

SYNTHESIS, SPECTROSCOPIC BEHAVIOR AND SCANNING ELECTRON MICROSCOPY OF NEW COMPLEXES OF THEOPHYLLINE WITH SOME TRANSITIONAL METALS IONS

ELEONORA MARIAN^{1*}, SIMONA CAVALU¹, TUNDE JURCA¹,
FLORIN BĂNICĂ¹, IOAN BRATU²

¹*University of Oradea, Medicine and Farmacy Faculty, Nicolae Jiga 29, Oradea, Romania*

²*National Institute for Research and Development of Isotopic and Molecular Technologies, Cluj-Napoca, Romania*

*corresponding author: emarian@uoradea.ro

Abstract

In this study, four newly synthesized theophylline (TEO) complexes with transition metal ions (Cu^{2+} , Cd^{2+} , Co^{2+} and Zn^{2+}) are characterized by Fourier transform infrared spectroscopy (FTIR), EPR (electron paramagnetic resonance) and SEM (scanning electron microscopy). FTIR analysis confirmed the existence of these complexes combination by the presence of the characteristic band metal-TEO in the range $500\text{-}700\text{ cm}^{-1}$, the metal being coordinatively coupled to the nitrogen atom no. 7 and to the oxygen atom no. 6 of theophylline acting as a ambidentate ligand. Scanning electron microscopy demonstrated that Co, Cd and Zn complexes present similar microstructures with parallelepiped shapes, whereas the Cu complex presents a mixture of needle-like forms, irregular and lamellar flakes.

Rezumat

În această lucrare, patru noi complecși sintetizați din teofilină și ioni ai metalelor tranziționale (Cu^{2+} , Cd^{2+} , Co^{2+} and Zn^{2+}) sunt caracterizați prin FTIR, EPR (rezonanță electronică de spin) și SEM (microscopie electronică de baleiaj). Analiza prin spectrometrie FTIR a confirmat existența acestor complecși prin prezența benzilor caracteristice legăturilor metal-teofilină în regiunea $500\text{-}700\text{ cm}^{-1}$, metalul coordinându-se la atomul de azot 7 și la oxigen 6 din teofilină, care este un ligand ambidentat. Microscopia electronică de baleiaj demonstrează o microstructură similară pentru complecșii de Co, Cd și Zn de formă paralelipipedică, în timp ce complexul de Cu este un amestec de aglomerări de particule cu diverse forme aciculare, neregulate și lamelare.

Keywords: theophylline complexes, transitional metals, FTIR, EPR, SEM.

Introduction

The metallic ions (especially those of the transitional metals from 3d series, natural chelate compounds) take part of all the fundamental biologic processes: solar energy capture, photosynthesis, oxygen and nitrogen fixation and transportation, coordination of all metabolic reactions and controlled discharge of the energy in the biologic systems. Furthermore, the formation of complex combinations has implications in the explanation

of some biologic processes or pathological states and may constitute models for the study of the drugs' pharmacodynamic effects. Because of their potential pathological effects, investigation of metal (II) compounds containing aromatic nitrogen bases [9] or biological ligand has recently provoked a pronounced interest. The complexation behavior of theophylline was intensely studied for modelling metal interactions with the guanine base of nucleic acids [13]. In neutral or basic environment, theophylline acts as a monodentate ligand and coordinates through the N7 atom, which is the preferred binding site in 6-oxopurines [1, 2, 3, 12].

Theophylline (3,7-dihydro-1,3-dimethyl-1H purine-2,6-dione(1,3-dimethyl-xanthine) (fig. 1a) is used in medicine as an antiasthmatic, coronary vasodilator, and diuretic. It is a xanthine alkaloid (methylxanthine) chemically related to caffeine and theobromine and is also an active constituent of tea (*Camellia sinensis*), commercially produced in the pharmaceutical field by chemical synthesis. Theophylline contains some atoms involved in coordinative bonds with different oligoelements, a support for the synthesis of some complexes combinations and consequently, the corresponding physico-chemical and structural analyses.

This study presents the spectroscopic and microscopic investigation of newly synthesized theophylline complexes with some transitional metal ions: Cu(II), Co(II), Cd (II) and Zn(II). Their physico-chemical properties were established by Fourier transform infrared spectroscopy (FTIR), electron paramagnetic resonance spectroscopy (EPR) and scanning electron microscopy (SEM).

Materials and methods

All materials were of reagent grade and used without further purification. Anhydrous theophylline was purchased from Arena Group S.A. București. $\text{Cu}(\text{CH}_3\text{COO})_2 \cdot \text{H}_2\text{O}$, $\text{Cd}(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$, $\text{Co}(\text{CH}_3\text{COO})_2 \cdot 4\text{H}_2\text{O}$, $\text{Zn}(\text{CH}_3\text{COO})_2 \cdot \text{H}_2\text{O}$ used for synthesis were purchased from Merck Germany.

The general procedure for the synthesis of the transition metal (II) complexes was: 1.80 g theophylline (TEO) was dissolved in a warm environment in 100mL distilled water heated up to 70°C. The obtained solution was treated with a solution that contained 0.005 mols (≈ 1.0 g) $\text{Cu}(\text{CH}_3\text{COO})_2 \cdot \text{H}_2\text{O}$ dissolved in 50 mL distilled water. The reaction mixture was stirred at room temperature for 30 min. The obtained green precipitate was filtered using a Büchner funnel, was washed several times with washing water that contains, for 100mL solution, 0.2 g theophylline and 0.2g copper acetate, and then with distilled water heated up to 70°C, then with alcohol and ether. Then, the substance was dried in open air on a filter paper or under vacuum.

In the same way, the other new complexes were obtained, but instead of the copper acetate, cobalt, zinc or cadmium acetate were used [5, 6, 11, 15]. The colours of new complexes synthesised was green for copper, light rose for cobalt, white for zinc and cadmium.

FTIR spectroscopic investigation were performed with a JASCO 6100 spectrometer using the KBr pellet technique in the spectral domain of 4000-400 cm^{-1} with a resolution of 2 cm^{-1} .

EPR spectra were obtained in the X band using standard JEOL-RES-3S equipment.

The morphological examinations were performed using the scanning electronic microscope type 5600 LV Jeol equipped with X-rays spectrometer type Oxford Instrument, with the following characteristics: resolution: 3.5 nm with secondary electrons; enlargement 300,000x; local quantitative chemical analyses based upon the X-rays characteristic spectrum (EDS) for the elements listed between boron and uranium, with the detection limit of 0.01%.

Results and discussion

Theophylline is able to form coloured complexes having distinct spectral characteristics with transition metal ions: Cu (II), Co (II), Cd (II), Zn (II). The solid products are soluble in acetone, chloroform, dimethylformamide and glacial acetic acid. The elemental analysis was previously reported and demonstrated a coordination ratio of 1:2 metal: theophylline, with the presence of crystallization water molecules [11].

FTIR spectrum of theophylline is presented in figure 1 along with the corresponding molecular structure and the FTIR spectra of the metallic complexes are displayed in figure 2 (a, b, c, d).

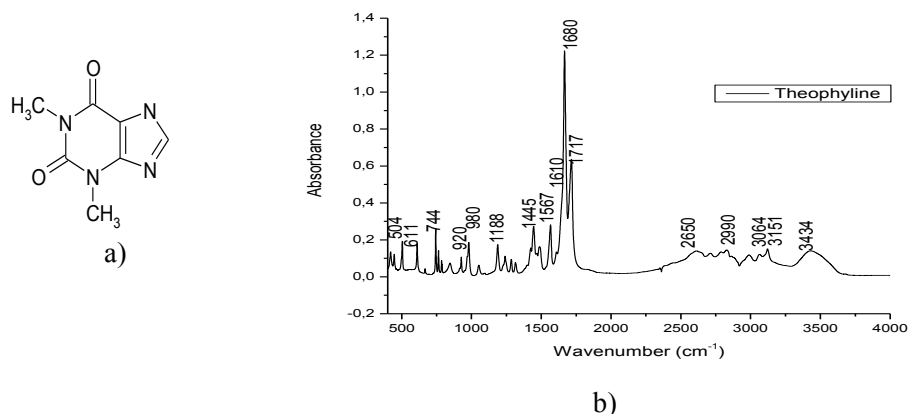
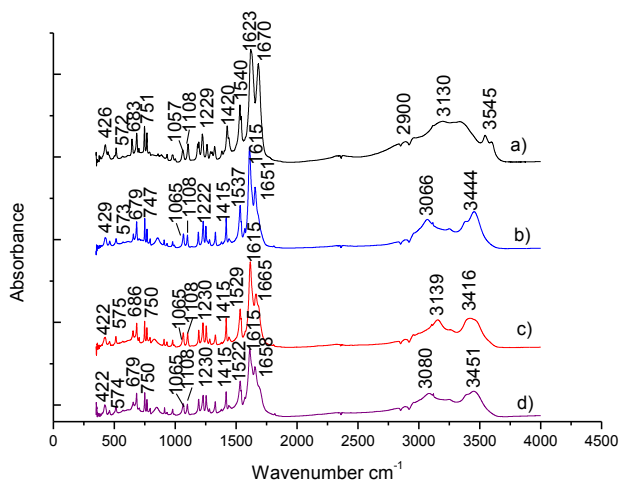


Figure 1. a) Molecular structure of theophylline, b) FTIR spectrum of theophylline

**Figure 2**

FTIR spectra of the complexes: a) $[\text{Cu}(\text{TEO})_2](\text{CH}_3\text{COO})_2$, b) $[\text{Cd}(\text{TEO})_2](\text{CH}_3\text{COO})_2$,
c) $[\text{Co}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ and d) $[\text{Zn}(\text{TEO})_2](\text{CH}_3\text{COO})_2$

The assignments of the characteristic vibrations are listed in Table I.

Table I

The assignments of vibrational frequencies for the investigated complexes

TEO (cm^{-1})	$[\text{Cu}(\text{TEO})_2]\text{Ac}_2$ (cm^{-1})	$[\text{Cd}(\text{TEO})_2]\text{Ac}_2$ (cm^{-1})	$[\text{Co}(\text{TEO})_2]\text{Ac}_2$ (cm^{-1})	$[\text{Zn}(\text{TEO})_2]\text{Ac}_2$ (cm^{-1})	Assignment (cm^{-1})
420	426(m)	429(w)	422(w)	422(w)	$\delta_{\text{C-N-C}}(\text{sim})$
-	572(w)	573(w)	575(w)	574(w)	$\nu_{\text{Me-L}}$
611	611(w)	611(w)	611(w)	611(w)	$\delta_{\text{C=C-N}}$
668	650(m)	645(m)	655(w)	653(w-m)	$\delta_{\text{O=C-N}}$
-	683(m-s)	679(m)	686(m)	679(m)	$\nu_{\text{Me-L}}$
744	751(s)	747(m)	750(m)	750(m)	$\delta_{\text{O=C-C}}$
764	769(m)	768(m)	768(m)	768(m)	γ_{ring}
786	791(w)	785(w)	794(w)	794(w)	$\gamma_{\text{C-H}}$
847	855(w)	859(w)	848(w)	828(w)	$\delta_{\text{C-N-H}}$
1241	1229(m)	1222(m)	1230(m)	1230(m)	$\nu_{\text{C-N}}(\text{asim})$
-	1251(m)	1261(m)	1254(m)	1255(m)	$\nu_{\text{COO}}(\text{-sim})$
-	1534(vs)	1537(s)	1529(s)	1522(s)	$\nu_{\text{COO}}(\text{-asim})$
1680	1670(vs)	1651(vs)	1665(vs)	1658(vs)	$\nu_{\text{C=O}}(\text{asim})$
1717	-	-	-	-	$\nu_{\text{C=O}}$
2650	-	-	-	-	$\nu_{\text{N-H}}$
2990	2900(w)	2901(w)	2902(w)	2903(w)	$\nu_{\text{N-H}}(\text{asim})$
3064	3040(m)	2997(w)	2970(w)	3001(m)	$\nu_{\text{C-H}}$
3151	3130(s)	3066(m)	3139(m)	3080(m)	$\nu_{\text{N-H}}(\text{sim})$
3434	3545(s)	3444(m)	3416(s)	3451(m)	ν_{ring}

vs: very strong, s: strong, m: medium, w: weak, v: stretching, δ : in plane bending, γ : out-of-plane bending

When interpreting the FTIR spectra, it must be taken into consideration the fact that theophylline consists of two heterocyclic systems, a six membered pyrimidine ring and a five membered imidazole ring [14]. The vibrational frequency assignment of theophylline and of the newly synthesized compounds was done using the vibrational frequencies of benzene, pyridine, pyrimidine, and imidazole as guidelines. Nitrogen heterocyclic aromatic compounds exhibit C-H stretching vibration bands at 3100-3000 cm^{-1} . Theophylline presents a C-H stretching vibration band at 3064 cm^{-1} , while the corresponding band of the complexes is low shifted at 3040 cm^{-1} , 2997 cm^{-1} , 2970 cm^{-1} and 3001 cm^{-1} respectively for Cu, Cd, Co and Zn. In plane deformation vibrations of the heterocyclic compounds are observed in the region 1110-1000 cm^{-1} , as bands of variable intensity are present in this area. Primary aliphatic amines usually absorb in the region 3550-3250 cm^{-1} in solids or liquids. For the secondary aromatic amines, a band between 3400-3300 cm^{-1} is observed. Generally, the bands assigned to the N-H stretching modes are sharper and weaker than those of O-H stretching vibrations. In the case of theophylline, the vibrational band at 3434 cm^{-1} is assigned to the N-H stretching vibration band of the xanthine ring, while the symmetric and asymmetric N-H stretching are located at 3151 and 2990 cm^{-1} respectively. The corresponding bands in complexes spectra are down shifted to 3130, 2900 cm^{-1} (Cu), 3066, 2901 cm^{-1} (Cd), 3139, 2902 cm^{-1} (Co) and 3080, 2903 cm^{-1} in Zn, but the asymmetric vibrations are drastically reduced in intensity.

The two C=O stretching vibration bands in theophylline, symmetric and asymmetric are located at 1717 and 1680 cm^{-1} respectively, presenting a very strong intensity. In the complexes spectra, the symmetric C=O stretching vibrations bands disappear, the only one remaining at 1670 cm^{-1} (Cu), 1651 cm^{-1} (Cd), 1665 cm^{-1} (Co) and 1658 cm^{-1} (Zn) being shifted to lower wavenumbers. The intensity of these bands is also reduced upon complexation.

The lower region of wavenumbers contains the bending vibrations of O=C-C, O=C-N, C=C-N and C-N-C assigned at 744, 668, 611 and 420 cm^{-1} in theophylline spectrum [7], with some few modifications in the complexes spectra. Metal-ligand coordination bonds usually occur in the FTIR spectra of the complex combinations at wavelengths ranging between 570-685 cm^{-1} . In our complexes spectra, the two coordination bonds occur around 570 cm^{-1} and 685 cm^{-1} (not in theophylline spectrum). It was also observed the disappearance of two absorption bands (1717 cm^{-1} and 2650 cm^{-1}) in the spectra of the complexes, assigned to C=O and N-H stretching vibrations. This fact may be correlated with chelates formation. Based on

the above results, we assume that N1 and N3 nitrogen atoms are being blocked due to the methyl groups attached on them and cannot participate to the coordination with the metal. In this case, only N7 and O6 atoms are able to participate to the coordination [8], the theophylline acting as an ambidentate ligand.

The complex formation is also demonstrated by the occurrence of COO^- stretching vibrations at $1260\text{--}1250\text{ cm}^{-1}$ (symmetric) and $1535\text{--}1530\text{ cm}^{-1}$ (asymmetric). The presence of some crystallization water molecules is demonstrated by the broad bands with shoulders in the range $3050\text{--}3500\text{ cm}^{-1}$.

In order to investigate the metallic ions environment, EPR spectra of powder samples were recorded at room temperature, in X band (9.4 GHz). The only sample exhibiting EPR signal in these conditions was the Cu(II)-theophylline complex (Figure 3) suggesting the presence of monomeric species with an axial symmetry. The values of the g tensor are $g_{\parallel} = 2.182$ and $g_{\perp} = 2.060$ showing a square-planar local symmetry around Cu(II) ion, with $d_{x^2-y^2}$ orbital as ground state for the paramagnetic electron [4, 10].

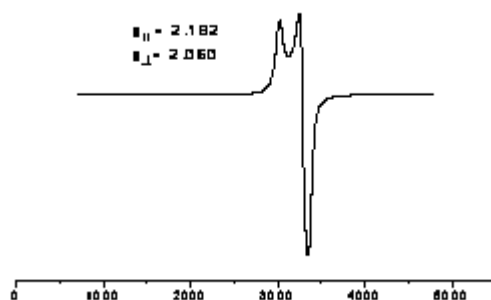
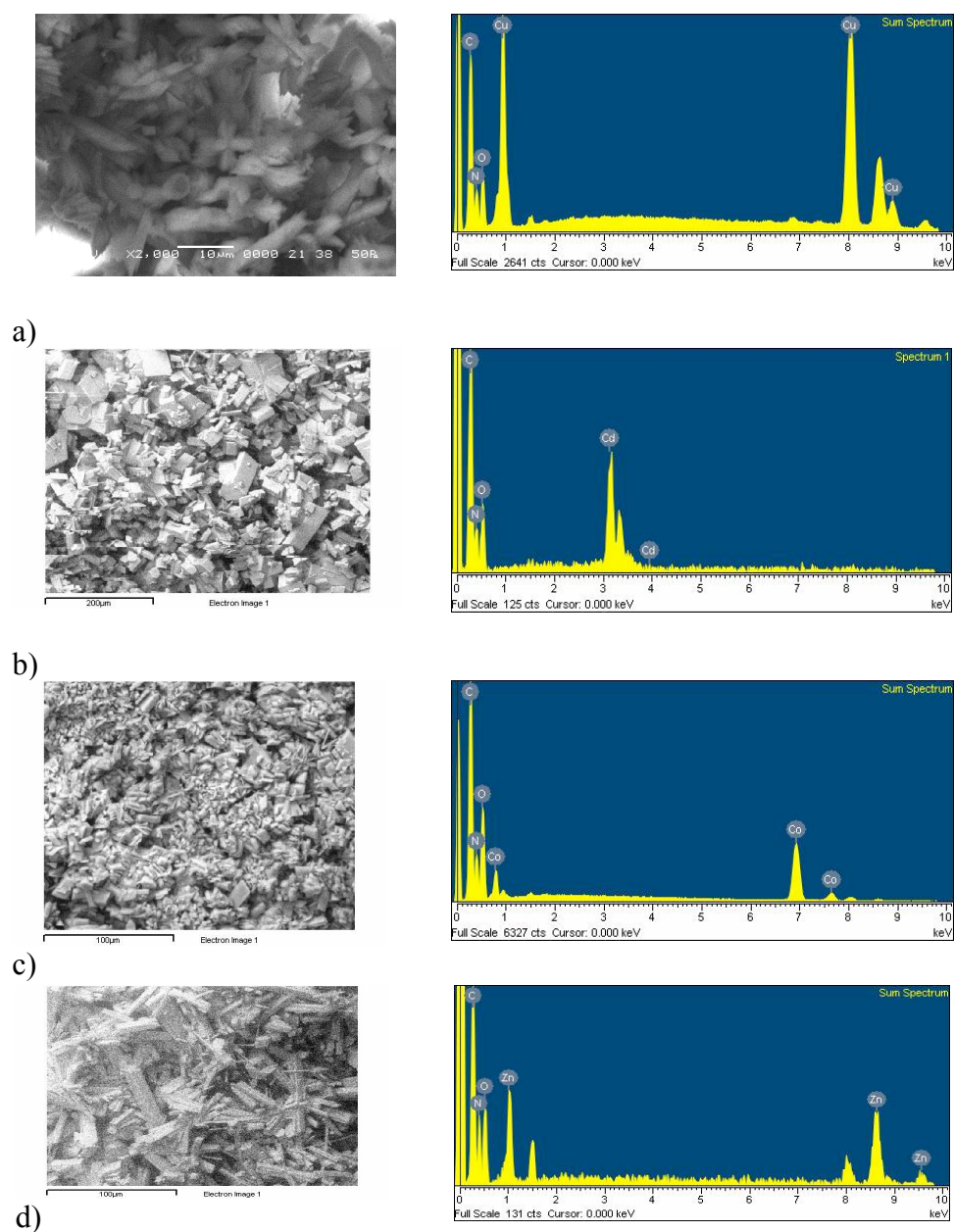


Figure 3
EPR powder spectrum of $[\text{Cu}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ complex

The morphology of the surfaces recorded by SEM reveals very different details comparing the four complexes (figure 4 a, b, c, d).

$[\text{Cu}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ presents a mixture of aciculate, flake-like, irregular and lamellar shapes, $15\text{--}20\text{ }\mu\text{m}$ long (fig. 4a).

$[\text{Cd}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ consists of microcrystals, mostly parallelepiped-shaped. The larger ones are $L = 80\text{ }\mu\text{m}$, $l = 20\text{ }\mu\text{m}$, and the smaller are $L = 10\text{ }\mu\text{m}$, $l = 3\text{ }\mu\text{m}$. The average size is about $10\text{--}30\text{ }\mu\text{m}$. Carefully analyzed, one can observe that they are monocrystals, the development and the arrangement of the atoms along the x, y and z axes being similar (fig. 4b).

**Figure 4**

The morphology of a) $[\text{Cu}(\text{TEO})_2](\text{CH}_3\text{COO})_2$, b) $[\text{Cd}(\text{TEO})_2](\text{CH}_3\text{COO})_2$, c) $[\text{Co}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ and d) $[\text{Zn}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ micro crystals surface and the corresponding EDS spectra (X rays) of the compound elements

In the $[\text{Co}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ complex the microcrystals present a tabular, parallelepiped shape, with dimensions ranging from $L = 30 \mu\text{m}$, $l = 10 \mu\text{m}$ to $L = 5 \mu\text{m}$, $l = 1 \mu\text{m}$. The average size of the microcrystals is about $20 \mu\text{m}$ (fig. 4c).

$[\text{Zn}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ microcrystals also present a tabular parallelepiped shape. Their dimensions vary from $L = 100 \mu\text{m}$, $l = 10 \mu\text{m}$ to $L = 25 \mu\text{m}$, $l = 5 \mu\text{m}$, with the average size of $70 - 80 \mu\text{m}$ (fig. 4d).

Conclusions

FTIR analysis confirms the existence of the complex combinations by the presence of the characteristic band of metal-theophylline in the $700\text{-}500 \text{ cm}^{-1}$ spectral region and the disappearance of the vibrations due to the atoms implied in the coordinative bonds. Consequently, the metal is coordinatively coupled to the nitrogen atom no. 7 and to the oxygen atom no. 6, N1 and N3 being blocked by the methyl groups engrafted on them [7, 8]. The spectral FTIR features demonstrated that each complex includes crystallization water molecules. Powder EPR spectrum of $[\text{Cu}(\text{TEO})_2](\text{CH}_3\text{COO})_2$ at room temperature suggests an axial local symmetry around the copper ion in a CuN_2O_2 arrangement and the $d_{x^2-y^2}$ orbital as ground state for the paramagnetic electron. Scanning electron microscopy demonstrated a similar morphology of Co, Cd and Zn complexes with tubular, parallelepiped shaped micro crystals, whereas the Cu complex presents a mixture of needle forms, irregular and lamellar flakes shapes.

References

1. Begum N.S., Manohar H.- Synthesis and X-ray crystal structure of a Cu^{II} -theophylline complex: $[\text{Cu}(\text{theo})_2 (\text{H}_2\text{O})_3] \cdot 2\text{H}_2\text{O}$ - *Polyhedron* 1994, 13, 307
2. Bombicz P., Madarasz J., Forizs E., Foch I.- Structure and thermal behaviour of (SPY-5-12)-(2-aminoethanol-N) (2-aminoethanol-N,O)-bis(theophyllinato) copper (II) dihydrate: a model for DNA-metal interactions- *Polyhedron* 1997, 16, 3601
3. Buncel E., Kumar R., Norris A.R., Beauchamp A.L.- Metal ion biomolecule interactions. PART VII. Synthesis, spectroscopic properties and crystal structure investigations of some bis (theophyllinato) tetraaquocadmium (II) compounds- *Can. J. Chem.* 1985, 63, 2575
4. David L., Cozar O., Craciun C., Chis V., Rezonanța electronică de spin, Presa Universitară Clujeană, Cluj Napoca, 2001
5. *** Europäisches Arzneibuch, Nachtrag, Berlin, 2000, 1430
6. *** Farmacopeea Română, Editia a X-a, Editura Medicală, București, 1993, 913
7. Gunasekaran S., Sankari G., Ponnusamy S.- A comparative study of caffeine and theophylline binding to $\text{Mg}(\text{II})$ and $\text{Ca}(\text{II})$ ions: studied by FTIR and UV spectroscopic methods, *J. Mol. Struct.* 608, Issue 1(202), 1-7

8. Gabriela Rău, George-Dan Mogoșanu, Camelia Elena Stănciulescu, Synthesis and characterization of some novel azoderivatives, *Farmacia* 2009, 57, 5, 648-658
9. Jovanovski G., Naumov P., Grupce O., Kaitner B.- Structural study of monoaquabis(pyridine)bis(saccharinato)copper(II), $[\text{Cu}(\text{H}_2\text{O})(\text{py})_2(\text{sac})_2]$, *Eur.J. Solid State Inorg. Chem.* 35, 1998, 231
10. Marcu A., Stanila A., Rusu D., Rusu M., Cozar O., David L.-Spectroscopic studies of copper (II) complexes with some amino acids- *J. Optoelectr. Adv. Mater.*, 2007, 9, 741
11. Marian E., Jurca T., Kacso I., Borodi Ghe., Bratu I.- Structure investigations of some complexes of theophylline with transitional metals- *Rev. de Chimie.* 2009, 60, 6, 599
12. Tünde Jurca, Eleonora Marian, Scanning electronic microscopy study of new pyrazinamide compounds with metallic ions, *Farmacia*, 2009, 57, 2, 247- 253
13. Orbell J. D., Taylor M.R., Birch S.L., Lowton S.E., Vilkins L.M., Keefe L.J., The crystal structures of four models for the binding to DNA of cisplatin derivatives containing a bidentate tertiary diamine, *Inorg. Chim. Acta* 1988, 152, 125
14. Socrates G., Infrared Characteristic Group Frequencies, Wiley, New York, 1980
15. *** USP 23, Twinbrook Parkway, Rockville, 1995, 1521

Manuscript received: June 10th 2009