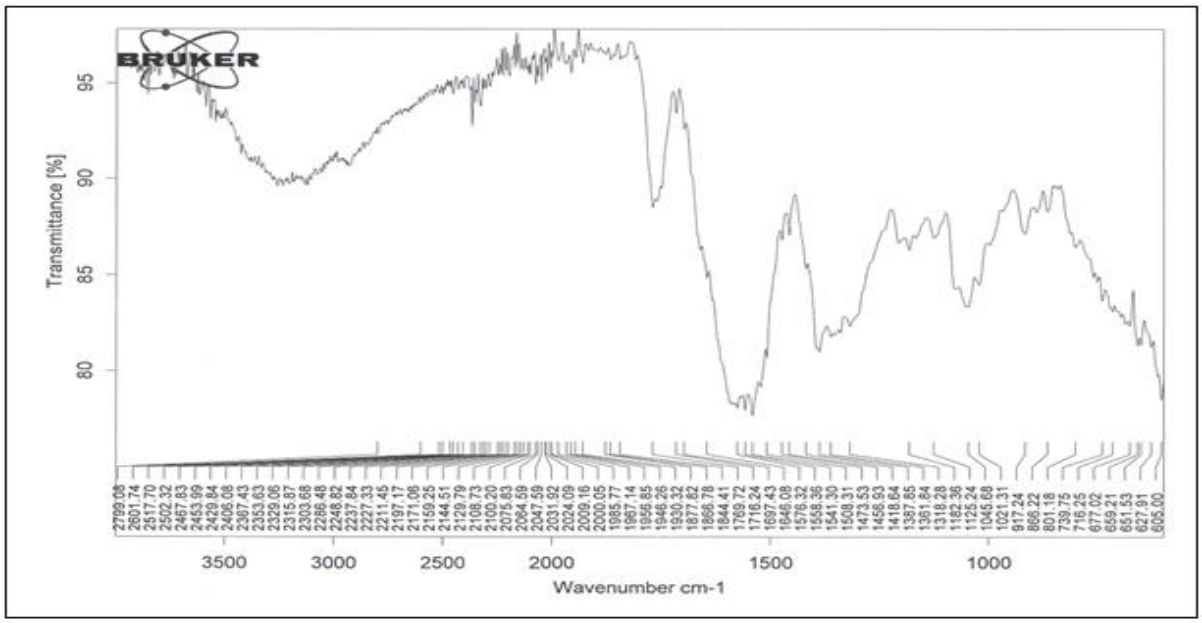


Fig. 4. FTIR spectrum of Complex 2.

Rf values were calculated by preparing separate solutions of both ligand and complex 3. Ligand Rf value was 0.7 and of complex 3 was 0.66. It showed that the product formed has less affinity for methanol as compared to ligand.

Following is the FT-IR spectra of complex 3 (KBr, cm^{-}) 1771 (C=O stretching vibrations of B-lactam carbonyl bond), 1738, 1716, 1653 (N-H stretching vibrations of B-lactam amide carbonyl bond), 1646-1616, 1558-1507 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond),1338 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond), 1296-1205, 1182, 1160, 1077-1043, 995-923, 886-805, 796-702, 691-664 and 1592 (COOH asymmetric vibrations of carboxylate of glycine), 1395 (COOH symmetric vibrations of carboxylate of glycine).

While comparing the FT-IR spectra of the complex 3 with the FT-IR spectra cefixime and glycine Fruitful

results were obtained regarding complexation of ligand with metal. The peak position of carbonyl stretching vibrations of the complex 3 occurred at 1771 cm^{-1} has remarkable difference from cefixime which appear at 1755 cm^{-1}. Which showed some changes had been occurred at this position(Anacona and Estacio, 2006).

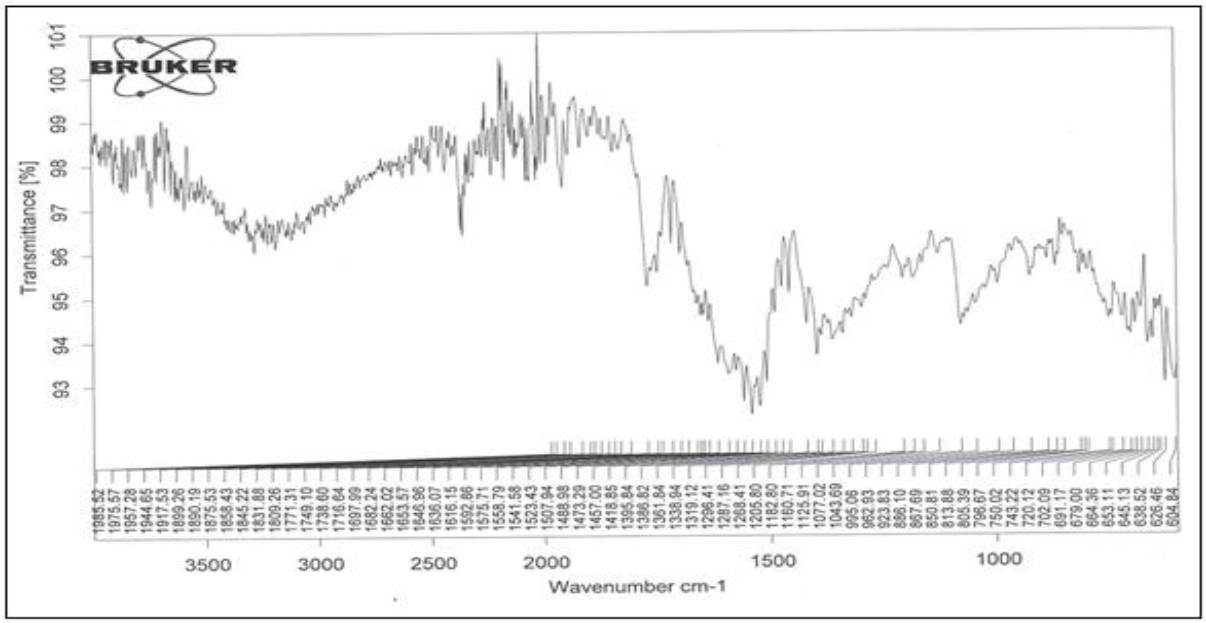


Fig. 5. FTIR spectrum of Complex 3.

The carbonyl oxygen has full chances for coordination to metal ion at this position. The stretching vibrations of b-lactam amide carbonyl bond also appear at different position than the free ligand. This also showed coordination occurred at this position(Pillai and Latha, 2012). Overlapped stretching vibrations also occurred at 1558-1516 cm^{-1} due to coordination. The glycine carboxylate band appeared at 1592 cm^{-1} which is different from carboxylate band in free glycine.

This showed that the glycine coordinated through carboxylic group with metal(El-Said *et al.*, 2009). The comparison of FT-IR spectra of the complex with free cefixime showed that cefixime act as meltdentate ligand while the secondary ligand glycine act as Monodentate ligand.

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Complex 4

The melting point (decomposition) of product measured was 265˚C while the melting point (decomposition) of ligand was 220˚C. It also showed there is a change in the compound or complexation. The pH of the ligand sol was 4 while the pH of complex 4 was 7.7.

Ligand and product separate sol were prepared and their RF values were calculated as 0.7 and 0.63 respectively using methanol as mobile phase. It shows that the product has less affinity for methanol which gives an evidence in the change in the nature of compound or formation of complex.

Following is the FT-IR spectra of complex 4 (KBr, cm^{-}) 3587 (O-H stretching vibrations of carboxylate), 3277-3202, 2203-2116 (C=H stretching vibrations of aromatic ring), 1749 (C=O stretching vibrations of B-lactam carbonyl bond), 1716, 1540 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond),1317 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond), 709, 692-616 and 1575 (COOH asymmetric vibrations of carboxylate of glycine), 1386 (COOH symmetric vibrations of carboxylate of glycine), 2927(N-H_2 vibrations of amino group of glycine).

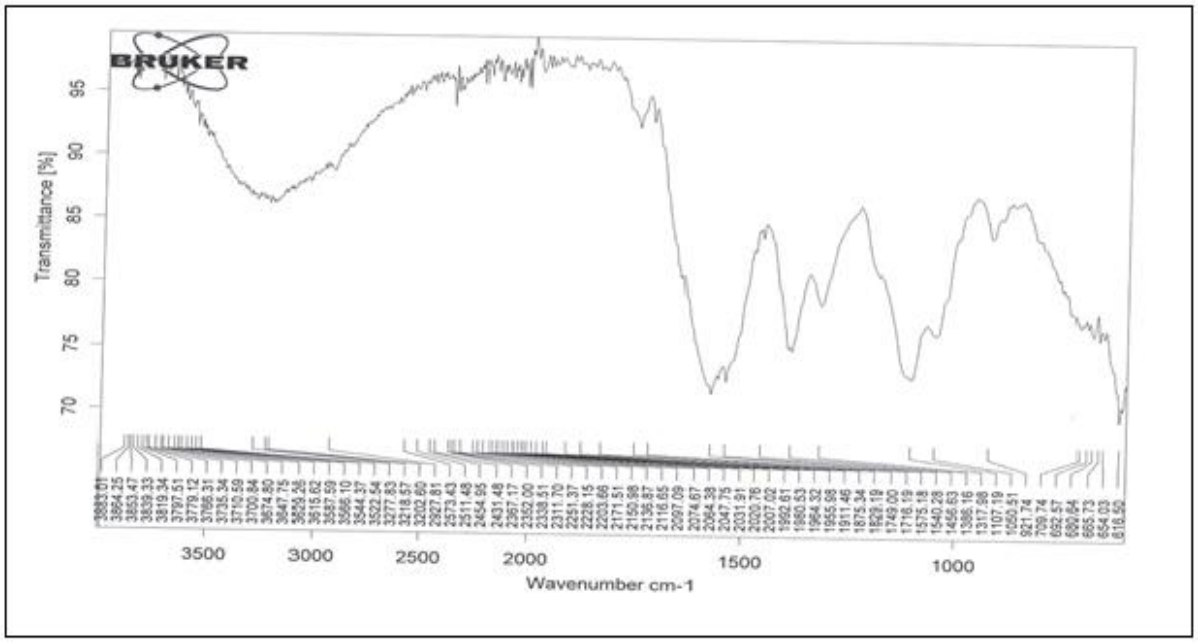

Fig. 6. FTIR spectrum of Complex 4.

The stretching vibrations of b-lactam amide carbonyl bond also appear at different position than the free ligand. This also showed coordination occurred at this position (Anacona and Estacio, 2006) overlapped stretching vibrations also occurred at 1540 cm^{-1} due to coordination. The glycine carboxylate band appeared at 1575 cm^{-1} which is different from carboxylate band in free glycine. This showed that the glycine coordinated through carboxylic group with metal(El-Said *et al.*, 2009).

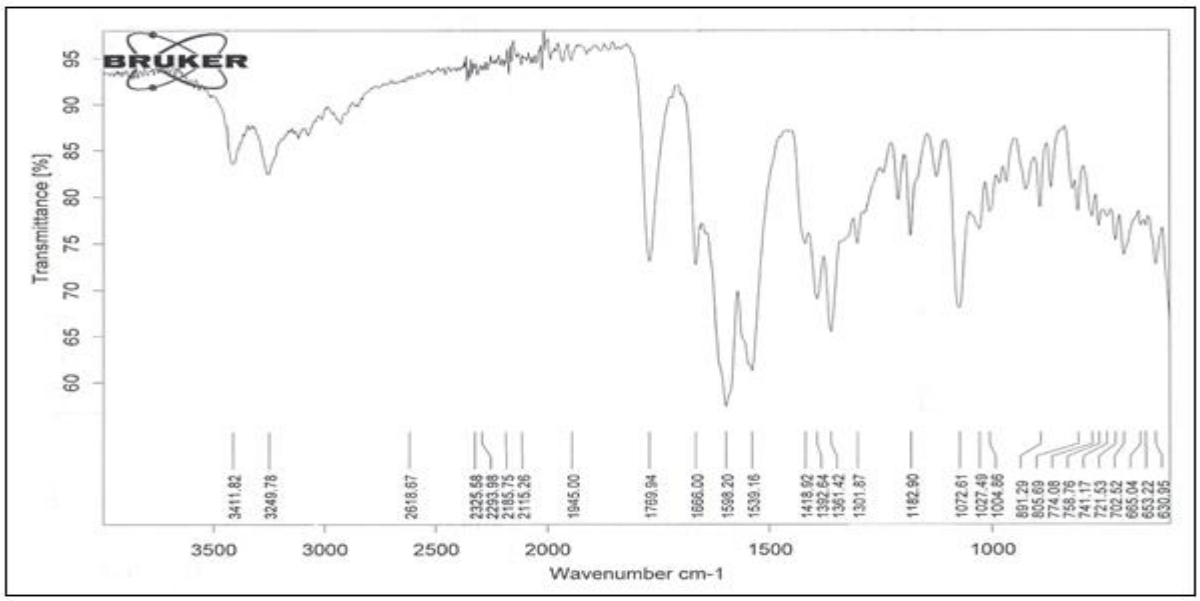

Fig. 7. FTIR spectrum of 5.

The comparison of FT-IR spectra of the complex with free cefixime showed that cefixime and glycine has coordinated through carboxylate group with metal atom. Complex 4 showed following antibacterial activity. No activity against Bacillus anthrax and Bacillus sabtilis. Zone of inhibition of 14 mm was measured against E.coli and 16 mm was measured against pseudomonas aeruginosa. The complex 4 showed no activity against Gram positive bacteria while the ligand showed mild activity. The complex 4

showed less activity against Gram negative bacteria as compared to free ligand.

Complex 5

The melting point (decomposition) of product measured was 250˚C while the melting point (decomposition) of ligand was 220˚C. The change in the decomposition point of complex reveals the formation of complex. The pH of the ligand sol was 4 while the pH of complex 5 was 7.2.

Rf values of the ligand and the complex 5 were measured as 0.7 and 0.64 respectively using methanol as mobile phase. The complex 5 showed less affinity for methanol as compared to ligand. This gives an evidence of the formation of complex.

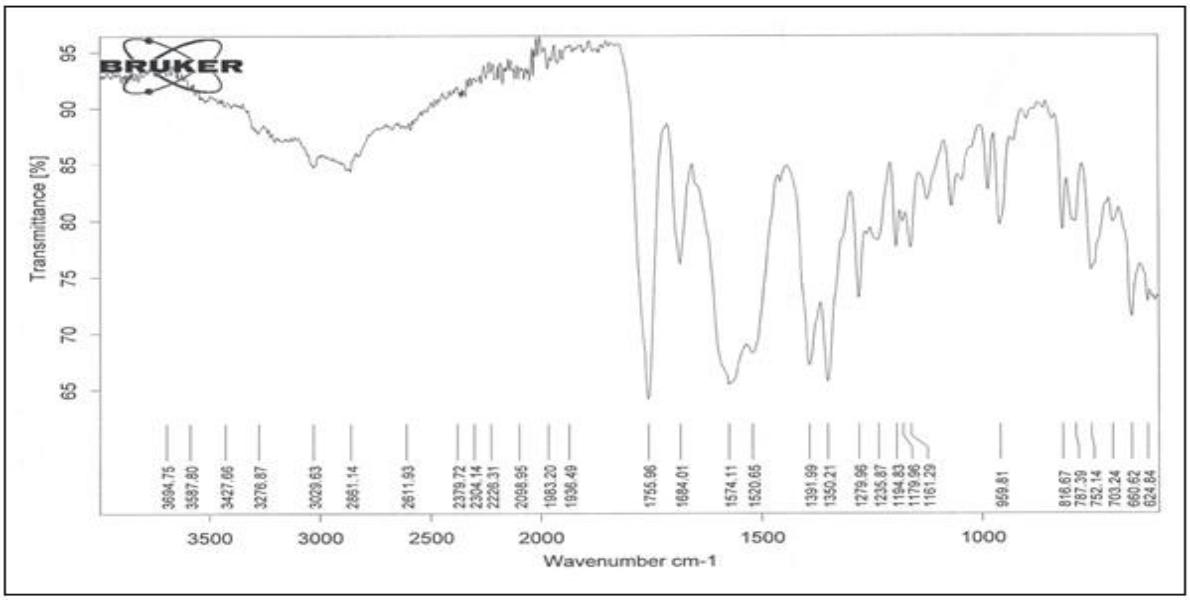

Fig. 8. FTIR spectrum of cefixime.

Following is the FT-IR spectra of complex 5 (KBr, cm^{-}) 3580 (O-H stretching vibrations of carboxylate), 3411, 3249, 2185-2115 (C=H stretching vibrations of aromatic ring), 1769 (C=O stretching vibrations of B-lactam carbonyl bond), 1666 (N-H stretching vibrations of B-lactam amide carbonyl bond),1539 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond),1361 (Overlapped asymmetric and symmetric stretching bands of coordinated carboxylate bond), 1182, 1072-1004, 977-903, 774-702, 665-630 and 1598 (COOH asymmetric vibrations of carboxylate of glycine), 1392 (COOH symmetric vibrations of carboxylate of glycine).

While comparing the FT-IR spectra of the complex 5 with the FT-IR spectra cefixime and glycine Fruitful results were obtained regarding complexation of the metal with ligand. The peak position of carbonyl stretching vibrations of the complex 5 occur at 1769 cm^{-1} has remarkable difference from cefixime which appear at 1755 cm^{-1}. Which showed some changes had been occurred at this position(Pillai and Latha, 2012). The carbonyl oxygen has full chances for coordination to metal ion at this position. The stretching vibrations of b-lactam amide carbonyl bond also appear at different position than the free ligand. This also showed coordination occurred at this position (Anacona and Estacio, 2006) . Overlapped stretching vibrations also occurred at 1539 cm^{-1} due to coordination. The glycine carboxylate band appeared at 1598 cm^{-1} which is different from carboxylate band in free glycine. This showed that the glycine coordinated through carboxylic group with metal (El-Said *et al.*, 2009). The comparison of FT-IR spectra of the complex with free cefixime showed that cefixime act as meltdentate ligand while the secondary ligand glycine act as Monodentate ligand.

When the antibacterial activities of complex 5 was

measured following were the zones of clearance calculated. Zone of inhibition against bacillus anthrax was 7 mm. Complex 5 showed no activity against Bacillus sabtilis. Zone of inhibition of 9 mm against E.coli and 10 mm against pseudomonas aeruginosa. Complex 5 showed less activity against bacteria as compared to free ligand.

References

Aiyelabola TO, Isaac O, Olugbenga A. 2012. Structural and antimicrobial studies of coordination compounds of phenylalanine and glycine. International Journal of Chemistry **4,** p 49.

ALI A. 2003. H2-Antagonist Cephalosporin Interactions. University of Karachi, Karachi.

Anacona JR, Estacio J. 2006. Synthesis and antibacterial activity of cefixime metal complexes. Transition metal chemistry **31,** 227-231. http://dx.doi.org/10.1007/s11243-005-6360-9

Arayne MS, Sultana N, Khanum F, Ali MA. 2002. Antibacterial studies of cefixime copper, zinc and cadmium complexes. Pakistan journal of pharmaceutical sciences **15,** 1-8.

Auda S, Mrestani Y, Fetouh M, Neubert R. 2008. Characterization and activity of cephalosporin metal complexes. Die Pharmazie-An International Journal of Pharmaceutical Sciences **63,** 555-561.

Cleare M, Hydes P, Hepburn D, Malerbi B, Prestayko A, Crooke S, Carter S. 1980. Cisplatin, Current Status and New Developments. New York, 149-170.

El-Said A, Aly A, El-Meligy M, Ibrahim M. 2009. Mixed ligand zinc (II) and cadmium (II) complexes containing ceftriaxone or cephradine antibiotics and different donors. The Journal of the Argentine Chemical Society **97.**

Fountain R, Russell A. 1969. Studies on the mode of action of some cephalosporin derivatives. Journal of Applied Bacteriology **32,** 312-321. http://dx.doi.org/10.1111/j.13652672.1969.tb00978.x

Hayes RL. The medical use of gallium radionuclides: a brief history with some comments. Seminars in nuclear medicine, 1978. Elsevier, 183-191.

Ingber DE. 1998. The architecture of life. Scientific American **278,** 48-57.

Kaushal R, Sheetal RK, Nehra K. Syntheses, Characterizations and Biological Screening of Titanium (IV) Complexes Derived from Antibiotic Drug (S).

Li J, Chen F, Cona MM, Feng Y, Himmelreich U, Oyen R, Verbruggen A, NI Y. 2012. A review on various targeted anticancer therapies. Targeted oncology **7,** 69-85. http://dx.doi.org/10.1007/s11523-012-0212-2

Li NC, Doody BE, White JM. 1957. Some Metal Complexes of Glycine Peptides, Histidine and Related Substances1. Journal of the American Chemical Society **79,** 5859-5863. http://dx.doi.org/10.1021/ja01579a006

Mahmoud WH, Mohamed GG, EL-Dessouky MM. 2015. Synthesis, structural characterization, in vitro antimicrobial and anticancer activity studies of ternary metal complexes containing glycine amino acid and the anti-inflammatory drug lornoxicam. Journal of Molecular Structure **1082,** 12-22. http://dx.doi.org/10.1016/j.molstruc.2014.10.014

O'callaghan CH. 1975. Classification of cephalosporins by their antibacterial activity and pharmacokinetic properties. Journal of Antimicrobial Chemotherapy **1,** 1-12. http://dx.doi.org/10.1093/jac/1.suppl_3.1

Patil SS, Thakur GA, Shaikh MM. 2012. Synthesis, spectral, thermal and antibacterial investigations of mixed ligand complexes of thorium (IV) derived from 8-hydroxyquinoline and some

amino acids. Acta poloniae pharmaceutica **69,** 1087-1093.

Pillai MS, Latha SP. 2012. Designing of some novel metallo antibiotics tuning biochemical behaviour towards therapeutics: Synthesis, characterisation and pharmacological studies of metal complexes of cefixime. Journal of Saudi Chemical Society. http://dx.doi.org/10.1016/j.jscs.2012.09.004

Reilly RM. 2007. The radiopharmaceutical science of monoclonal antibodies and peptides for imaging and targeted in situ radiotherapy of malignancies. Pharmaceutical Sciences Encyclopedia.

Sanoja W, Martínez JD, Araujo ML, Brito F, Hernández L, Del Carpio E, Lubes V. 2014. Stability constants of mixed ligand complexes of vanadium (III) with 8-hydroxyquinoline and the amino acids glycine, proline, α-alanine and β-alanine. Journal of Molecular Liquids, **197,** 223-225. http://dx.doi.org/10.1016/j.molliq.2014.05.012

Soliman M, Hindy A, Mohamed G. 2014. Thermal decomposition and biological activity studies of some transition metal complexes derived from mixed ligands sparfloxacin and glycine. Journal of Thermal Analysis and Calorimetry **115,** 987-1001. http://dx.doi.org/10.1007/s10973-013-3466-8

Sugiura Y, Takita T, Umezawa H. 1985. Bleomycin antibiotics: metal complexes and their biological action, Marcel Dekker: New York.

Vaidyanathan VG, Nair BU. 2003. Photooxidation of DNA by a cobalt (II) tridentate complex. Journal of inorganic biochemistry **94,** 121-126. http://dx.doi.org/10.1016/S0162-0134(02)00620-7