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Some Luminescence Characteristics of Ytterbium-Acridines

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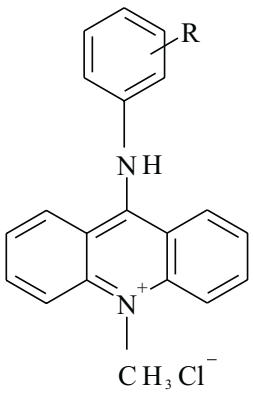
Luminescence properties of Ln(III) ions in solutions of complexes with the different types of organic ligands are interesting, as they are the base for the creation of labels for immunofluorescence assay [1,2]. It is known that among indicating ions are those, which emitted in the near IR spectral region, where interfering radiation from bio-objects is minimal. First of all, these are the ytterbium Yb(III) and neodymium Nd(III) ions [3–5].

Anilinoacridines and some of their derivatives are known owing a high anti-tumor activity [6]. It was determined that they have a higher bio-availability and lower toxicity [7]. These peculiarities are of great interest. This work concerns a study of some luminescence properties of new ytterbium complexes with anilinoacridine derivatives. Preliminary results of investigations of their cytotoxicity are reported also. The structures of the ligands are given in Fig. 1.

All starting materials were of analytical grade. The ligands L_1–L_6 (acridinium salts) were synthesized according to [8] *via* alkylation of acridan-9-one by different alkylation agents and the subsequent transformation of 10-alkylacridan-9-ones by thionyl chloride into 10-alkyl-9-chloracridinium salt condensed with the anilines in alcohol. The composition of all ligands was confirmed by elemental analyses for C, H, and N (within the range ± 0.2%). Mass spectroscopy, ^{1}H NMR and TLC controlled



	R
L_1	*o*- OH
L_2	*m*-OH
L_3	*p*- OH
L_4	*o*- COOH
L_5	*m*-COOH
L_6	*p*- COOH

Figure 1. Structures of the ligands studied.

the purity of prepared compounds. The 1 mmol solutions of complexes were prepared by dissolving the appropriate weights of the solid compounds in the solvents and diluted as needed.

Absorption spectra of ligands and complexes solutions were recorded on a Specord M-40 UV/VIS (Carl Zeiss, Jena, Germany) spectrophotometer. Luminescence measurements were carried out on an SDL-2 spectrofluorimeter equipped with a photon counting system (Leningrad Optic-Mechanic Association, St. Petersburg, Russia) [9]. All spectra were corrected with a standard lamp. The 254-, 313- and 365-nm lines cut with filters from the light of a DRSh-250 mercury quartz lamp or a xenon lamp (Xe-150) excited luminescence. The solutions of complexes (1 ml) were placed into 10-mm quartz cuvette. The emission of the solutions was recorded at an angle of 90° to the exciting radiation. The relative quantum yields of luminescence (ϕ) for the ytterbium complexes (λ_{exc} = 280 nm) were calculated by the method described in [10], with the use of Zn-tetraphenylporphyrin (ϕ = 0.0315 in ethanol) as a standard [11].

The influence of ligands and complexes on cell division and growth were studied by the root test with use of sprouts of *Cucumis sp*. [8]. The cytotoxicity of the compounds was detected by decreasing the lengths of the main root and area of the side root growth. The cytotoxicity activity was expressed as concentration of the complex in $\mu g\ ml^{-1}$ (IC_{50}). At this concentration growth of *Cucumis sprouts* showed 50% inhibition, and was calculated by linear regression analysis [12].

Absorption spectra of the ligands L_1–L_6 in water-ethanol solutions (50:50) are characterized by one strong band (λ_{max} = 220 nm, extinction coefficients $\varepsilon = 32–36 \times 10^4$) and one band with middle intensity (λ_{max} = 260 nm, $\varepsilon = 10–17 \times 10^4$) in the UV spectral region corresponding to the transitions in the aromatic groups. Moreover, a weak band ($3–10 \times 10^4$) at the 390–420 nm has been observed for the all ligands. This last band is significantly changed in shape for the ytterbium complexes and the maximum is shifted by 25–35 nm relative to the free ligands. For instance, Fig. 2 demonstrates the absorption spectra of free L_6 and Yb–L_6 complex. Above mentioned differences in these spectra prove that complexation takes place.

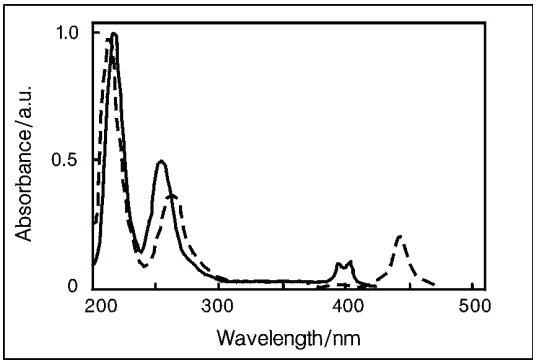


Figure 2. Absorption spectra of L_6 (——) and Yb–L_6 (-----) in water-ethanol solution (50:50).

Excitation in the maximum of the long wave absorption band of the complexes causes 4f-luminescence of Yb(III) ions. A similarity between the absorption and excitation spectra of the complexes indicates that ligand-to-lanthanide ion energy transfer takes place [13]. The positions of the ligand triplet levels lie in the range of 19350–20800 cm^{-1}. These results show that all ligands are able to transfer effectively excitation energy to the Yb(III) ion, whose emitting level is located at 10250 cm^{-1} (Fig. 3). All the ligands display molecular luminescence upon the excitation at 254 nm. Their luminescence structureless bands are situated in the range 460–540 nm. The ligand luminescence is essentially (85–90%) reduced in the complexes. These data demonstrate the effective energy transfer from the organic moiety of the molecule to the Yb(III) ion [13]. The values of the relative quantum yields for Yb(III) in the complexes considered are given in Table 1. The Yb–L_3 and Yb–L_6 complexes show the highest quantum efficiencies compared with the others. Obviously, the reason is the following: As determined by the fluorescence method [14], the relationship Yb:L_3 and Yb:L_6 are equal 1:2. For the rest of complexes it is 1:1. Undoubtedly, the steric factor plays the main role. Thus, the inner coordination sphere of the complexes with the ligands L_3 and L_6 has less water molecules, than in the case of the ligands, containing in the phenyl ring *o*- and *m*- coordinately active substituents. It is known that highly frequent (3500 cm^{-1}) O-H oscillations essentially reduce the intensity of 4f-luminescence of Ln(III) ions. This assumption correlates with the fact, that in D_2O medium, where quenching of luminescence is not strong (oscillation frequency O-D is 2600 cm^{-1}), the increase of the luminescence intensity (*I*) at conversion from H_2O to D_2O is more considerable just for *o*- and *m*-isomers.

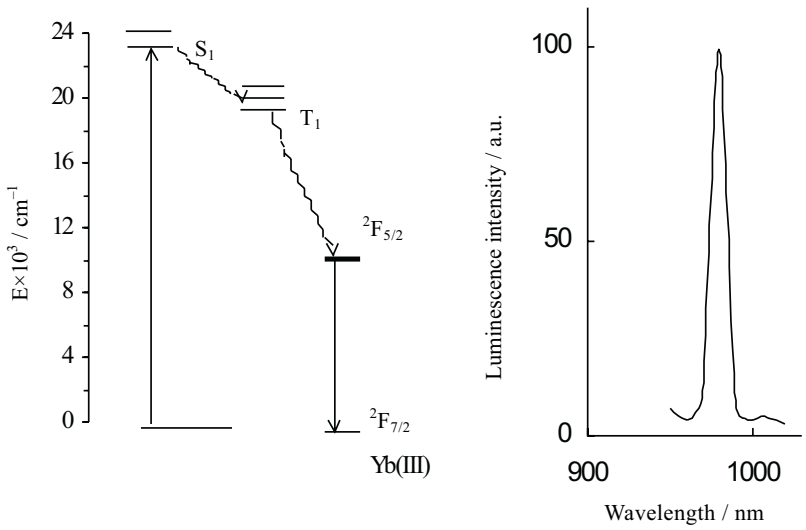


Figure 3. Energy transfer diagram for the sensitized emission of Yb(III) by acridines and the luminescence spectrum of Yb(III).

Table 1. Some luminescence characteristics of Yb-acridines at 25°C.

Complex	$\phi \times 10^{-5}$	I_{D_2O/H_2O}	I_{n-s/H_2O}
L_1	1.6	19.2	7.8
L_2	0.3	21.5	7.2
L_3	2.6	18.4	7.0
L_4	0.7	20.7	7.4
L_5	1.7	16.1	6.2
L_6	3.4	15.8	6.0

Data about increasing of *I* in the media of nonionic surfactants ($I_{n/s}$) have also been given in Table 1. They indicate that increase of the media viscosity leads to the increase of the 4f-luminescence. Most *I* of complexes were obtained in neutral media. The luminescence intensity was practically not reduced during 20 min of radiation light. The latter circumstances are especially important for the further use of Yb(III)-luminescence in the complexes with the acridines in biomedicine.

However, the following should be noted. Relationship Yb:L (1:1 and 1:2), low denticity of the ligands and high coordination number of ytterbium testify for that the inner coordination sphere of complexes has no more than four-five water molecules. Namely, this fact decreased the 4f-luminescence efficiency. Therefore, it should be considered that given ligands are not optimal for manifestation of high luminescent properties of Yb(III) ions in their complexes. But the way for this optimization can be predicted on the base of obtained results [15,16], which allow to use acridine-derivatives. The highest quantum yields of 4f-luminescence for lanthanide ions, emitting in the near-IR spectral region, is observed in the complexes with the macrocyclic ligands containing the chromophore-groups as substituents. These substituents do not participate directly in complexation, but they are so called "photo-antenna" – the accumulators of excitation energy. This energy is transferred to radiative levels of Ln(III) ions by intermolecular mechanism. The molecules of acridines by their nature are appropriate for such photo-antennae. Examples of such structures are for instance some of crown ether-linked acridine derivatives [17]. In the nearest time we are going to publish our recent results, demonstrating that in ytterbium complexes with the linked azamacrocyclic-acridine-chromophores ligands the 4f-luminescence quantum yields are higher by more than one order as compared to the values listed in Table 1.

It is noteworthy, that according to the data on the cytotoxicity (Table 2), there are rather strong inhibitors of cell division among considered compounds (L_1 and L_2), which demonstrate the inhibiting effect on the micromolar range of concentrations. The complexes have a lower cytotoxicity in comparison with ligands. The IC_{50} of complexes with *o*- and *m*-substituents increases approximate 2–2.5 times, whereas this value for *p*-derivatives does not change. Undoubtedly, more detailed spectroscopic and biological investigations are required in order to recommend some of ytterbium-acridines as luminescence labels.

Table 2. Cytotoxicity data for ligands and their complexes.

Ligand	IC_{50}, μm	Complex	IC_{50}, μm
L_1	33	Yb–L_1	64
L_2	27	Yb–L_2	52
L_3	62	Yb–L_3	79
L_4	112	Yb–L_4	209
L_5	190	Yb–L_5	> 500
L_6	79	Yb–L_6	83

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REFERENCES

1. Bünzli J.C.G., in: Lanthanide Probes in Life, Chemical and Earth Sciences. Theory and Practice (Eds. J.C.G. Bünzli and G.R. Choppin), Elsevier, Amsterdam, 1989, p. 219.
2. Gudgin Dickson E.F., Pollak A. and Diamandis E.P., *J. Photochem. Photobiol. B: Biol*., **27**, 3 (1995).
3. Shevchuk S.V., Rusakova N.V., Turianskaya A.M., Korovin Yu.V., Nazarenko N.A. and Gren A.I., *J. Fluorescence*, **8**, 225 (1998).
4. Werts M.H.V., Woudenberg R.H., Emmerink P.G., van Gassel R., Hofstraat J.W. and Verhoeven J.W., *Angew. Chem. Int. Ed*., **39**, 4542 (2000).
5. Korovin Yu. and Rusakova N., *Rev. Inorg. Chem*., **21**, 299 (2001).
6. Albert A., in: The Acridines. Their Preparation, Physical, Chemical, and Biological Properties and Uses, 2nd ed., (Ed. E. Arnold), William Clowes and Sons, London, 1966, Ch. 23, p. 517.
7. Cain B.F., Atwell G.J. and Denny W.A., *J. Med. Chem*., **19**, 772 (1976).
8. Suveyzdis Y.I., Kostenchuk M.M. and Rusakova M.Yu., *J. Pharmaceutical*, 59 (2000).
9. Beltyukova S.V., Balamtsarashvili G.M. and Kravchenko T.B., *Analyst*, **117**, 807 (1992).
10. Haas Y. and Stein G., *J. Phys. Chem*., **75**, 3677 (1971).
11. Tsvirko M.P., Stelmakh G.F., Pyatosin V.E., Solovyov K.N., Kachura T.F., Piskarskas A.S. and Gadonas R.A., *Chem. Phys*., **106**, 467 (1986).
12. Tallarida R.J. and Murray R.B., in: Manual of Pharmacological Calculations with Computer Programs, Springer, NY, 1987, p. 35.
13. Tsvirko M.P., Stelmakh G.F., Pyatosin V.E., Solovyov K.N. and Kachura T.F., *Chem. Phys. Lett*., **73**, 80 (1980).
14. Meshkova S.B., *J. Fluorescence*, **10**, 333 (2000).
15. Sabbatini N. and Guardigli M., *Coord. Chem. Rev.*, **123**, 201 (1993).
16. Oude Wolbers M.P., van Veggel F.C.J.M, Peters F.G.A., van Beelen E.S.E., Hofstraat J.W., Geurts F.A.J. and Reinhoudt D.N., *Chem. Eur. J*., **4**, 772 (1998).
17. Fukuda R., Takenaka S. and Takagi M., *J. Chem. Soc., Chem. Commun*., 1028 (1990).