Synthesis, Characterization and Fluorescent Properties of New Complexes of 5-hydroxyflavone with Some Divalent Metal Ions

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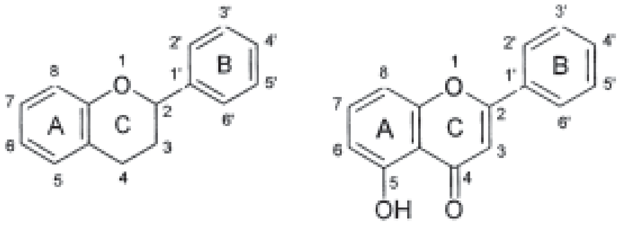
New complexes of Mg(II), Zn(II), Cu(II) and Co(II) (1-4) with the natural flavonoid 5-hydroxyflavone have been synthesized. The characterization of the newly formed compounds was done by UV-Vis, IR, fluorescent spectroscopy, elemental analysis and molar electric conductivity. The complexes of Mg(II) and Zn(II) are highly fluorescent.

Keywords: complexes, 5-hydroxyflavone, divalent metal ions, fluorescence

Flavonoids (2-phenyl-benzo-γ-pyrones, fig. 1a) represent a large group of polyphenolic natural compounds widespread in seeds, fruit skin, peel, and bark of plants [1]. Their valuable cardioprotective [2], antioxidant [3, 4], anticancer [5-7], anti-inflammatory [8, 9], antiviral [10], anti-allergic [11], anti-osteoporotic [12] or antiulcer [13] properties attracted a major interest for studying the chemistry of the favonoid molecules.

Flavonoids are good chelating ligands for various metal ions, and many of their chelates were investigated for the antioxidant [14-16], antitumor [17-19], antimicrobial and antiinflammatory [20] activities.

Among the flavonoids, 5-hydroxyflavone (5-hydroxy-2-phenyl-4*H*-1-benzopyran-4-one, primuletin, fig. 1b) is a bio-derivative with anti-inflammatory [21], antiatherogenic [22], vasorelaxing [23] effects, and is a well-known androgen receptor antagonist used to treat prostate cancer [24].



a b

Fig. 1. (a) General structure of flavonoids. (b) Chemical structure of 5-hydroxyflavone

Due to the presence of a chelating 5-hydroxy-4-keto group, 5-hydroxyflavone can act as a bidentate ligand toward metal ions. Considering the low solubility in water of 5-hydroxyflavone, on the one hand, and its good solubility in alcohol on the other hand, most part of solution complexation studies were carried out in neat methanol or in methanol-water mixtures. In pure methanol, complexes of 1:1 stoichiometry were obtained following the interaction of 5-hydroxyflavone with Al(III) [25], Pb(II) [26] and Zn(II) [27].

The aim of this study was to obtain and characterize in solid state new complexes of 5-hydroxyflavone with some divalent metal ions, namely Mg(II), Zn(II), Co(II) and Cu(II). Given the photophysical properties of 5-hydroxyflavone [28, 29], the fluorescence properties of the complexes have been also investigated.

Experimental part

Materials

All reagents and solvents were of analytical reagent grade and were used withou tfurther purification. 5-hydroxyflavone was purchased from Aldrich Chemical Co., Schnelldorf, Germany, $MgSO_4{\cdot}7H_2O$, $ZnSO_4{\cdot}7H_2O$, $CuSO_4{\cdot}5H_2O$, $CoCl_2{\cdot}6H_2O$, and KOH were obtained from Merck, Germany.

Synthesis of the complexes

Synthesis of complexes was carried out following the general procedure: a methanolic solution (10 mL) of metal salt (0.2 mmol, 0.0493 g $MgSO_4 \cdot 7H_2O$; or 0.0575 g $ZnSO_4 \cdot 7H_2O$; 0.0499 g $CuSO_4 \cdot 5H_2O$; 0.0476 $CoCl_2 \cdot 6H_2O$ respectively) was added to a methanolic solution (15 mL) of 5-hydroxyflavone (0.4 mmol, 0.0953 g), deprotonated with KOH (0.4 mmol, 22 mg). The reaction mixture was refluxed for 1 h. The solid product formed was filtered off, washed several times with small amounts of methanol, and dried in air.

Instrumentation –physical measurements

The C, H, S-microanalyses were performed by a Perkin–Elmer 2400 Series II CHNS analyzer. Melting points were determined using a digital capillary melting point apparatus, Optimelt- Stanford Research System. Electrical conductivity measurements of the complexes were recorded at 20°C for 10^{-3}M solution of the sample in DMSO using a Consort C830 (Turnhout, Belgium) conductimeter with an SK10T platinum electrode embedded in glass (cell constant 1.0 cm^{-1}). IR spectra were recorded using KBr pellets on a FT-IR VERTEX 70 (Bruker) spectrometer in the range 400–4000 cm^{-1}. Electronic spectra by diffuse reflectance technique, with Spectralon as reference sample, were recorded in the range 200–900 nm, on a Jasco V 650 spectrophotometer. Fluorescence spectra were recorded on a Jasco FP 6500 spectrofluorometer.

Results and discussions

Synthesis and analytical data

The complexes were prepared by a simple reaction involving depronated ligand and a salt of a divalent metal

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Table 1
ANALYTICAL DATA AND MOLAR CONDUCTANCE VALUES FOR METAL COMPLEXES

Compound / Molecular formula	Molecular weight (g mol^{-1})	Anal. found (calcd.) (%)			Colour	Melting point (°C)	Molar conductance Λ_M (Ω^{-1} cm^2 mol^{-1})
		C	H	S			
$[Mg_2L_2(SO_4)(H_2O)_6] \cdot 2H_2O$ $Mg_2C_{30}H_{34}O_{18}S$ **(1)**	762.54	46.98 47.24	3.93 4.23	4.09 4.20	yellow-orange	>400	7.1
$[Zn_2L_2(SO_4)(H_2O)_6] \cdot 2H_2O$ $Zn_2C_{30}H_{34}O_{18}S$ **(2)**	845.32	42.25 42.61	3.99 4.06	3.52 3.79	yellow-orange	247	3.2
$[Cu_2L_2(SO_4)]$ $Cu_2C_{30}H_{18}O_{10}S$ **(3)**	697.64	51.27 51.64	2.90 2.60	4.56 4.89	yellow-olive	>400	6.7
$[CoLCl(H_2O)] \cdot 0{,}5\ H_2O$ $CoC_{15}H_{12}ClO_{4.5}$ **(4)**	358.64	50.22 50.23	2.91 3.37	-	yellow	220-222	34.5

Table 2
IR DATA (cm^{-1}) FOR LIGAND AND COMPLEXES

Compound	ν(O-H)	ν(C=O)	ν(C=C)	ν(C-O) + δ(OH)	ν(C-O-C)
HL	3200-2600 b, m	1654 s; 1615 s	1587 s	1357 m; 1298 s	1255 s
(1)	3600-2600 b, m	1639 s	1582 s	1361 m; 1299 w	1252 s
(2)	3600-2600 b, m	1633 s	1581 s	1357 m; 1299 w	1253 s
(3)	-	1626 s	1578 s	1360 m; 1296 w	1256 s
(4)	3600-2600 b, m	1632 s	1581 s	1352 m; 1297 w	1251 s

b: broad; m: medium; s: strong; w: weak.

ion (sulphate in the case of Mg(II), Zn(II), Cu(II), and cobalt (II) chloride, respectively). Although a metal-to-ligand molar ratio of 1:2 was used, elemental analysis revealed a metal:ligand:gegenion ratio of 2:2:1 for complexes (**1**), (**2**), and (**3**), and a metal:ligand ratio of 1:1 for complex (**4**). The analytical data of the complexes correspond to the compositions given in table 1. The formulae proposed for these compounds were established on the basis of elemental chemical analysis, correlated with physicochemical investigations (FT-IR and UV-Vis spectroscopy and molar conductance values). The coloured compounds obtained are all hardly soluble in water, soluble in DMSO, except for copper (II) complex which dissolved moderately in DMSO. The higher melting points of the complexes than those of the ligand (158-161°C) can be taken as an evidence for the bonding to the metal ion with chelate ring formation. Chelates of divalent metal ions obtained appeared to be non-electrolytes according to the molar conductance values ranging between 3.2-34.5 Ω^{-1} cm^2 mol^{-1} [30, 31].

Infrared spectra

IR spectra of the free ligand and its complexes (fig. 2) are useful for determining the mode of coordination of the ligand. The most important vibration bands along with their assignments are given in table 2.

In the high wavenumber region of the IR spectrum of 5-hydroxyflavone (HL), the intense broad band observed between 2600 and 3200 cm^{-1} was assigned to the strong intramolecular hydrogen bond involving the OH group, a characteristic feature of 5-hydroxylated chromones [32]. In this region, a sharp and intense band at 3059 cm^{-1} is also present corresponding to the stretching vibration n(C-H). In the IR spectra of the complexes (**1**), (**2**), and (**4**) a broad band is found between 2600 and 3600 cm^{-1}, assigned to the presence of water molecule in the structure of these complexes. The absence of this band in the IR spectrum of complex (**3**) sustains its anhydrous nature.

The ν(C=O) vibration of 5-hydroxyflavone generates two intense bands, placed at 1654 and 1615 cm^{-1}. In the IR spectra of the complexes, a single strong band appears at around 1633 cm^{-1}; the shifting of ~20 cm^{-1} suggests the involvement of C=O group in coordination. The strong band which appears at 1587 cm^{-1} in the IR spectrum of the ligand, characteristic for n(C=C), is slightly shifted in the IR spectra of complexes.

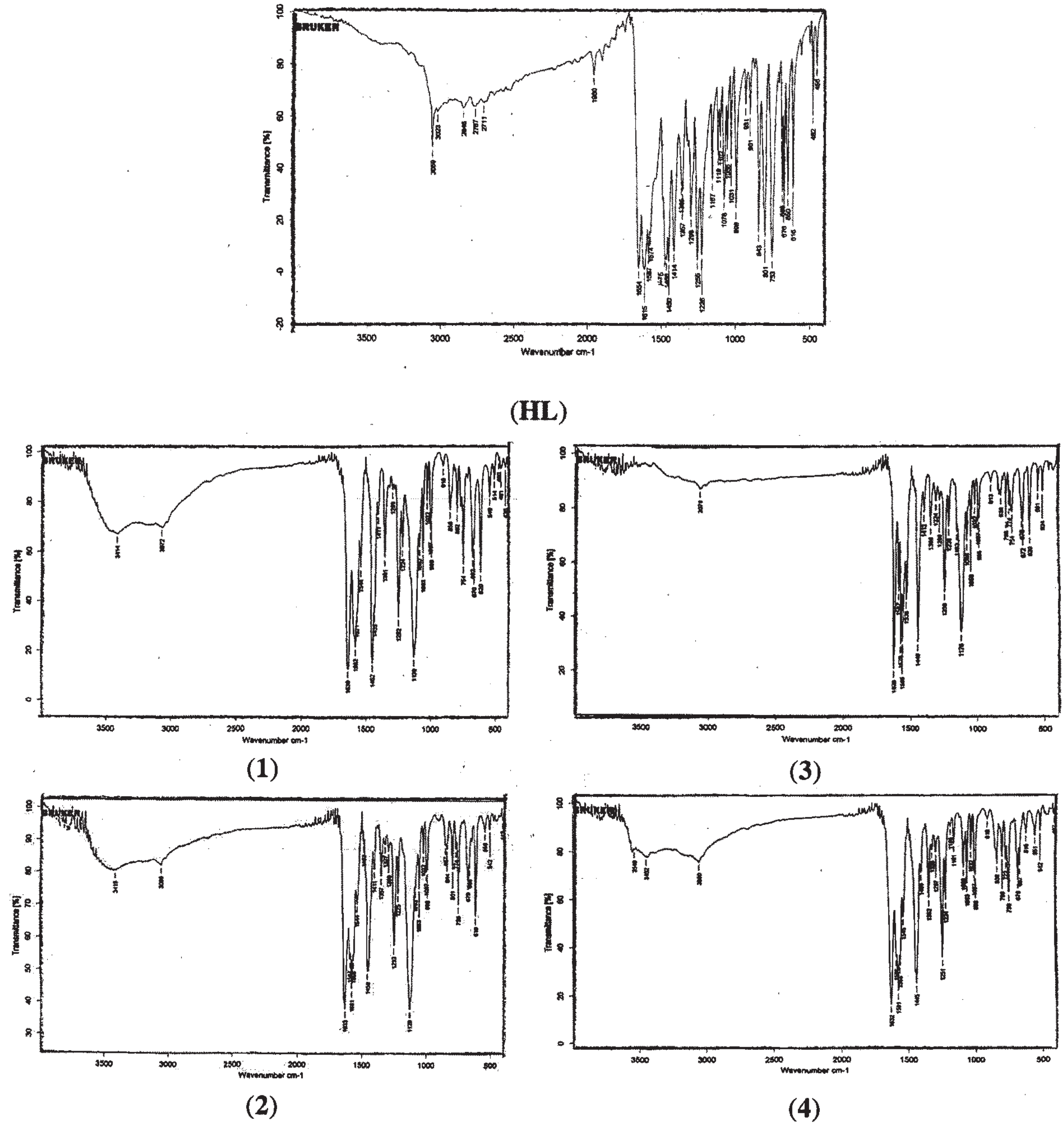
In the IR spectrum of 5-hydroxyflavone, between 1000 and 1500 cm^{-1}, bands associated with d(OH) mode are presented, mixed with ν(C=O), ν(C=C) and aromatic ring deformation. The strong band at 1298 cm^{-1} corresponding to the coupled vibration ν(C-O) + δ(OH) [33] in the IR spectrum of the ligand, appears much weakened in the IR spectra of complexes. This behavior suggests the coordination of the ligand in its deprotonated form. The ν(C-O-C) frequency is unaffected upon complexation, indicating that the ring oxygen is not involved in coordination process.

In the sulfato complexes (**1**), (**2**), and (**3**), the ν_3 vibration is observed at ~ 1129 cm^{-1} as a very intense and enlarged band, while the ν_4 vibration is observed near 620 cm^{-1} as a strong band. The positions of these bands better match with the coordination of sulphato anion as bidentate bridging ligand [34]. However, not all the characteristic bands can be observed, because of the overlap with those of the ligand.

Electronic spectra

The electronic spectra recorded for ligand and its complexes are presented in figure 3, and the data obtained are summarized in table 2.

The diffuse reflectance electronic spectrum of 5-hydroxyflavone exhibits two major absorption maxima, typically observed for the flavonoid structure. Both bands originate from π–π* transitions: the band centred at 396 nm (band I) is due to the transitions within the B ring, a cinnamoyl system, whereas the band at 280 nm (band II) may be assigned to the transitions in the ring A, a benzoyl system [35]. Both bands are bathochromically shifted in the electronic spectra of complexes, due to the extension of the conjugated system with the complexation process (fig. 4). Especially the band I is broadened, as a result of overlapping with LMCT band after coordination, and is much shifted to higher wavelengths.



(HL)

(1)

(3)

(2)

(4)

Fig. 2. IR spectra of 5-hydroxyflavone (HL) and its complexes with Mg(II) (**1**), Zn(II) (**2**), Cu(II) (**3**) and Co(II) (**4**)

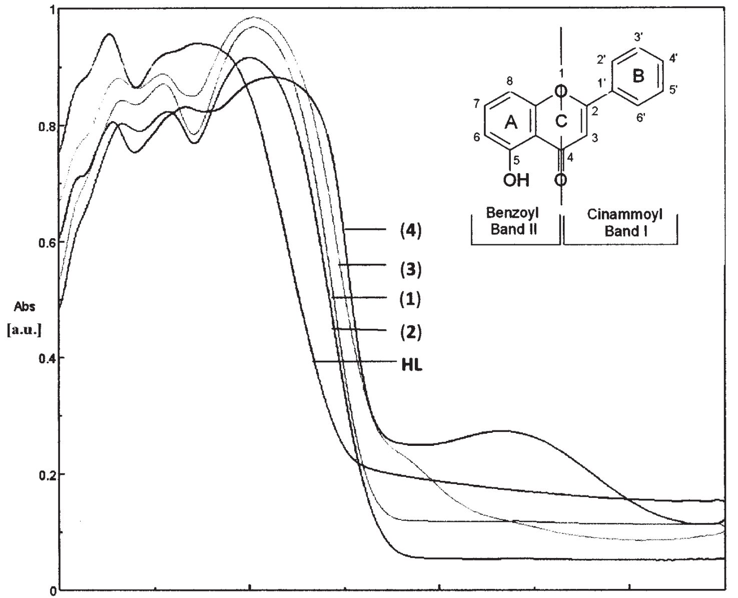


Fig. 3. Diffuse reflectance electronic spectra of 5-hydroxyflavone (HL) and its complexes with Mg(II) (**1**), Zn(II) (**2**), Cu(II) (**3**) and Co(II) (**4**)

Table 3
UV-VIS DATA FOR LIGAND AND COMPLEXES

Compound	λ_{max}(nm)		
	d–d	Band I	Band II
HL	-	345	253
(1)	-	406	267
(2)	-	401	267
(3)	667	424	258
(4)	680 (sh) 574	456	263

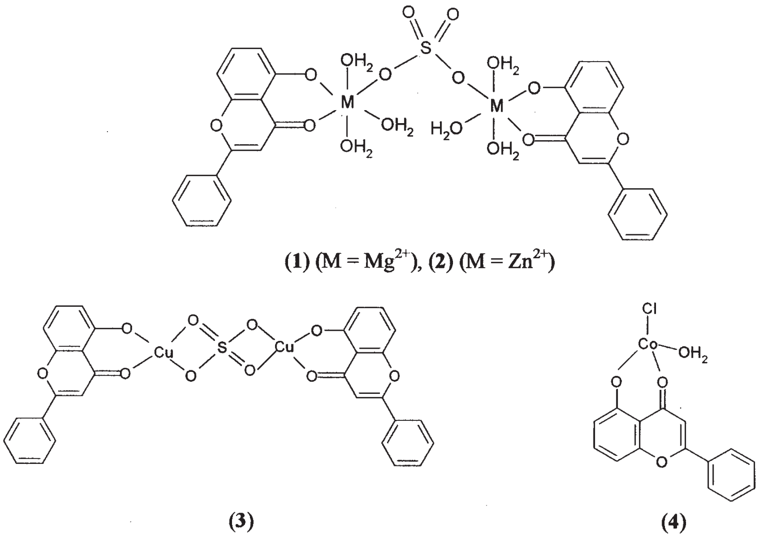


Fig. 4. Proposed structures for the complexes of 5-hydroxyflavone with Mg(II) (**1**), Zn(II) (**2**), Cu(II) (**3**) and Co(II) (**4**)

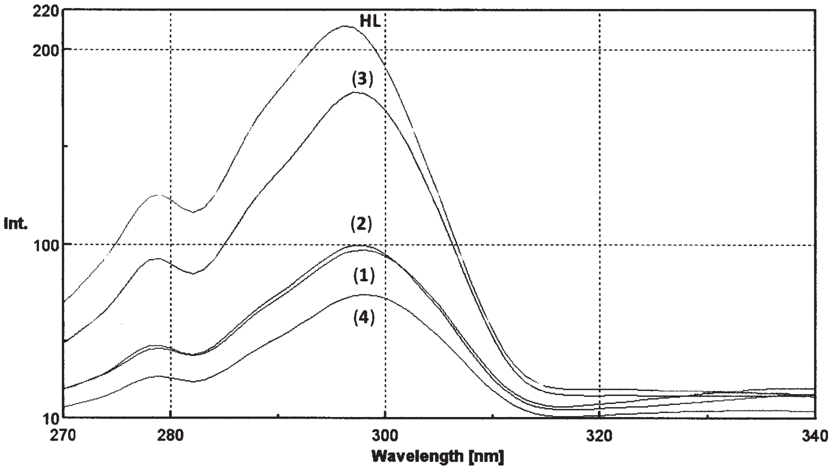


Fig. 5. The emission spectra of 5-hydroxyflavone and the complexes at λ_{exc} = 253 nm

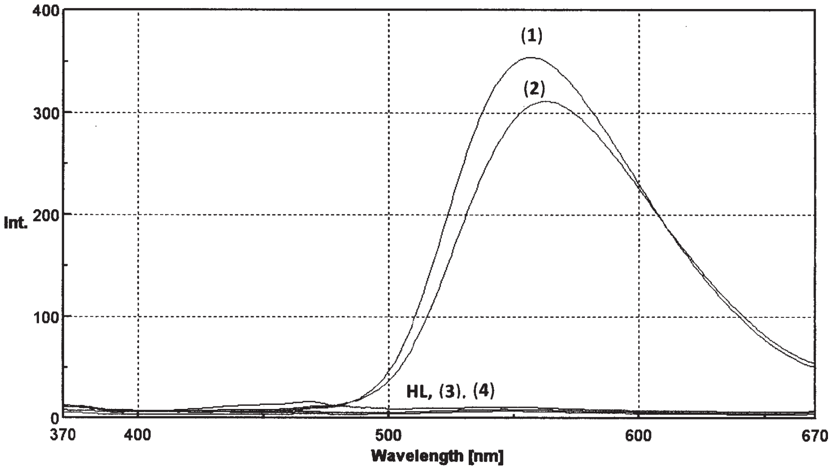


Fig.6. The emission spectra of 5-hydroxyflavone and the complexes at λ_{exc} = 345 nm

The characteristic features in the visible region of the electronic spectra of complexes (**3**) and (**4**) allow appraisals about coordination environment of metal ion. The spectrum of the copper (II) complex (**3**) exhibits a broad and low energy band at 667 nm which is attributed to d-d transition ($^2B_{1g} \rightarrow {}^2A_{1g}$) typical for Cu(II) in a square planar environment [36]. In the low energy region, the spectrum of cobalt (II) complex (**4**) exhibits a shoulder at 680 nm and a low intensity band centered at 574 nm. These bands may be assigned to the ${}^4A_2 \rightarrow {}^4T_1(F)$ (ν_2) and ${}^4A_2 \rightarrow {}^4T_1(P)$ (ν_3) transitions, respectively, supporting the tetrahedral geometry [37].

Considering the analytical and spectral data, the most probable structures of the obtained complexes are presented in figure 4.

Fluorescent properties

The fluorescence emission spectra were recorded at four excitation wavelengths, 253 nm (fig. 5), 345 nm (fig. 6), 400 nm (fig. 7) and 429 nm (fig. 8), respectively, both for ligand and complexes. The data presented in table 4 revealed the following: (i) 5-hydroxyflavone exhibits a strong fluorescence when it was excited at 253 and 429 nm, respectively; (ii) all complexes displayed a quenching of fluorescence at λ_{exc} = 253 nm (λ_{exc} = 298 nm) and at

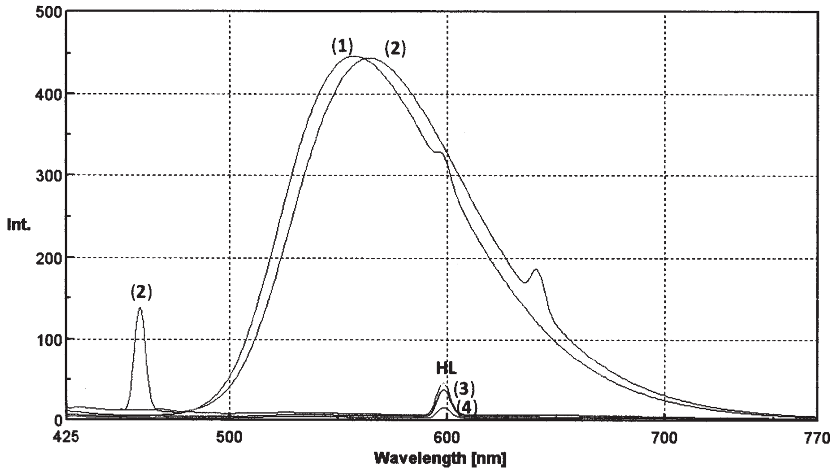


Fig.7. The emission spectra of 5-hydroxyflavone and the complexes at λ_{exc} = 400 nm

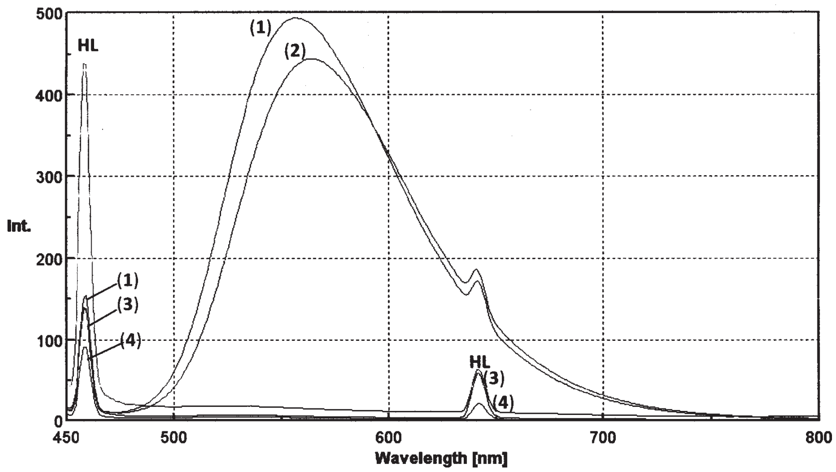


Fig. 8. The emission spectra of 5-hydroxyflavone and the complexes at λ_{exc} = 429 nm

Table 4
FLUORESCENCE DATA FOR LIGAND AND COMPLEXES

Compound	λ_{exc} = 253 nm		λ_{exc} = 345 nm		λ_{exc} = 400 nm		λ_{exc} = 429 nm	
	λ_{em} (nm)	I (a.u.)	λ_{em} (nm)	I (a.u.)	λ_{em} (nm)	I (a.u.)	λ_{em} (nm)	I (a.u.)
HL	298	212	468	17	599	47	459 642	438 64
(1)	298	97	556	354	599	448	459 557 642	154 494 172
(2)	298	99	562	311	599	438	459 564 642	138 444 186
(3)	298	178	468	7	599	39	459 642	139 59
(4)	298	74	551	7.5	599	17	459 642	91 22

λ_{exc} = excitation wavelength; λ_{em} = emission wavelength; I = relative fluorescence intensity

λ_{exc} = 429 nm (λ_{exc} = 459 nm); (iii) the Mg(II) and Zn(II) complexes had a very strong fluorescence for three of the excitation wavelength used (345, 400, 429 nm).

The differences observed for Mg(II) and Zn(II) complexes could be correlated not only with their structure, but also with the saturated structure of the metal ion.

Conclusions

Primuletin (5-hydroxyflavone) is a flavonoid derivative with important biological applications. It could form chelates with metal ions acting as bidentate ligand. In this work, the synthesis and properties of the complexes with Mg(II), Zn(II), Cu(II) and Co(II) were investigated. The new complexes have been characterized by elemental analysis, melting point determination, molar conductivity measurements, spectral data (IR, UV-Vis). Considering the analytical and spectral data, as well as the molar conductance values, we concluded that the complexes with Mg(II), Zn(II) and Cu(II) are dimers with a metal:ligand:gegenion ratio of 2:2:1, whereas the compound with Co(II) is a mononuclear complex. The Mg(II) and Zn(II) complexes exhibit a strong fluorescence, valuable for analytical applications.

References

1.ANDERSEN, O.M., MARKHAM, K.R., Flavonoids – chemistry, biochemistry and applications. CRC Taylor & Francis, Boca Raton (2006)

2.MIDDLETON, E., JR., KANDASWAMI, C., THEOHARIDES T.C., Pharmacol. Rev., 52, nr.4, 2000, p. 673.

3.AGATI, G., AZZARELLO, E., POLLASTRI, S., TATTIN, M., Plant Sci., 196, 2012, p.67.

4.LUNGU, L., POPA, C.-V., SAVOIU, M., DANET, A F., DINOIU, V., Rev. Chim. (Bucharest), **61**, nr. 10, 2010, p.911.

5.KANDASWAMI, C., LEE, L.T., LEE, P.P., KE, F.C., HUANG, Y.T., LEE, M.T., In vivo, 19 nr. 5, 2005, p.895.
6.CHAHAR, M.P., SHARMA, N., DOBHAL, M.P., JOSHI, Y.C., Pharmacogn. Rev., 5, nr. 9, 2011, p1.
7.LIU, H.L., JIANG, W.B., XIE, M.X., Recent Pat. Anticancer Drug Discov., 5, nr. 2, 2010, p.152.
8.RATHEE, P., CHAUDHARY, H., RATHEE, S., RATHEE, D., KUMAR, V., KOHLI, K., Inflamm. Allergy Drug Targ., 8, nr. 3, 2009, p.229.
9.SERAFINI, M., PELUSO, I., RAGUZZINI, A., Proc. Nutr. Soc., 69, nr. 3, 2010, p.273
10.SITHISAM, P., MICHAELIS, M., SCHUBERT-ZSILAVECZ, M., CINATL, J., JR., Antiviral Res. 97, nr. 1, 2013, p.41.
11.KAWAI, M., HIRANO, T., HIGA, S., ARIMITSU, J., MARUTA, M., KUWAHAR, Y., OKHAWARA, T., HAGIHARA, K., YAMADORI, T., SHIMA, Y., OGATA, A., KAWASE, I., TANAKA, T., Allergol. Int., 56, nr. 2, 2007, p.113.
12.MA, X.-Q., ZHENG, C.-J., ZHANG, Y., HU, C.-L., LIN, B., FU, X.-Y., HAN, L.-Y., XU, L.-S., RAHMAN, K., QIN, L.-P., Phytochem. Lett., 6, nr. 1, 2013, p.118.
13.MOTA, K.S.L., DIAS, G.E.N., PINTO, M.E.F., LUIZ-FERREIRA, Â., MONTEIRO SOUZA-BRITO, A.R., HIRUMA LIMA, C.A., BARBOSA FILHO, J.M., BATISTA, L.M., Molecules, 14, nr. 3, 2009, p.979.
14.DE SOUZA, R.F., DE GIOVANI, W.F., Redox. Rep., 9, nr. 2, 2004, p.97.
15.BIRJEES, BUKHARI, S., MEMON, S., MAHROOF-TAHIR, M., BHANGER, M.I., Spectrochim. Acta A Mol. Biomol. Spectroscop., 71, nr. 5, 2009, p.1901.
16.DIAS, K., NIKOLAOU, S., Nat. Prod. Commun., 6, nr. 11, 2011, p.1673.
17.ETCHEVERRY, S.B., FERRER, E.G., NASO, L., RIVADENEIRA, J., SALINAS, V., WILLIAMS, P.A., J. Biol. Inorg. Chem., 13, nr. 3, 2008, p.435.
18.FERRER, E.G., SALINAS, M.V., CORREA, M.J., NASO, L, BARRIO, D.A,, ETCHEVERRY, S.B., LEZAMA, L., ROJO, T., WILLIAMS, P.A., J. Biol. Inorg. Chem., 11, nr. 6, 2006, p.791
19.ZHOU, J., WANG, L.F., WANG, J.Y., TANG, N., J Inorg Biochem., 83, nr. 1, 2001, p.41.
20.PEREIRA, R.M., ANDRADES, N.E., PAULINO, N., SAWAYA, A.C., EBERLIN, M.N., MARCUCCI, M.C., FAVERO, G.M., NOVAK, E.M., BYDLOWSKI, S.P., Molecules, 12, nr. 7, 2007, p.1352.
21.SHIN, S.Y., WOO, Y., HYUN, J., YONG, Y., KOH, D., LEE, Y.H., LIM, Y., Bioorg. Med. Chem. Lett., 15, nr. 20, 2011, p.6036.
22.OLSZANECKI, R., GEBSKA, A., KOZLOVSKI, V.I., GRYGLEWSKI, R.J., J. Physiol. Pharmacol., 53, nr. 4, 2002, p.571.
23.CALDERONE, V., CHERICONI, S., MARTINELLI, C., TESTAI, L., NARDI, A., MORELLI, I., BRESCHI, M.C., MARTINOTTI, E., Naunyn Schmiedebergs Arch. Pharmacol., 370, nr. 4, 2004, p.290.
24.NISHIZAKI, Y., ISHIMOTO, Y., HOTTA, Y., HOSODA, A., YOSHIKAWA, H., AKAMATSU, M., TAMURA, H., Bioorg. Med. Chem. Lett., 19, nr. 6, 2009, p.4706.
25.CORNARD, J.P., MERLIN, J.C., J. Mol. Struct., 569, nr. 1-3, 2001, p.129.
26.DANGLETERRE, L., CORNARD, J.P., Polyhedron, 24, nr. 12, 2005, p.1593.
27. LAPOUGE, C., DANGLETERRE, L., CORNARD, J.P., J. Phys. Chem. A, 110, nr. 45, 2006, p.12494.
28.NORIKANE, Y., ITOH, H., ARAI, T., J. Photochem. Photobiol. A: Chem. 161, nr, 2-3, 2004, p.163.
29.DEL VALLE, J.C., J. Chem. Phys., 124, nr. 10, 2006, p.104506.
30.GEARY, W. J., Coord. Chem. Rev., 7, 1971, p.81.
31.TABASSUM, S., NISHAT, N., ARJMAND, F., LUTFULLAH, SIDDIQI, K.S., Transition Met. Chem., 21, nr, 2, 1996, p. 97.
32.MACHADO, N.F.L., BATISTA DE CARVALHO, L.A.E., OTERO, J.C., MARQUES, M.P.M., Spectrochim. Acta A, 109, 2013, p.116.
33.KEMP, W., Organic Spectroscopy, 3rd ed., McMillan: London, UK, 1991, p. 66.
34.NAKAMOTO, K., Infrared and Raman spectra of Inorganic Coordination Compounds, 4th ed.; John Wiley and Sons: New York, NY, USA, 2006, p. 248–251.
35.MALESEV, D., KUNTIC, V., J. Serb. Chem. Soc., 72, nr. 10, 2007, p. 921.
36.LEVER, A. B. P. Inorganic Electronic Spectroscopy, Elsevier, Publishing Co., New York, 1968, p. 333.
37.GABER, M., HASSANEIN, A.M., LOTFALLA, A.A., J.Mol. Struct., 875, nr. 1-3, 2008, p. 322

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