



Synthesis, Characterisation And Microbial Activity of Mixed 'Transition Metal - Calcium Tartarate' Complexes

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ABSTRACT

Many applications of tartrate compounds are reported globally. In the present research work we have synthesized a series of six new mixed transition metal complexes of the type $[MM'(C_4H_4O_6)_2 \cdot xH_2O]$ (where $M=Ca$ and $M= Mn, Fe, Co, Ni, Cu, Zn$) by using tartarate as a ligand and are characterised by different analytical techniques such as elemental analysis, TGA, FTIR, XRD, SEM, magnetic susceptibility study, UV-visible spectroscopy etc. Analytical data shows that all complexes exhibited 1:1 (metal: ligand) ratio. IR spectral data shows that bidentate ligand coordinate with metal ion in a bi dentate manner through the two 'O' atoms. TGA of complexes shows that degradation pattern of complexes were in good agreement with recommended formulae of the complexes. XRD technique shows that all the complexes are in polycrystalline in nature. SEM shows morphological features and surface characteristics of complexes. The synthesised metal complexes were then tasted in vitro for their biological activity against *Bacillus subtilis*, *Saccharomyces cerevisiae*, *Aspergillus niger* and *Escherchia coli* to assess their antibacterial and antifungal effects. The bioassays of all the complexes showed a greater inhibitory effect in the form of broad activity spectrum than the individual ligand, which indicates after the coordination the antimicrobial activity is increased.

Keywords: Alkaline earth transition metal complexes, TGA, Octahedral geometry, Biological activity.

INTRODUCTION

Applications of different tartarate complexes are reported on large scale [1], for example: sodium-potassium tartrate is having ferroelectric application [3]. Potassium- chromium tartarate is used in medicine [4] and antimony- barium tartarate in veterinary drugs [5]. Calcium- strontium mixed leavo tartarate [6], iron- manganese leavo tartarate are also reported. The tartarates also find applications in science and technology such as ferroelectric applications [11,12,15,16,17]. They are used for transducers and many linear and non-linear mechanical devices [18,19,26,]. So many metal complexes of Schiff base ligands with N and O donor atoms with their important properties such as catalytic activity and different

biological activities like antimicrobial, antifungal, antiviral [8,13,14] are reported in literature, but the metal complexes of ligand containing 'O' donor atoms are reported very less.

Although a wide variety of metal ligand complexes were studied for their antimicrobial activity, the literature survey reveals that very less work has been done on combination of mixed metal -ligand complex to study the biological activity. To find some new active metal complexes we have synthesized and characterised mixed metal tartarates containing tartarate as a ligand which may coordinate through carboxylate and carbonyl oxygen with both the metal ions such as Mn (II), Fe (II), Co(II), Ni(II) [1,2]. The biological activity of metal ions with the tartarate ligand has improved as compared to the ligand. The synthesis of complex by co precipitation technique using water as a solvent is a low cost and offer high yield [1,2].

The present work deals with synthesis, characterisation and antimicrobial activity study of some mixed metal tartrate complexes of the type $[MM'(C_4H_4O_6)_2 \cdot xH_2O]$ which contain $M' = Ca$ as a fixed metal ion along with the series of transition metal ions i.e. $M = Mn, Fe, Co, Ni, Cu, Zn$ bonded together through bidentate tartarate ligand. Tartarate is having donor site of carboxyl 'O' atom. These complexes are characterised by different analytical techniques such as elemental analysis, TGA, FTIR, XRD, SEM, magnetic susceptibility study, UV-visible spectroscopy etc. The complexes have been screened for their biological activity [22-25].

MATERIALS AND METHODS

Materials: All the hydrated metal salts ($CaCl_2 \cdot 2H_2O$, $MnCl_2 \cdot 4H_2O$, $FeCl_3$, $CoCl_2 \cdot 6H_2O$, $NiCl_2 \cdot 6H_2O$, $CuCl_2 \cdot 2H_2O$ and $ZnCl_2$) and reagents are of AR grade. Distilled water is used as a solvent for preparation of solutions of metal salts and complexes.

Synthesis of precursors: All the complexes were prepared by a simple co precipitation method. The two metal salts aqueous solution i.e., $CaCl_2 \cdot 2H_2O$ and $MnCl_2 \cdot 4H_2O$ were mixed homogeneously in a fixed molar ratio. The pH of the medium was adjusted to a low enough value ($pH < 6$) so that precipitate of hydroxide does not form. The solution is stirred with the help of magnetic stirrer. The Sodium tartarate (15%) solution is then added slowly with constant stirring till a permanent precipitate occurs. Acetone is added in equal amount to solution, where addition of acetone not only ensures a high yield, but also influences more homogeneous, stoichiometric, fine grained powders. The precipitate is filtered after stirring it for 30 minutes. The filtrate is checked for Ca^{2+} and Mn^{2+} ions whose absence ensured complete co-precipitation. The precipitate is washed with acetone and then dried under normal atmospheric conditions, and then stored in desiccators. All the metal complexes are of different colours which are prepared using this similar general procedure.

Antimicrobial activity: Mixed metal complexes are well known for their biological activity [22-25,29-34]. All the synthesized mixed metal complexes were screened for antibacterial as well as antifungal activity.

Materials and methods: Using dilute HCl 0.1 % solution of each chemical was prepared. For every chemical solution four nutrient agar plates were used and labelled for four bacterial cultures of *E. Coli*, *Bacillus subtilis*, *Saccharomyces cerevisiae* and *Aspergillus niger* in total 8 sets of plates (2 plates in each set and 3 samples in each plate) were prepared. In each set of plates, 0.5 mL of above bacterial cultures were spread, inoculated and incubated at $37^\circ C$ for 30 min to absorb the culture on medium surface. Using well borer, 4 wells are bored (3 for samples and 1 for control) in each plate, aseptically. 0.1 mL of solution of each chemical is poured aseptically in each respective well and incubated for diffusion at $4^\circ C$ for 1 h. All the plates were incubated at $37^\circ C$ for 48 h and results are recorded as zone of inhibition.

RESULTS AND DISCUSSION

Characterization of precursors: All synthesised metal complexes are obtained with good percentage yield. Data is summarised in table 1. Complexes are powdered solids, quite stable towards air and moisture at room temperature and decomposition temperature is quite high.

Table 1: Complex composition

Complex	Symbol	Mol.Wt	Amount of CaCl ₂ .2H ₂ O (g)	Amount of metal salt (g)	Tartarate solution added	% Yield of complex
MnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A1	481	4.584	6.174	15%	70
FeCa(C ₄ H ₄ O ₆) ₂ 10H ₂ O	A2	571.85	3.859	4.254	15%	63
CoCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A3	485	4.546	7.358	15%	75
NiCa(C ₄ H ₄ O ₆) ₂ 8H ₂ O	A4	538.69	4.093	6.618	15%	69
CuCa(C ₄ H ₄ O ₆) ₂ 2H ₂ O	A5	435.5	5.063	5.871	15%	72.5
ZnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A6	491.4	4.487	4.159	15%	77

Elemental analysis: C, H analysis done on vario MICRO CHNS, serial number: 151 14035. The elemental analysis made in weight percent of tartarate precursors for metals, C and H are very well matched with the calculated ones. The data is shown in table 2. Analytical data shows that metal chelates have 1:1:2 stoichiometry. So the general molecular formula of the synthesized complexes can be given as [MM'(C₄H₄O₆)₂.xH₂O] (where M'=Ca and M= Mn , Fe, Co, Ni, Cu, Zn)

Table 2: Elemental analysis data

Formula	Symbol	Mol. Wt.	C		H		Transition Metal (%)		Ca		Magn. Mom. (μ)B.M
			Obs.	Cal.	Obs.	Cal.	Obs.	Cal.	Obs.	Cal.	
MnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A1	481	18.34	19.96	4.48	3.74	9.20	9.87	6.67	7.18	5.65
FeCa(C ₄ H ₄ O ₆) ₂ 10H ₂ O	A2	571.85	15.47	16.7	3.18	4.8	8.80	9.40	5.82	6.73	4.62
CoCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A3	485	19.52	19.79	3.66	3.71	9.87	10.18	6.23	6.90	3.55
NiCa(C ₄ H ₄ O ₆) ₂ 8H ₂ O	A4	538.69	16.14	17.82	3.46	4.45	9.50	10.14	7.30	6.91	2.60
CuCa(C ₄ H ₄ O ₆) ₂ 2H ₂ O	A5	435.5	21.07	22.04	2.13	2.75	10.54	11.22	7.87	7.07	1.75
ZnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A6	491.4	18.75	19.5	4.20	3.66	10.60	11.17	6.01	6.83	00

Thermo gravimetric analysis: The thermo analytical measurements of complexes were obtained with Perkin-Elmer (Delta series-TGA7) instrument. All complexes are studied under static atmosphere of air using 30-50 mg samples and providing heating rate up to 10 deg min⁻¹ in the range of ambient to 500°C. TGA data is summarized in table 3. TGA curves of the complexes are shown in the figure 1. All the precursors has different number of water of hydration, hence it shows continuous mass loss between 60 to 150°C indicating the loss of water molecules from the complexes at this temperature [7]. The % loss of water molecules for all precursors is well matched with the theoretical values. The oxidative decomposition of the ligand is observed between different temperature range 150 to 250°C (table 3).

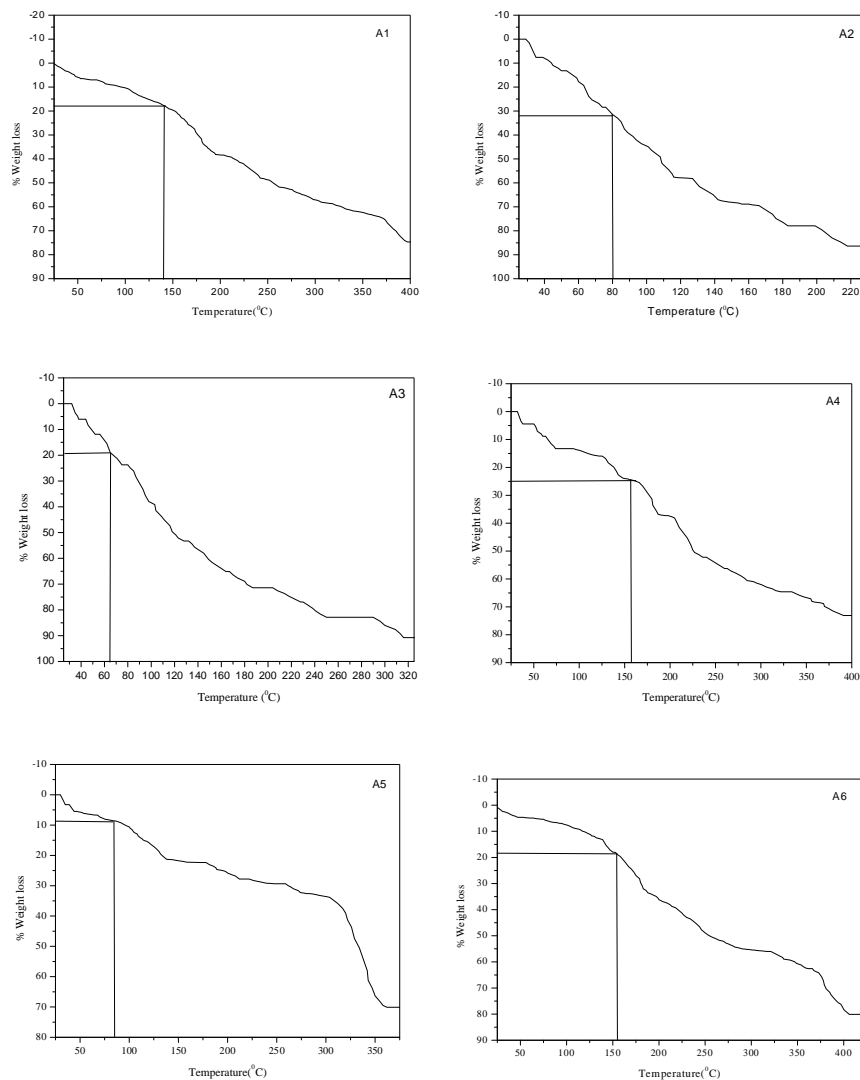


Figure 1 : TGA of complex A1 to A6

Table 3: TGA data of complexes under static air

Complex	Sample	% mass loss	% mass loss 2	Temp. Range °c
		Obsd.	Calcd.	
MnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A1	17		60-140
		38.66	18.71	140-195
		48.06		195-245
FeCa(C ₄ H ₄ O ₆) ₂ 10H ₂ O	A2	32.50		55-85
		57.97	31.47	85-120
		70.38		120-170
CoCa(C ₄ H ₄ O ₆) ₅ H ₂ O	A3	19.61		60-70
		53.53	18.55	70-140
		71.00		140-200
NiCa(C ₄ H ₄ O ₆) ₂ 8H ₂ O	A4	25.19		90-160
		38.44	26.73	160-200
		60.48		200-285
CuCa(C ₄ H ₄ O ₆) ₂ 2H ₂ O	A5	8.82		60-90
		22.36	8.26	90-150
		34.00		150-300

ZnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	A6	18.26		100-150
		33.30	18.31	150-190
		50.95		190-260

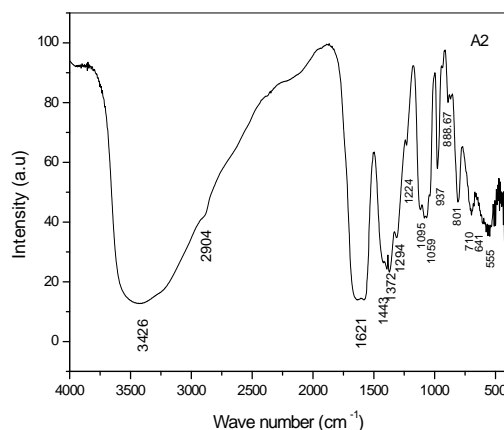
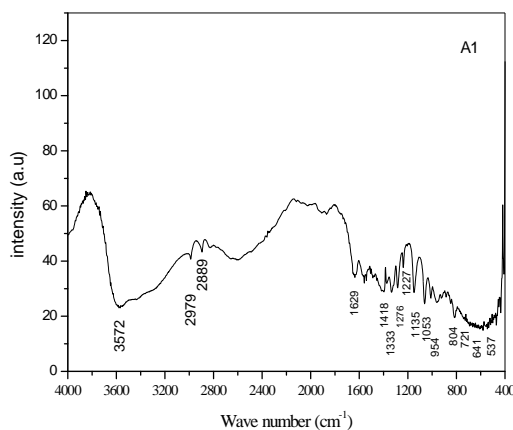
IR spectral study: IR spectrum of precursors was taken by the instrument JASCO FTIR 4100 SEREIS, and is shown in the fig.2 for all the complexes. The bidentate linkage of metal ions at donor sites of the ligand tartrate was confirmed by IR spectral assessment of the ligand and its metal complexes. The observed absorption frequencies and their assignments in relation to their characteristic vibrational modes are given in table 4. The strong intensity band at 1750 cm⁻¹ in di- tartarate is shifted to lower wave numbers at Vasy (OCO) 1611 cm⁻¹ and Vasy (OCO) 1428-1372cm⁻¹ for complexes reveals the coordination of both COO- group in tartarate molecule to metallic ions.

It was also reported that bidentate coordination of carboxylate group with metal results in lowering of both ν (COO) frequencies due to drainage of electron density from carboxylate group to the metal ion. Hence on the basis of difference between antisymmetric and symmetric (C=O) stretching frequencies of tartarate complexes, bidentate nature of attachment of ligand to metal ion can be predicted. The broad and strong trough positioned in between 3427-3414 cm⁻¹ assigned to the -OH vibration frequency of water and secondary alcohol group which shows that complexes are hydrous. The bands at 543-520 cm⁻¹ correspond to the M-O bonding. The bands ranging from 1380-1040 cm⁻¹ are the bands corresponding to the C-O bonding in the tartarate of the complexes [7,9,10,20,21,31,34].

Table 4: Infra-Red spectral bands and their probable assignments in [cm⁻¹]

Sr. No	A1	A2	A3	A4	A5	A6	Assignments
1	3414	3417	3414	3414	3414	3427	ν asy (H-OH)
2	1613	1615	1611	1611	1612	1614	ν asy (C=O)
3	1421	1423	1418	1428	1419	1411	ν sy(C=O),asy(c-c)
4	1373	1372	1368	1368	1368	1331	ν sy c-o(carboxyl)
5	1276	1230	1282	1286	1236	1280	ν asy C-C
6	1235	1230	1236	1236	1226	1230	ν asy C-O
7	1087	1090	1114	1104	1104	1087	ν C-O(alcohol)
8	1060	1060	1053	1047	1053	1057	ν C-O(alcohol)
9	915	928	922	922	922	926	ν sym C-O),d(o-c=o)
10	730	725	729	729	739	722	ν sym C-C
11	642	634	638	638	648	641	ν (H-O-H)
12	541	543	527	527	527	520	ν (M-O) (C-C)

A1- Mn Ca(C₄H₄O₆)₂5H₂O, A2- Fe Ca(C₄H₄O₆)₂10H₂O, A3- Co Ca(C₄H₄O₆)₂5H₂O, A4- Ni Ca(C₄H₄O₆)₂8H₂O, A5- Cu Ca(C₄H₄O₆)₂2H₂O, A6- Zn Ca(C₄H₄O₆)₂5H₂O



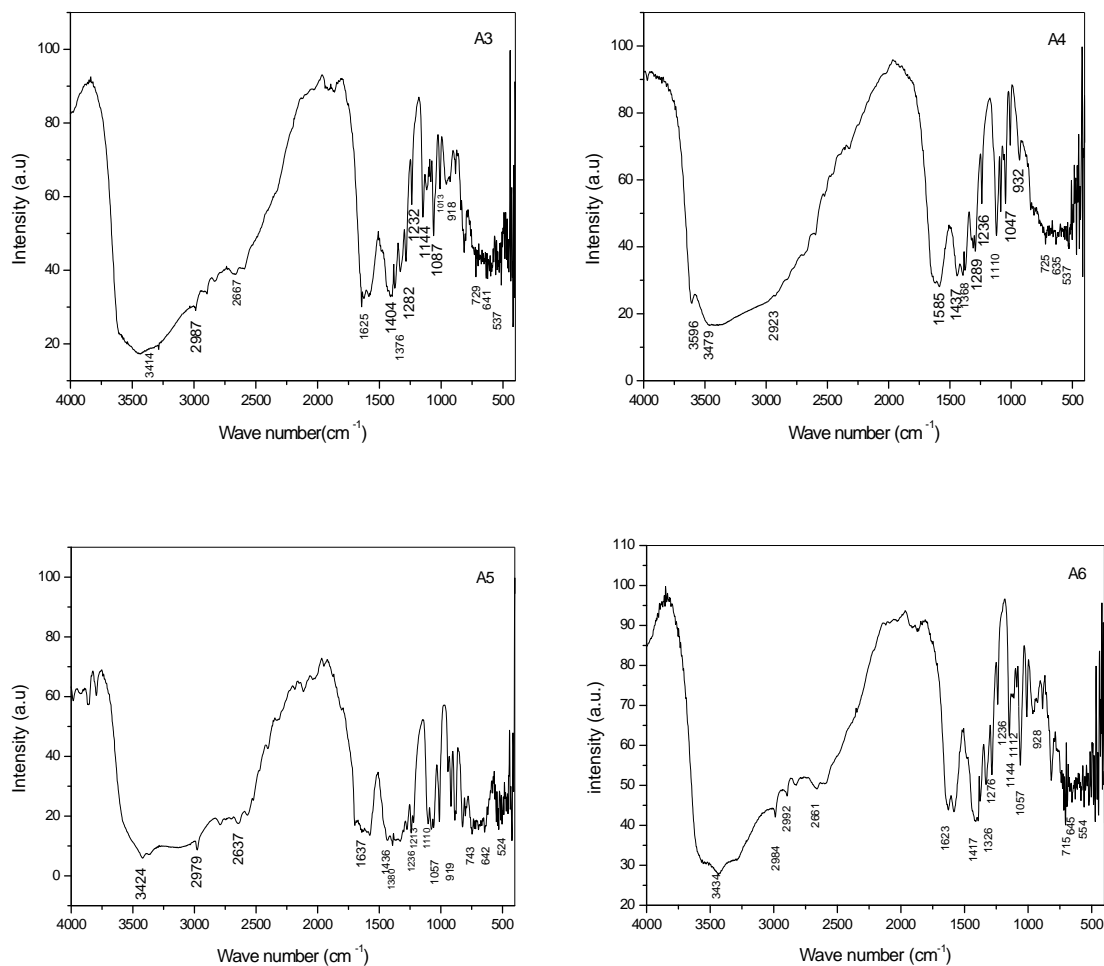


Figure 2: Infra-Red spectra of complexes A1 to A6

X-Ray diffraction: The X-Ray powder pattern of these complexes were taken on BRUKER D8 ADVANCE XRD instrument and shown in fig.3. The XRD pattern shows complexes are polycrystalline in nature. The observed 'd' spacing values and Particle size calculated using formula ($D=0.89\lambda / \beta \cos\theta$) are given in table 5 and 6 respectively [7,27,28,30,33].

Table 5: Observed 'd' spacing values of complexes in Å^o

A1	A2	A3	A4	A5	A6
4.1347	3.6027	4.0953	4.0116	4.3483	4.0953
3.9108	3.3927	3.6907	3.6667	4.0768	3.4891
3.2685	2.8010	3.5215	3.4598	3.4972	3.2755
3.0261	2.5439	2.9905	3.1274	2.9127	3.0261
2.6682	2.4235	2.8584	2.8061	2.7824	2.8044
2.5245		2.6698	2.6259	2.5439	2.6791
2.3434		2.5508	2.4412	2.4387	2.5481
2.3376		2.2333	2.3043	2.3493	2.3330
2.2333		2.0635	2.1950	2.1404	2.2323
2.0537		1.9598	2.1531	2.0528	2.0555
1.9424		1.8748	1.9262	1.9132	1.9582
1.8726		1.7294	1.8597	1.6497	1.8697
		1.6563			

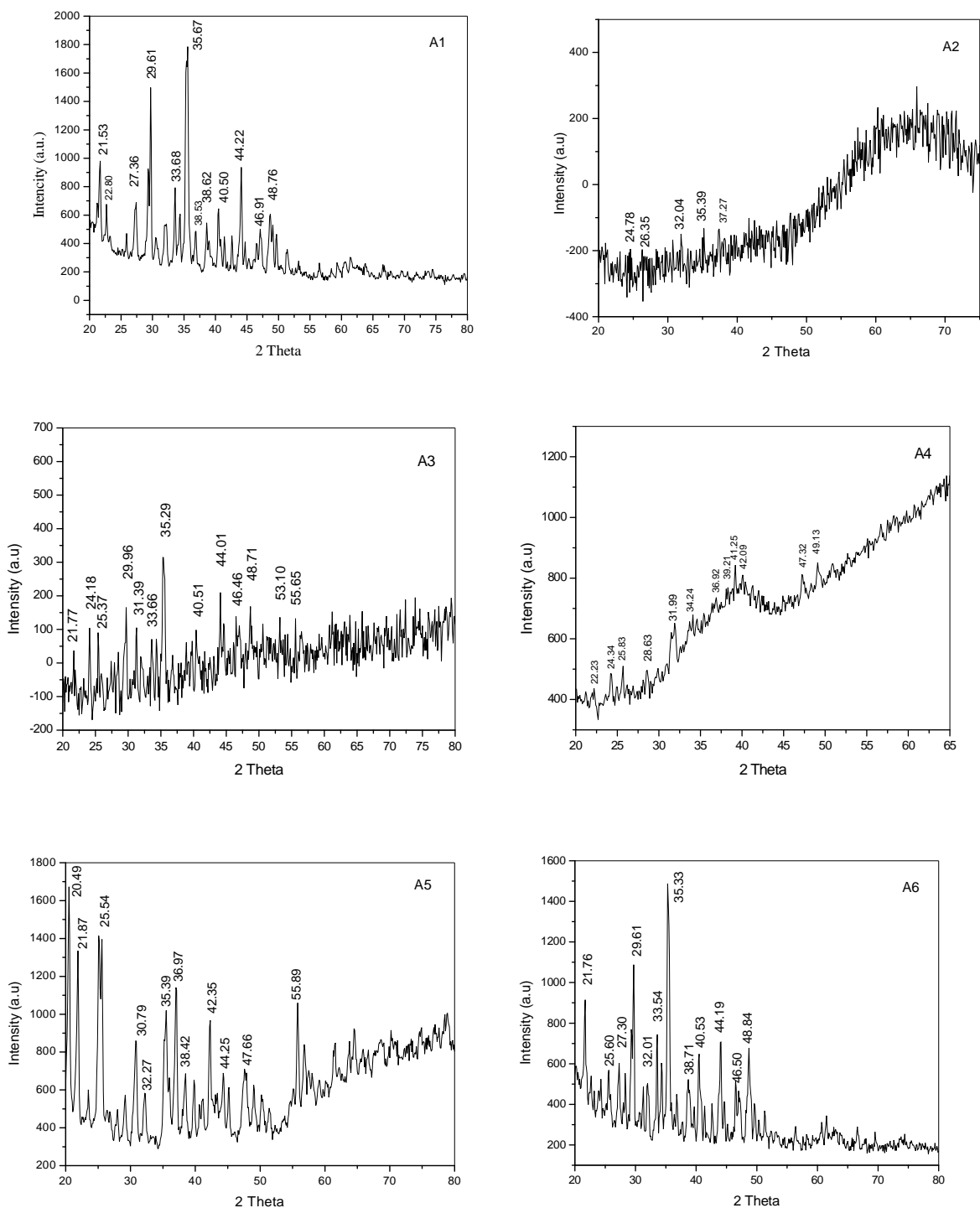


Figure 3: X-Ray diffraction patterns of complexes

Table 6: Observed particle size ($D = 0.89 \lambda / B \cdot \cos \theta$)

A1	A2	A3	A4	A5	A6
311.07\AA^0	139.87\AA^0	411.69\AA^0	182.52\AA^0	522.11\AA^0	391.21\AA^0

SEM: The SEM images of complexes were taken on JOEL SEM 6300 and are shown in fig.4. The particle size of complexes calculated varies in the range shown in table7. The scanning electron microscope reveals the morphology of the particle as octahedral particles embedded in rock like structure [29].

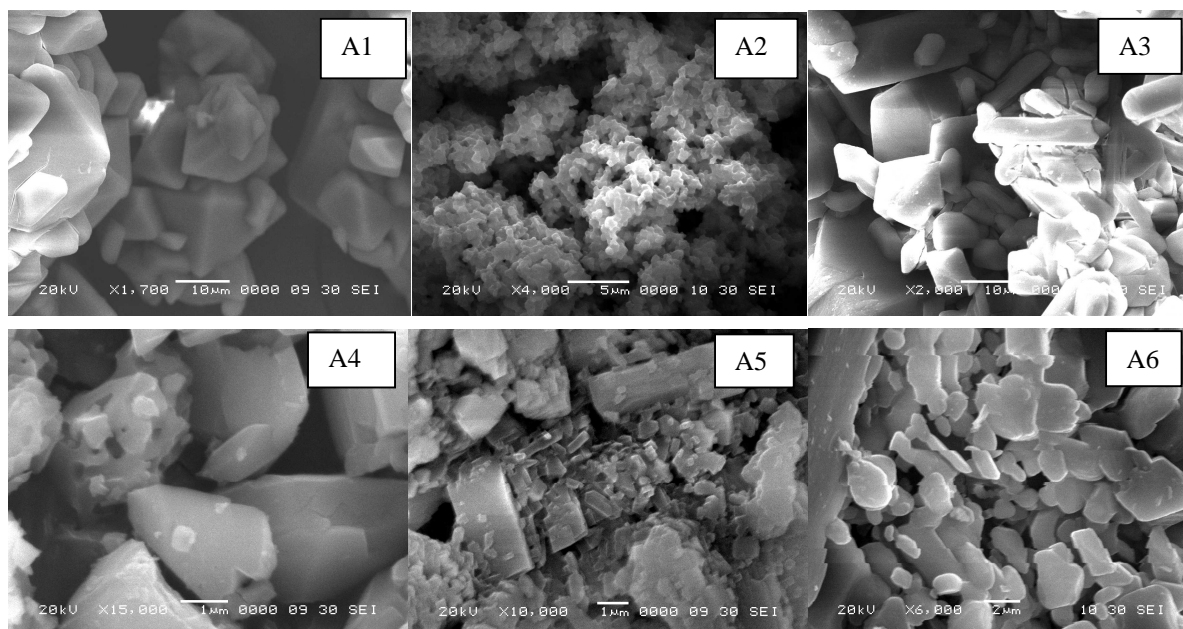


Figure 4: SEM images of complexes A1 to A6

Table 7: Range of particle size for A1 TO A6 (in μM) from SEM

Sr. No	Complex	Range in μm
1	$\text{MnCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 5\text{H}_2\text{O}$	3.03 to 9.69
2	$\text{FeCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 10\text{H}_2\text{O}$	0.65 to 2.23
3	$\text{CoCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 5\text{H}_2\text{O}$	4.68 to 9.37
4	$\text{NiCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 8\text{H}_2\text{O}$	0.43 to 0.78
5	$\text{CuCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 2\text{H}_2\text{O}$	0.3 to 0.5
6	$\text{ZnCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 5\text{H}_2\text{O}$	1.0 to 1.9

Magnetic Moments: The magnetic susceptibility measurements for metal (II)-Ca (II) tartarate have been done at 27 ± 1 °C on Faraday balance using 7000 gauss magnetic field. With respect to the number of unpaired electrons present in the complex their magnetic moment values are mentioned in table 2. For Zinc (II) Ca (II) tartarate precursor, the magnetic moment value is zero which reveals that no unpaired electrons are present in the precursor and hence results diamagnetism [32].

UV-Visible: The UV-Vis. spectrums of all complexes were taken on JASCO UV-VISIBLE 670 Series the plot of absorbance versus wavelength for all complexes are shown in fig.5. It shows that the complexes A2, A4 and A5 have high absorption in the ultra violet region at about 374.438nm, 398.633nm and 319.22nm of the spectrum (table 8). This makes the material to be suitable for devices for good absorption of UV radiation that is it can be used as a UV filters. The band gap (E_g) for the complexes are calculated by formula $E = hc / \lambda$, where c is velocity of light and λ is the wavelength. The E_g for the complexes A2 and A4 found 3.31 eV and 3.11 eV respectively which shows that complexes are semi conductor (table 8) [10].

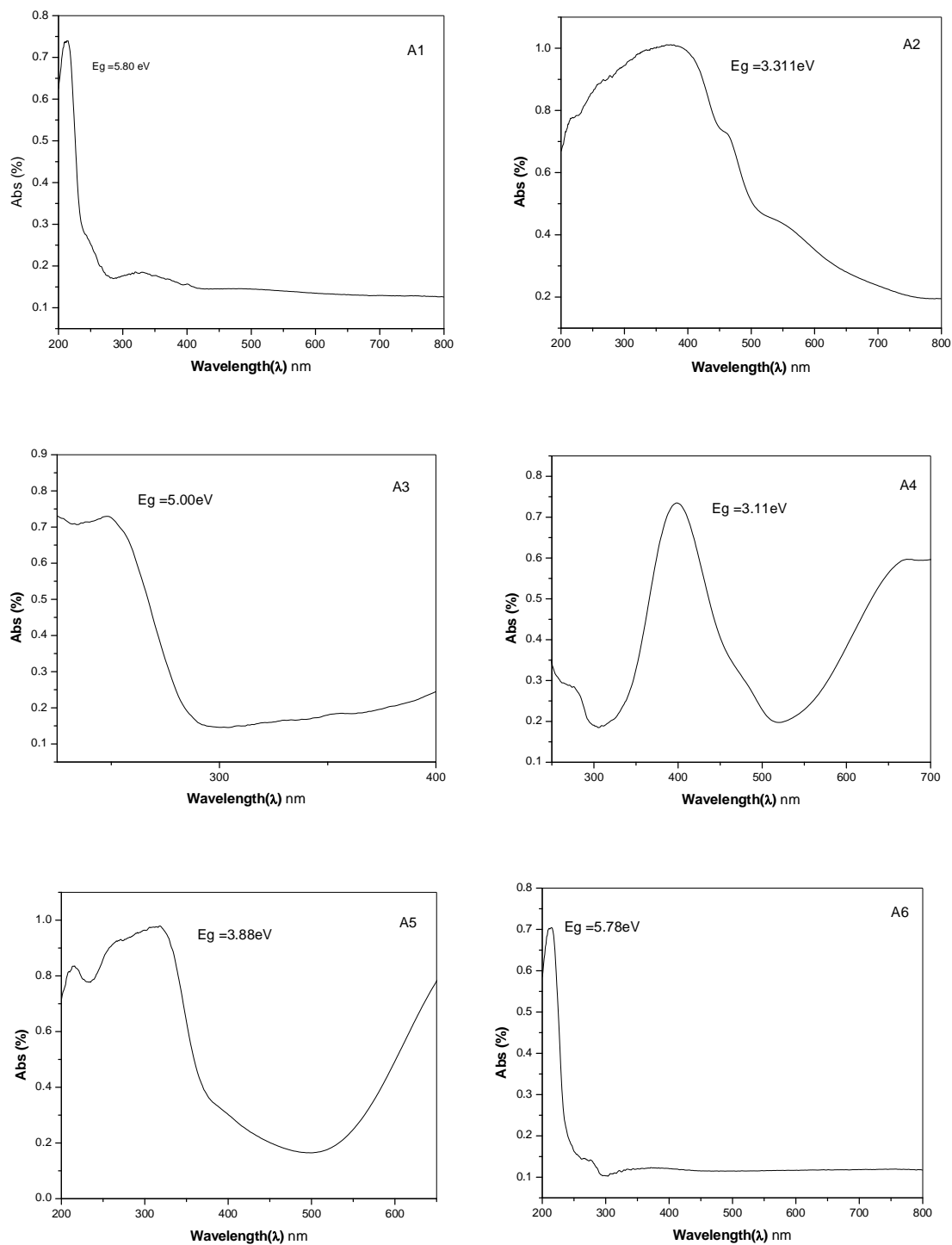


Figure 5: The plot of absorbance versus wavelength of complexes A1 to A6

Table 8: λ Max and corresponding E_g (band gap) of A1 TO A6

Sr.No	Complex	λ max (nm)	E_g (eV)	conductivity
1	$\text{MnCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 5\text{H}_2\text{O}$	213.480	5.80	insulator
2	$\text{FeCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 10\text{H}_2\text{O}$	374.438	3.31	semiconductor
3	$\text{CoCa}(\text{C}_4\text{H}_4\text{O}_6)_2 \cdot 5\text{H}_2\text{O}$	247.988	5.00	insulator

4	NiCa(C ₄ H ₄ O ₆) ₂ 8H ₂ O	398.633	3.11	semiconductor
5	CuCa(C ₄ H ₄ O ₆) ₂ 2H ₂ O	319.22	3.88	insulator
6	ZnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	214.258	5.78	insulator

Structure of the complex: The probable structure of complexes is shown in figure 6 [7].

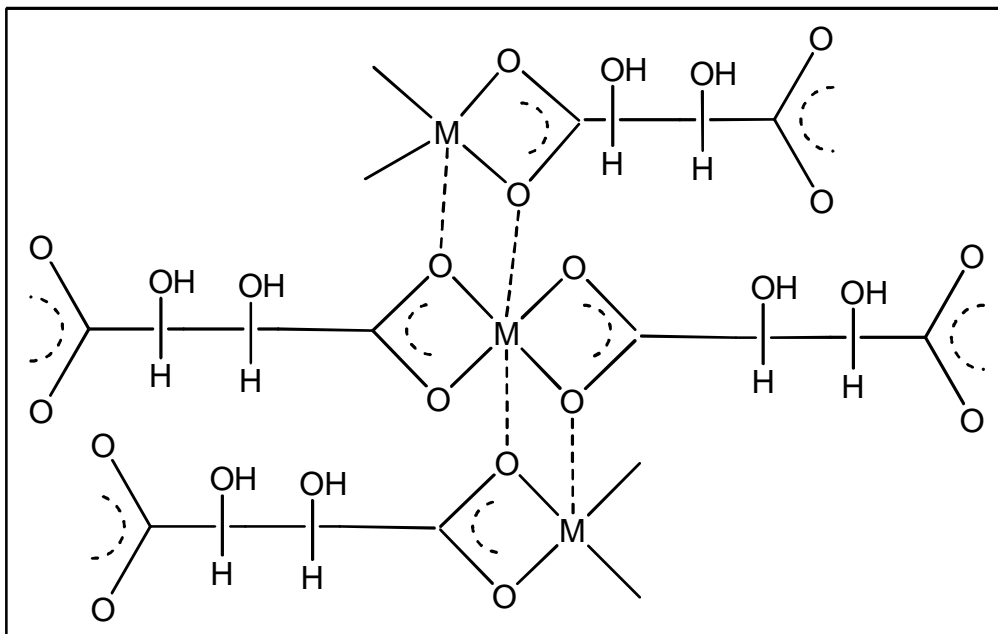
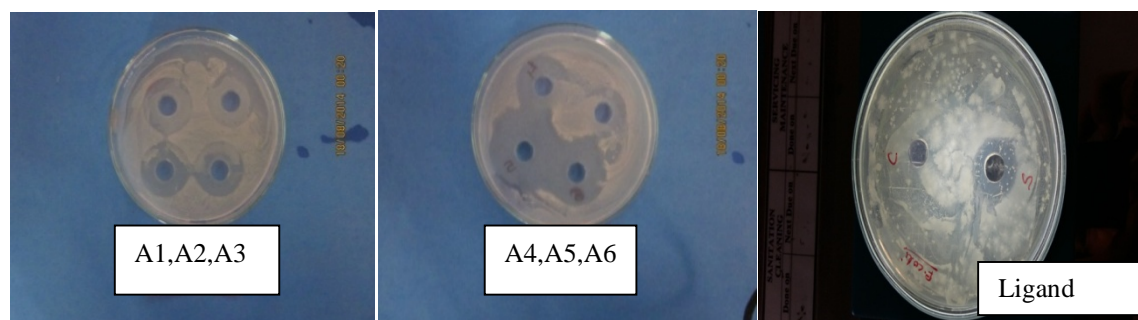
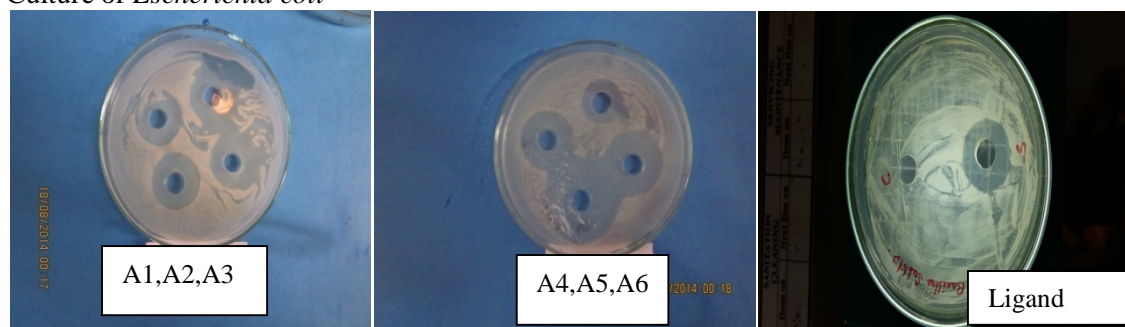
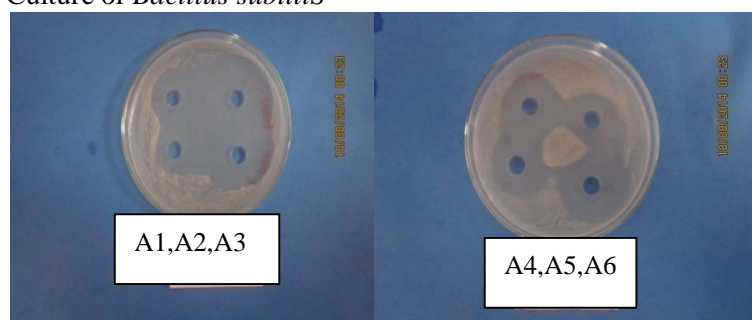
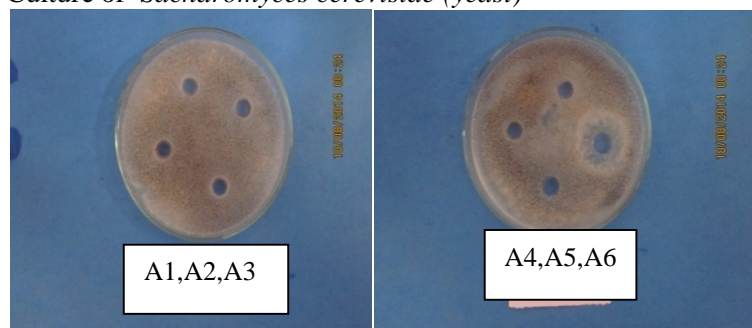


Figure 6: Probable structure of complex

APPLICATIONS

Antimicrobial activity: All the complexes (A1-A6) showed antibacterial and antifungal activity against microorganism (table 9).

It is evident from result that in case of bacteria gram +ve *Bacillus subtilis* is more sensitive than gram -ve *E.Coli*. Both the bacteria have shown good sensitivity to all the six complexes ranging from 3-22 mm. inhibition in *E.Coli* (excluding control zone) and 6-22mm in case *Bacillus subtilis*. The best activity against both bacteria was shown by complex A5. In case of yeast and mould, very good sensitivity was shown by yeast *Saccharomyces cerevisiae* ranging from 20mm-34mm (excluding control zone) and best against complexes A1 and A2, while *Aspergillus niger* has shown fair resistance to complexes A1, A2, A5, and A6, while low sensitivity to complex A3 (4mm) and complex A4 (12mm) (excluding control zone). It seems that complexes are fairly active against gram negative and gram positive bacteria and yeast, while least active against mould. As per literature, increased antimicrobial activity of metal complexes may be due to the chelation of metal ions with tartarate ligand which gives delocalization of pi-electrons across the entire chelate ring and hence lipophilicity is increased. This increased lipophilicity enhances the penetration of complexes in to the lipid membranes of microorganism and also disturbs their respiration process. Hence further synthesis of proteins get blocked which cause restriction in extension of growth of the organisms. The whole data assessment shows that these chemical complexes have potential to be used in the drug formulations against bacteria and yeast. The photo plates are shown in fig.7. The ligand has less activity towards bacteria.

Culture of *Escherichia coli*Culture of *Bacillus subtilis*Culture of *Sacharomyces cerevisiae* (yeast)Culture of *Aspergillus niger* (mold)**Figure 7 :** Antimicrobial activity**Table 9:** Antibacterial and antifungal activity studies of complexes: zone of inhibition (in mm)

Sr.No	Name of complex	Formula Wt.	<i>E.coli</i> (bact. +ve)	<i>B.subtilis</i> (bact. -ve)	<i>Sacharomyces cerevisiae</i> (yeast)	<i>Aspergillus niger</i> (mold)
1	MnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	481	21	25	45	11
2	FeCa(C ₄ H ₄ O ₆) ₂ 10H ₂ O	571.85	25	24	51	10
3	CoCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	485	25	26	27	14
4	NiCa(C ₄ H ₄ O ₆) ₂ 8H ₂ O	538.69	27	28	30	22
5	CuCa(C ₄ H ₄ O ₆) ₂ 2H ₂ O	435.5	40	38	28	10

6	ZnCa(C ₄ H ₄ O ₆) ₂ 5H ₂ O	491.4	32	30	40	10
7	Control		18	18	18	10
8	ligand		10	10	-	-

CONCLUSIONS

The results obtained can be summarised as follows

The elemental analysis of all complexes and C, H data is well matched with expected ones. The IR spectral study suggest that the tartarate ligand coordinate with the metal ions in bidentate fashion through O, O donor sites, forming a continuous polymeric structure with each metal ion surrounded by ligand in octahedral manner. The X-ray diffraction patterns of these complexes suggest that complexes are polycrystalline in nature. Thermo gram gives the information about number of water molecules and oxidative decomposition of the ligand. Over all study suggest that the metal chelate exhibited polymeric octahedral structure (fig.6) The SEM of complexes has shown considerable uniform distribution of octahedral particles with variable sizes.

A magnetic moment value suggests the paramagnetism and diamagnetism of precursors. The FTIR spectrum of the complexes has shown the broad trough positioned in between 3427-3414 cm⁻¹ corresponds to O-H bonding confirming the hydrous nature of the complexes. The spectrum has shown the presence of M-O bond. The complexes A2, A4 and A5 have high absorption at about 374.438nm, 398.633nm and 319.22nm respectively in the UV region of the spectrum makes the material to be suitable for UV filters. The energy gap of the complexes A2 and A4 is deduced as 3.31eV and 3.11eV which confirms the semiconducting nature of the compound. Antimicrobial study reveals that complexes possesses antibacterial as well as antifungal activity and have potential to use in the drug formulations against bacteria and yeast.

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