

THE NEW RUTHENIUM(II)-BIPYRIDYL COMPLEX WITH *O,O'*-DIETHYL-(*S,S*)-ETHYLENEDIAMINE-*N,N'*-DI-2-(3-CYCLOHEXYL)PROPANOATE: SYNTHESIS AND CHARACTERIZATION

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The new bipyridyl complex of ruthenium(II) with *O,O'*-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate was synthesized. The reaction of *cis*-[RuCl₂(bpy)₂] and its ligand was performed in water/ethanol solution, in the presence of lithium hydroxide, under reflux. After the addition of ammonium hexafluorophosphate, the complex was precipitated. The complex, *cis*-[Ru(bpy)₂L](PF₆)₂, was characterized by ¹H and ¹³C NMR, UV-Vis, IR spectroscopy, ESI-MS spectrometry and elemental analysis. Results indicate an octahedral geometry of the complex, with *N,N'*-coordinated *O,O'*-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate. Complexes of this type are particularly important in terms of potential cytotoxicity and application in photodynamic therapy. Using this therapy, many side effects can be reduced, which may allow the administration of higher drug dosages.

Keywords: ruthenium(II) complexes; edda-type ligand; photodynamic therapy

НОВ КОМПЛЕКС НА РУТЕНИУМ(II)-БИПИРИДИЛ СО *O,O'*-ДИЕТИЛ-(*S,S*)-ЕТИЛЕНДИАМИН-*N,N'*-ДИ-2-(3-ЦИКЛОХЕКСИЛ)ПРОПАНОАТ: СИНТЕЗА И КАРАКТЕРИЗАЦИЈА

Синтетизиран е нов комплекс на рутениум(II) со *O,O'*-диетил-(*S,S*)-етилендиамин-*N,N'*-ди-2-(3-циклохексил)пропаноат. Реакцијата на *cis*-[RuCl₂(bpy)₂] и лигандот е изведена при рефлуksирање на раствор вода/етанол во присуство на литиумхидроксид. Комплексот се таложи по додавање на амониумхексафлуорофосфат. Комплексот, *cis*-[Ru(bpy)₂L](PF₆)₂, е карактеризиран по пат на ¹H и ¹³C NMR, UV-Vis, IR-спектроскопија, ESI-MS-спектрометрија и елементарна анализа. Резултатите укажуваат дека комплексот има октагонална геометрија со *N,N'*-координиран *O,O'*-diethyl-(*S,S*)-етилендиамин-*N,N'*-ди-2-(3-циклохексил)пропаноат. Комплексите од овој тип се особено важни во смисла на нивната потенцијална цитотоксичност и примена за фотодинамична терапија. Со примена на оваа терапија се намалуваат многу споредни ефекти, што овозможува давање на повисоки дози на лекот.

Клучни зборови: комплекси на рутениум(II); лиганд од типот на edda; фотодинамична терапија

1. INTRODUCTION

Bioinorganic chemistry is a field of inorganic chemistry which is constantly evolving, particularly in recent decades, as a growing number of synthesized compounds are widely used for medical and pharmaceutical purposes [1]. Since Rosenberg's discovery of *cisplatin*, thousands of platinum complexes have been synthesized, in order to

find a more suitable anti-tumour drug. Only a small number has entered clinical trials and only two other compounds, *carboplatin* and *oxaliplatin*, are currently used worldwide for the treatment of cancer. These three compounds are used in the treatment of approximately 50% of all cancer cases [2].

The use of these compounds is restricted because of severe dose-limiting side effects; nausea, vomiting, nephrotoxicity, neurotoxicity, myelo-

suppression, and ototoxicity are frequently observed [3]. To overcome these problems, the development of novel chemotherapeutics with a different activity profile and mode of action, with lower toxicity and higher selectivity, is essential for the improved treatment of cancer. As a consequence, complexes with other metal ions have become an area of intensive research. The cytotoxic potential of ruthenium complexes was found three decades ago and the utilisation of ruthenium offers several advantages over platinum-based chemo-

therapy [4, 5]. Ruthenium has the ability to mimic iron in binding to biomolecules, which leads to lower toxic side effects and a different mode of action. Many ruthenium-based drugs are now promising anti-tumour agents, showing lower toxicity *in vitro* and high activity in *in vivo* models. Some of them, KP1019 and NAMI-A (Figure 1), have passed clinical trials and given promising results. The complexes have demonstrated lower general toxicity and activity towards platinum-based drug-resistant tumours [6].

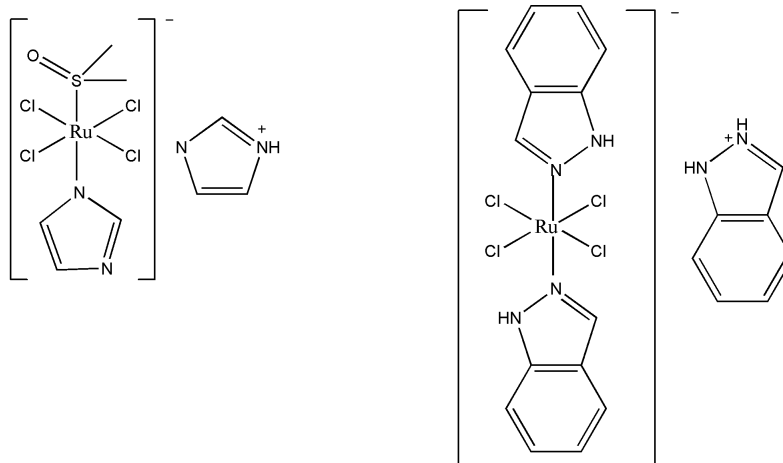


Fig. 1. Ru(III) complexes in clinical trials; NAMI-A and KP1019

It is known that Ru(II) readily binds to ligands containing sulphur and nitrogen donor atoms. In recent years, the Ru(II) bi- and polypyridyl complexes have been a major focus, as a result of their good photochemical properties and their exhibited anticancer activity [7, 8].

During the last two decades, large studies have focused on the field of photodynamic therapy (PDT), in which cancer cells are treated with photo-activatable drugs [9]. In this way, side effects can be reduced, which may allow the administration of higher dosages of drugs. The principle of PDT involves the administration of the photoactive agent followed by local illumination of the tumour with light of appropriate wavelength to activate the specific drug. Upon illumination, the PDT agent is excited from its ground state to a singlet excited state, from where it may decay directly back to the ground state, or undergo intersystem crossing (isc) to its long-lived triplet excited state [10, 11] (Figure 2). The long-lived triplet excited state of the PDT agent sensitises the production of singlet oxygen ($^1\text{O}_2$) or produce other reactive oxygen species (ROS), both of which can damage biomolecules [12].

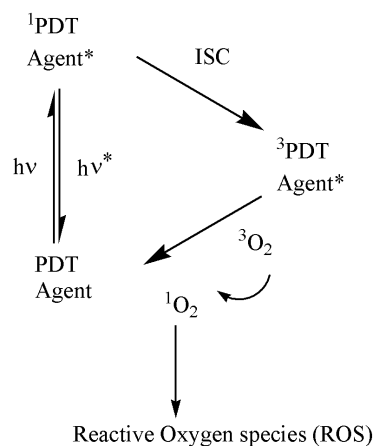


Fig. 2. Photosensitization pathways of a PDT agent

Complexes of rhodium [13, 14] and ruthenium [15–17] have been extensively studied as agents for PDT. Some of them showed much better activity than *cisplatin* [18]. A special group of ruthenium complexes are compounds of the formula $[\text{Ru}(\text{bpy})_2(\text{L-L})]^{2+}$, with L-L representing the chelating *N,N'*-ligand. Since Chaberek and Martell first described the chelating ability of ethylenediamine-*N,N'*-diacetic acid (H_2edda), it has been of interest

for investigation of the mode of coordination of this type of ligand to various metal ions [19]. This class acts toward different compounds including the esters of (*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoic acid. The biological activity of these ligands was investigated towards different cancer cell lines. The results obtained indicate the remarkable anticancer activity of these compounds [20], which is comparable to *cisplatin*, or in the case of ethyl ester, even better than *cisplatin*.

In this paper, we report on the synthesis and characterization of ruthenium(II)-bipyridyl complex with *O,O'*-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate. This ligand is bidentate and neutral, with an aliphatic cyclohexyl substituent. Due to the bulky ligand and its conformation, it was hypothesized that the synthesized complex has octahedral, *cis*-geometry (Figure 3). The coordination mode of the ligand was proposed on the basis of UV-Vis, IR, ^1H , ^{13}C NMR spectroscopy, and ESI-MS spectrometry, compared with the already described analogous complexes and ligands.

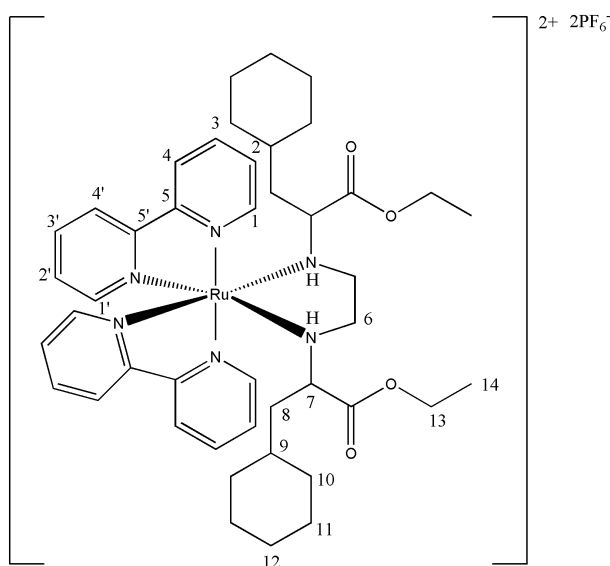


Fig. 3. The proposed structure of the synthesized complex

2. EXPERIMENTAL

2.1. Materials and methods

(*S*)-2-amino-3-cyclohexyl-propanoic acid hydrochloride was purchased from Senn Chemicals (Dielsdorf, Switzerland). *O,O'*-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate [20] and *cis*-[RuCl₂(bpy)₂] [21] were obtained as described. Solvents were obtained commercially and used without further purification.

Elemental analyses were carried out with the Elemental Vario EL III microanalyzer. Infrared spectra were recorded on a Nicolet 6700 FT-IR spectrometer using the ATR technique. The NMR spectra were recorded on a Varian Gemini 200 spectrometer. Chemical shifts for ^1H and ^{13}C spectra were referenced to residual ^1H and ^{13}C presented in deuterated dimethylsulphoxide. Mass spectra were carried out with a 6210 Time-of-Flight LC-MS instrument (G1969A, Agilent Technologies) in acetonitrile. An electronic spectrum was carried out on a GBC UV-Visible Cintra 6 spectrometer, in acetonitrile, 1×10^{-4} mol dm⁻³ solution of complex. Melting point was determined on Electrothermal melting point apparatus.

2.2. Synthesis of ligand dihydrochloride, L·2HCl

O,O'-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate dihydrochloride was synthesized by following an already described procedure [20]. Thionyl chloride, 2.00 ml (2.70 mmol) and (*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoic acid dihydrochloride, 1.20 g (2.70 mmol) were added to 15 ml ice cooled absolute EtOH. After stirring for 1 h at 0 °C, the suspension was held at reflux for 16 h. The resulting mixture was filtered and left to crystallise. The product was filtered, washed with EtOH and air-dried. Compound L·2HCl is a white powder, 0.56 g, (41.42 %); m.p: 208 °C; ^1H NMR (200 MHz, DMSO-*d*₆) δ /ppm: 0.89 (*m*, C7, 4H), 1.26 (*m*, C5, C6, 8H; CH₃CH₂OOC-, 6 H), 1.72 (*m*, -CH₂Cy; C4, 2H; C5, 4H; C6, 4 H), 3.96 (*s*, CH₃CH₂OOC-, 4H), 4.05 (*m*, -NH₂CH₂CH₂NH₂-, 4H), 4.25 (*m*, -OOCCHNH₂-, 2H), 10.09 ppm (*m*, -NH₂CH₂CH₂NH₂-, 4 H); ^{13}C NMR (50 MHz, DMSO-*d*₆,) δ /ppm: 14.1 CH₃CH₂OOC-, 25.6 (C7), 25.6 (C4), 31.8 (C6), 33.4 (C5), 36.9 (C3), 57.3 (C8), 62.3 (C2; CH₃CH₂OOC-), 169.3 ppm (C1); IR (ATR, cm⁻¹): 2900–2500, 1739, 1450, 1215, 802; MS (LC-MS, 4000 V) *m/z*: 425 ([M-2HCl+H]⁺, 45.01%). Calcd. mass fractions of elements, w/%, for C₂₄H₄₆O₄N₂Cl₂·0.5 H₂O (*M*_r = 497.54) are: C 56.90, H 9.35, N 5.53; found: C 56.40, H 8.92, N 5.50.

2.3. Synthesis of starting complex, *cis*-[RuCl₂(bpy)₂]

The synthesis was performed according to a slightly modified method of Sullivan [21]. RuCl₃·xH₂O 1.30 g (5 mmol), 2,2'-bipyridine 1.56 g (10 mmol) and LiCl 2.1 g (50 mmol) were added in 12.5 ml DMF and refluxed for 8 h with stirring. Afterwards, the solution was cooled to room temperature and 100 ml of a mixture of acetone/water

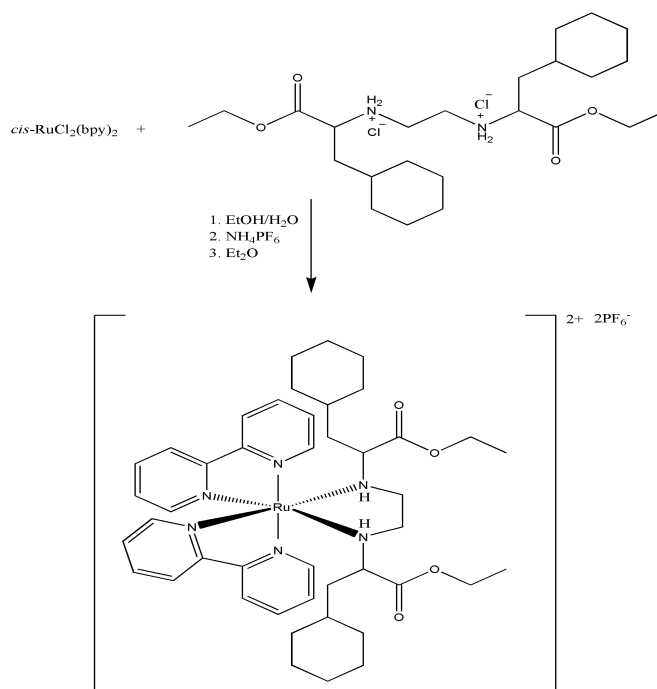
(1:1) was added. A green-black solid was separated by filtration. The solid was added into 100 ml water and stirred for 10 h at r.t., followed by filtration. The product was then washed three times with 12.5 ml of water and ether. The yield was 80% based on the ruthenium salt; $^1\text{H NMR}$ (200 MHz, $\text{DMSO-}d_6$) δ/ppm : 7.15 (*s*, C2', 2H), 7.34 (*m*, C1', 2H), 7.60 (*m*, C3', 2H), 7.71 (*s*, C2, 2H), 8.06 (*t*, C3, 2H), 8.51 (*d*, C4', 2H), 8.62 (*d*, C4, 2H), 9.82 ppm (*s*, C2, 2H); $^{13}\text{C NMR}$ (50 MHz, $\text{DMSO-}d_6$) δ/ppm : 122.78 (C4 and C4'), 125.55 (C2 and C2'), 134.76 (C3 and C3'), 153.04 (C1 and C1'), 160.91 ppm (C5 and C5'); IR (ATR, cm^{-1}): 3491.4, 3098.9, 3068.5, 3037.6, 1604.4, 1450.4, 1417.3, 1261.4, 1014.3, 762.0. Calcd. mass fractions of elements, *w*%, for $\text{C}_{20}\text{H}_{16}\text{N}_4\text{Cl}_2\text{Ru}\cdot 1.5\text{H}_2\text{O}$ ($M_r = 511.37$) are: C 47.72, H 3.80, N 11.13; found: C 47.89, H 3.59, N 11.13.

2.4. Synthesis of ruthenium(II) complex, *cis*-[Ru(bpy)₂L](PF₆)₂

cis-[RuCl₂(bpy)₂] $\cdot 1.5\text{H}_2\text{O}$ 0.10 g (0.20 mmol) was dissolved in 10 ml mixture of ethanol/water (1:1), with heating at 80°C. Suspension of the ligand *O,O'*-diethyl-(*S,S*)-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate dihydrochloride 0.10 g (0.20 mmol) in ethanol (10 ml) was neutralized with $1.66\cdot 10^{-2}$ g (0.4 mmol) of LiOH $\cdot\text{H}_2\text{O}$. During 1 h, the ligand was dropped slowly, and the reaction mixture was left for 4 h under reflux. Af-

ter reaction, solution was concentrated *in vacuo* to 10 ml and 0.06 g solid NH₄PF₆ (0.37 mmol) was added. The mixture was stirred at r.t. for 1 h. The formed precipitate was filtered off and washed with 5.0 ml ether, and left in a desiccator to dry.

cis-[Ru(bpy)₂L]: 63.32 %; dec. temp. 182°C; UV-Vis (CH_3CN) $\lambda_{\text{max}}/\text{nm}$: 380 and 471 ($\epsilon/\text{dm}^3\text{mol}^{-1}\text{cm}^{-1}$: 7386.76 and 4845.59); $^1\text{H NMR}$ (200 MHz, $\text{DMSO-}d_6$) δ/ppm : 0.87 (*t*, $\text{CH}_3\text{CH}_2\text{OOC-}$, 6H), 0.90–1.38 (*m*, C10, 4H; C11, 4H; C12, 4H; $-\text{CH}_2\text{Cy}$, 4H; C9, 2H), 1.38–1.96 (*m*, C10, 4H; C11, 4H; $-\text{NHCH}_2\text{CH}_2\text{NH-}$, 4H), 2.09 and 2.92 (*m*, $-\text{OOCCHNH}_2$, 2H), 4.19 (*m*, $\text{CH}_3\text{CH}_2\text{OOC-}$, 4H), 7.18 (*m*, C2 and C2', 4H), 7.91 (*m*, C1 and C1', 4H), 8.25 (*m*, C3 and C3', 4H), 8.60 (*m*, C4 and C4', 4H), 8.87 and 9.84 ppm (*d*, $-\text{NHCH}_2\text{CH}_2\text{NH-}$, 2H); $^{13}\text{C NMR}$ (50 MHz, $\text{DMSO-}d_6$, δ/ppm) 13.48 and 14.08 ($\text{CH}_3\text{CH}_2\text{OOC-}$), 25.88 (C₁₁ and C₁₂), 33.38 (C₉ and C₁₀), 34.90 ($-\text{CH}_2\text{Cy}$), 57.12 ($-\text{NHCH}_2\text{CH}_2\text{NH-}$), 59.75 ($-\text{OOCCHNH}_2$), 61.40 ($\text{CH}_3\text{CH}_2\text{OOC-}$), 123.93 (C₄ and C_{4'}), 127.59 (C₂ and C_{2'}), 137.28 (C₃ and C_{3'}), 151.10 and 152.79 (C₁ and C_{1'}), 157.43 and 158.36 (C₅ and C_{5'}), 171.52 ppm ($\text{CH}_3\text{CH}_2\text{OOC-}$); IR (ATR, cm^{-1}): 3420.7, 2927.1, 2852.4, 1735.6, 1634.2, 1604.1, 1447.8, 1268.9, 1024.5, 842.4, 767.8, 734.8; MS *m/z*: 419.18 (M^{2+} , 100%) and 983.33 ($[\text{MPPF}_6]^+$). Calcd. mass fractions of elements, *w*%, for $\text{C}_{44}\text{H}_{60}\text{O}_4\text{N}_6\text{P}_2\text{F}_{12}\text{Ru}$ ($M_r = 1127.98$) are: C 46.85; H 5.36; N 7.45; found: C 46.54; H 5.26; N 7.18.



Scheme: Synthesis of the complex

3. RESULTS AND DISCUSSION

Complex *cis*-[Ru(bpy)₂L](PF₆)₂ was synthesized by the reaction of *cis*-[RuCl₂(bpy)₂] and L·2HCl in the presence of two equivalents of the base, LiOH, in a mixture of ethanol/water solution (Scheme). The desired compounds were precipitated after the addition of ammonium hexafluorophosphate from the reaction mixture. The compound is soluble in acetonitrile and partially soluble in dimethylsulphoxide, but not in water, ethanol or chloroform.

3.1. Spectroscopic characterization

The IR spectrum of the free ligand (L·2HCl) was compared with the corresponding spectrum of ruthenium(II) complex, *cis*-[Ru(bpy)₂L](PF₆)₂ to confirm supposed coordination to the metal ion. In the infrared spectrum of the complex, a band around 3420 cm⁻¹, which is typical for NH vibrations of secondary amines, was observed. In the region 2927–2852 cm⁻¹, strong and weak bands correspond to CH/CH₂ vibrations of the cyclohexyl group and aliphatic chains. A lack of valence vibrations in the region 2586–2213 cm⁻¹, which corresponds to valence vibrations of secondary ammonium salt in the free ligand, indicates the coordination *via* nitrogen donor atoms of the ester ligand. Strong bands corresponding to valence C=O vibrations are in the same position as in the spectrum of the ligand, suggesting that the ligand is not coordinated *via* oxygen. Other bands are at approximately the same position as in the spectrum of the ligand. The ¹H NMR spectrum of complex showed characteristic bands from the 2,2'-bipyridine ligand in the region 7–10 ppm. Also, NH protons from ligand are, in this region and the sum of the integral absorption corresponds to the number of protons from 2,2'-bipyridine ligand and two NH protons from ester. Protons from ethylene part of ring of complex and protons from –CHNH– group are moved to lower value of shifts, as a consequence of the coordination of the ligand to metal. In the complex, the symmetry of ligand environment is lowered. In the spectrum of complex this is manifested by the non-equivalence of the chemically equivalent protons in free ligands, –(ROOC)-CHNHCH₂CH₂NHCH(COOR)–. Signals in the range of 0.87–1.96 ppm correspond to cyclohexyl protons, protons from alkyl group (CH₃CH₂OOC–) and protons from –CH₂Cy and ethylene moiety of ligand. Based on the interpretation of the ¹³C NMR spectra of the starting complex, free ligand and synthesized complex, it is clear that only carbons of the ethylene part of the ring in the complex are

significantly moved (in the free ligand these are at 41.81 ppm, but in the complex are at 57.12 ppm). Since these carbons are directly bonded for nitrogen atoms, these shifted signals are the result of coordination of the ligand to metal. The signal for the carbonyl group is at 171.52 ppm. Signals at 158.36, 157.43, 152.79, 151.10, 137.29, 127.59 and 123.93 ppm correspond to carbon atoms from 2,2'-bipyridine ligands and these signals show similar shifts in the starting complex. The signal at 61.40 ppm corresponds to carbons from ester parts, which are directly bonded to oxygen, and signals at 14.08 and 13.48 ppm correspond to terminal carbons from the ester part. Signals at 25.86 and 33.38 ppm are from carbons from the cyclohexyl group, and the signal at 59.75 ppm is attributed to carbon from the –OOCCHNH₂– part of the ligand. Since, the complex cation ion, *cis*-[Ru(bpy)₂L]²⁺ (molecular mass: 838.36), is twice positively charged, the molecular ion was detected at a value of *m/z* = 419.18. The ion ([MPF₆]⁺) was also detected at 983.33.

The synthesized complex has two peaks, at 380 and 471 nm, which correspond to the *d-d* transition. The appearance of two peaks in the electronic spectrum of *d⁶* system is typical for octahedral geometry. This transition is in the range of transition bands for the *d⁶* system and ¹A_{1g} ground state for octahedral geometry of ruthenium(II) complexes [22]. Octahedral complexes demonstrate less intensive absorptions than complexes with tetrahedral geometries. Since Δ_t is smaller than Δ_o, the *d-d* transition in the octahedral complex occurs at a higher energy, and hence at a shorter wavelength.

The assumption that this complex has octahedral geometry is supported by its electronic spectrum (Figure 4).

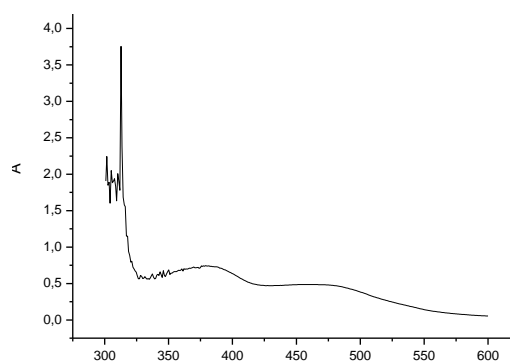


Fig. 4. Electronic spectrum of the synthesized complex

Efforts to obtain crystals suitable for X-ray analysis included: evaporation of solvent (acetonitrile or DMSO or acetonitrile/DMSO) at room temperature, cooling a saturated solution of the

complex in acetonitrile or acetonitrile/DMSO and by solvent diffusion, where we used: water, ethanol, or chloroform as an anti-solvent. None of these methods gave the crystals of desired quality.

4. CONCLUSION

The complex *cis*-[Ru(bpy)₂L](PF₆)₂, where the L is *O,O'*-diethyl-*(S,S)*-ethylenediamine-*N,N'*-di-2-(3-cyclohexyl)propanoate, has been characterized by means of ¹H and ¹³C NMR, elemental analysis, ESI-MS, IR and UV-Vis spectroscopy. This complex precipitates as a red powder, which is soluble only in acetonitrile and dimethylsulphoxide. The bidentate ligand is coordinated in the *cis* position, yielding the complex of octahedral geometry. This was confirmed on the basis of the IR, UV-Vis, ¹H NMR, ¹³C NMR spectroscopy and ESI-MS spectrometry. The aims of our further work are the investigation of cytotoxicity and the possibility that this complex is used as a PDT agent.

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REFERENCES

- [1] Z. Guo, P. J. Sadler, Medicinal inorganic chemistry. *Adv. Inorg. Chem.*, **49**, 183–306 (2000).
- [2] M. A. Jakupec, M. Galanski, V. B. Arion, C. G. Hartinger, B. K. Keppler, Anti-tumour metal compounds: more than theme and variations. *Dalton Trans.*, 183–194 (2007).
- [3] M. Galanski, M. J. Jakupec, B. K. Keppler, Update of the preclinical situation of anticancer platinum complexes: novel design strategies and innovative analytical approaches. *Curr. Med. Chem.*, **18**, 2075–2094 (2005).
- [4] R. Margalit, H. B. Gray, M. J. Clarke, L. Podbielski, Chemical and biological properties of pentaammine-ruthenium-bleomycin complexes. *Chem.-Biol. Interact.*, **59**, 231–245 (1986).
- [5] G. Y. Park, J. J. Wilson, Y. Song, S. J. Lippard, Phenanthriplatin, a monofunctional DNA-binding platinum anticancer drug candidate with unusual potency and cellular activity profile. *P. Natl. Acad. Sci., USA*, **109**, 11987–11992 (2012).
- [6] A. Bergamo, G. Sava, Ruthenium anticancer compounds: myths and realities of the emerging metal-based drugs. *Dalton Trans.*, **40**, 7817–7823 (2011).
- [7] K. Du, J. Q. Wang, J. F. Kou, G. Y. Li, L. L. Wang, H. Chao, L. N. Ji., Synthesis, DNA-binding and topoisomerase inhibitory activity of ruthenium(II) polypyridyl complexes, *Eur. J. Med. Chem.*, **46**, 1056–1065 (2011).
- [8] L. Salassa, Polypyridyl metal complexes with biological activity. *Eur. J. Inorg. Chem.*, **32**, 4931–4947 (2011).
- [9] C. M. Allen, W. M. Sharman, J. E. V. Lier, Current status of phthalocyanines in the photodynamic therapy of cancer. *J. Porphyr. Phthalocya.*, **5**, 161–169 (2001).
- [10] R. E. Cuenca, R. R. Allison, C. Sibata, G. H. Downie, Breast cancer with chest wall progression: treatment with photodynamic therapy, *Ann. Surg. Oncol.*, **11**, 322–327 (2004).
- [11] Z. Lin, W. Shaohua, G. Xuefeng, Z. Jiahong, Y. Boyang, S. Jian, External heavy-atomic construction of photosensitizer nanoparticles for enhanced in vitro photodynamic therapy of cancer, *J. Phys. Chem. B.*, **116**, 12744–12749 (2012).
- [12] S. K. Pushpan, S. Venkatraman, V. G. Anand, J. Sankar, D. Parmeswaran, S. Ganesan, T. K. Chandrashekr, Porphyrins in photodynamic therapy – a search for ideal photosensitisers. *Curr. Med. Chem.-Anti-Cancer Agents*, **2**, 187–207 (2002).
- [13] J. Wang, S. L. H. Higgins, B. S. J. Winkel, K. J. Brewer, A new Os, Rh bimetallic with O₂ independent DNA cleavage and DNA photobinding with red therapeutic light excitation, *Chem. Commun.*, **47**, 9786–9788 (2011).
- [14] M. Kang, H. T. Chifotides, K. R. Dunbar, 2D NMR study of the DNA duplex d(CTCTCAACTTCC)-d(GGAAGTTGAGAG) cross-linked by the antitumor-active dirhodium(II,II) unit at the cytosine-adenine step, *Biochemistry*, **47**, 2265–2276 (2008).
- [15] L. Yao, R. Hammitt, D. A. Lutterman, L. E. Joyce, R. P. Thummel, C. Turro, Ru(II) Complexes of new tridentate ligands: Unexpected high yield of sensitised ¹O₂. *Inorg. Chem.*, **48**, 375–385 (2009).
- [16] E. Corral, A. C. Hotze, A. Magistrato, J. Reedijk, Interaction between the DNA model base 9-ethylguanine and a group of ruthenium polypyridyl complexes: kinetics and conformational temperature dependence, *Inorg. Chem.*, **46**, 6715–6722 (2007).
- [17] K. Karidi, A. Garoufis, N. Hadjiliadis, M. Lutz, A. L. Spek, J. Reedijk, Synthesis, characterization, and DNA-binding studies of nitro(oligopyridine)ruthenium(II) complexes, *Inorg. Chem.*, **45**, 10282–10292 (2006).
- [18] D. A. Lutterman, P. K. Fu, C. Turro, *cis*-[Rh₂(μ-O₂CCH₃)₂(CH₃CN)₆]²⁺ as a photoactivated cisplatin analogue. *J. Amer. Chem. Soc.*, **128**, 738–739 (2006).
- [19] S. J. Chaberek, A. E. Martell, Ethylenediaminediacetic acid and *N,N'*-ethylenediaminediacetic-*N,N'*-dipropionic acid, *J. Am. Chem. Soc.*, **74**, 6228–6231 (1952).
- [20] J. M. Lazić, Lj. Vučićević, S. Grgurić-Šipka, K. Janjetović, G. N. Kaludjerović, M. Misirkić, M. Gruden-Pavlović, D. Popadić, R. Paschke, V. Trajković, T. J. Sabo, Synthesis and *in vitro* anticancer activity of octahedral platinum(IV) complexes with cyclohexyl-functionalised ethylenediamine-*N,N'*-diacetate-type ligands. *Chem. Med. Chem.* **5**, 881–889 (2010).
- [21] B. P. Sullivan, D. J. Salmon, T. J. Meyer, Mixed phosphine 2,2'-bipyridine complexes of ruthenium, *Inorg. Chem.* **17**, 3334–3341 (1978).
- [22] D. Mishra, S. Naskar, M. G. B. Drew, S. K. Chattopadhyah, Synthesis, spectroscopic and redox properties of some ruthenium(II) thiosemicarbazone complexes: Structural description of four of these complexes, *Inorg. Chim. Acta*, **359**, 585–592 (2006).