Clicked dipicolinic antennae for lanthanide luminescent probes†

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4-Triazolyl-dipicolinic acids, prepared efficiently by CuAAC, proved to act as efficient antennae for optical lanthanide probes containing Eu³⁺ or Tb³⁺, under linear and non linear excitations (one, two and three photons provided by laser induced luminescence). The versatility offered by the substitution of the triazole subunit is, in addition, useful for covalent linkage to slide glass providing luminescent films.

Introduction

The recent emergence of "click chemistry" as a new way of categorizing organic reactions has facilitated an extraordinary expansion in the number of molecules available for medicinal chemistry, biology and material sciences.¹ Among "click" synthetic tools, the copper(I)-catalyzed azide alkyne cycloaddition (CuAAC) has received unrivalled attention and has been found to be of paramount importance to attach fluorescent probes to films, nanoparticles and biomaterials.² Fluorescent lanthanide complexes present several advantages³ compared to regular organic dyes: insensitivity, to oxygen and moderate warming, and to the local environment, narrow electronic transitions with large Stokes shifts, long excitedstate lifetimes and high luminescence intensities induced by increasing significantly the absorption cross-section (in these systems, energy is absorbed by the ligand and efficiently transferred to the emitting metal ion via the so-called antenna effect or Absorption-Transfer-Emission - ATE - mechanism). All these characteristics explain that considerable attention is presently devoted to the design of Light Conversion Molecular Devices based on lanthanides for bioimaging applications.⁴ Pyridines and terpyridines subunits are by far the most common chromophores used for luminescent lanthanide complexes, as these chemical antennae enjoy strong light absorption and triplet sensitizing properties. A wide range of conjugated dipicolinic acid (DPA) derivatives (Fig. 1), recently synthesized, exhibit a high absorption coefficient, broad excitation range, and efficient energy transfer giving rise to significant improvements in detection sensitivity.⁵





These ligands are classically synthesized by palladium coupling (1-3), however, their synthesis is not straightforward, especially if further functionalizations are envisaged.⁶ Moreover, the absorption wavelength depends on the attached group. Changing the absorption range, if needed, would require additional synthetic work. To circumvent the aforementioned efforts, we assumed that a 1–4 subsituted 1,2,3-triazole moiety could serve as relevant and adjustable antenna, allowing good triplet sensitizing properties on a large absorption domain (Fig. 2).⁷



Fig. 2 Click strategy to construct new antennae and materials.

We then decided to synthesise a set of 4-triazolyl-DPA, containing electron withdrawing and electron donating groups in order to study the electronic influence on the luminescence properties of corresponding Tb(III) and Eu(III) chelates.

To allow for their use as chromophores in fluorescence based bioassays or as biomarkers for medical imaging, these Ln complexes should be covalently grafted to solid supports. Furthermore, these fluorescent materials must withstand the heating temperature upon light radiations. To fulfil such requirements,

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organic-inorganic hybrids have been achieved by a sol-gel process from Ln complexes bearing an alkoxysilane moiety. The sol-gel process is a common way to make such materials due to its mild conditions.^{5b,d} The optical properties are provided by the Ln-doped organic moiety while the shaping as well as the mechanical and thermal resistances are brought by the silica network. In this paper are reported preliminary investigations undertaken to prepare strongly fluorescent films, based on lanthanide activated hybrid materials coated onto glass substrates.

Results and discussion

Ligand synthesis

The 4-azido DPA derivative **4a** was synthesized in high yield from dimethyl 4-chlorodipocolinate⁸ by an aromatic nucleophilic substitution with sodium azide, in DMF at 50 °C during 24 h. The pure and stable product **4a**, prone to precipitate in water, is easily obtained by a simple filtration, making this procedure amenable on a multigram scale (Scheme 1).



Scheme 1 Synthesis of dimethyl 4-azidodipicolinate.

The CuAAC reaction was firstly tested with CuSO₄ (30 mol %) and ascorbic acid (60 mol %). Unfortunately, this reaction proved to be too slow (48 h) and yields were disappointing, probably because of a competitive chelation with the pyridyl substrate. To circumvent this matter, we turned our attention to ligands known to bind copper(I) and displaying strong rate acceleration effects. Indeed, several nitrogen containing ligands were considered, such as TBAT (tris-(benzyltriazolylmethyl)amine), (*tris*(3-hydroxypropyltriazolylmethyl)amine) THPTA and $(BimC_4A)_3$, all of them requiring the presence of a sacrificial reducing agent.9 Newly reported catalytic systems, to allow a CuAAC reaction in reducing agent free conditions, were also considered, (namely: $[Cu(C_{18}6tren)]Br$, tris(triazolyl)methanol copper(I), (Aminoarenethiolato) copper(I) and the copper(I) N-heterocyclic carbenes [CuBr(SIMes)], [CuCl(Phen)(SIMes)], [CuCl(SIMes)(4,7-dichloro-1,10-phenanthroline)]).10 Among these possibilities, THPTA and [CuCl(SIMes)(4,7-dichloro-1,10phenanthroline)] were tested. To our delight, the triazoles 7a-d formed in a very efficient manner from 4a using both catalysts. The reactions proceed very efficiently and extremely fast as completion is reached in approximately 30 min for the former and 10 min for the latter (Fig. 3).

It has to be noticed that the reactions are conducted in an open vessel and, in most cases, the products precipitate at completion of the process, satisfying the Click criteria.^{1b} The exception is ligand **7d** that contains an alcohol function for which a simple extraction may be necessary. The yields for the CuAAC reactions are gathered in Table 1.

Finally, targeted DPA derivatives **8a–d** were obtained quantitatively, cleanly and without purification, by saponification of diester **7a–d** followed by acidification (Fig. 4).

Entry	Alkyne	Cu(I)/THPTA	Cu(I)/NHC
1	6a phenylacetylene	94%	94%
2	6b 4-methoxyphenylacetylene	95%	98%
3	6c 4-nitrophenylacetylene	93%	100%
4	6d propargyl alcohol	92%	90%



Fig. 3 General synthesis of 1,2,3-triazoles.



Fig. 4 Saponification of clicked dipicolinate esters.

Complex synthesis

Corresponding solid lanthanide complexes **9a–d** and **10a–d** were prepared using, respectively, europium chloride and terbium nitrate, following Andraud and Maury's procedure.¹¹ Accordingly, complexes **9–10** were obtained within a few minutes by addition of the lanthanide to an aqueous solution of **8a–d** guanidinium salts, and were quantitatively isolated by simple filtration (Scheme 2).

Overall, the synthesis of the ligands and the complexes were conducted without any tedious purification.

Luminescence properties

All the measurements were performed on solid state samples.

Each complex exhibits an excitation spectrum featuring a broad absorption band within the near-UV range, with a shift of the maximum depending on the substituting groups on the triazole subunit. This signal was assigned to an $S_0 \rightarrow S_1$ absorption transition within the organic chromophore. On the basis of excitation spectra and for any rare earth ion, the presence of electron withdrawing groups results in a redshift of the maximum absorption. For both types of lanthanide complexes, the maximum



Scheme 2 Synthesis of clicked DPA complexes

excitation wavelengths are gathered in Table 2 while excitation and emission spectra of compounds **9a–d** are presented in Fig. 5 (for **10** see ESI[†]).

Emission spectra were recorded within the visible range upon excitation at the maximum absorption wavelength for europium and terbium complexes. They consist, respectively, of characteristic ${}^{5}D_{0} \rightarrow {}^{7}F_{J(J=1-2)}$ and ${}^{5}D_{4} \rightarrow {}^{7}F_{J(J=6-3)}$ emission bands of Eu³⁺ and

 Table 2
 Maximum excitation wavelengths for lanthanide complexes

Europium complexes	
9a: 341 nm 10a: 3- 9b: 374 nm 10b: 3' 9c: 387 nm 10c: w 9d: 301 nm 10d: 3-	40 nm 70 nm eak fluorescence 40 nm

Tb³⁺ transitions. Samples **9a–d**, **10a–b**, and **10d** exhibit remarkable fluorescence confirming that a very efficient ATE mechanism takes place. Very weak fluorescence was observed for compound **10c**. This feature is probably due to the strong electron withdrawing effect of the nitro group that stabilizes the singlet and triplet states of the ligand. Thus, the triplet state probably lies too low for an efficient electron transfer to the ⁵D₄ level of the terbium. The decays for complexes **9a–d** and **10a–d** are given in Table 3.

Despite the fact that quantum yields are not reported (due to absence of specific device), two complexes demonstrate promising properties with fluorescence intensities much stronger than other samples *i.e.* complexes **9b** and **10d**. They were retained to measure fluorescence properties upon pulsed OPE (One Photon Excitation), TPE (Two Photon Excitation) and THPE (Three Photon Excitation) regimes. Linear (OPE) and non-linear (TPE



Fig. 5 (a) Excitation and (b) emission spectra recorded at RT from europium complexes. (c) Excitation and (d) emission spectra recorded at RT from terbium complexes. Emission spectra were recorded upon excitation at wavelengths given in Table 2.

Table 3 Constants of decay times for europium and terbium complexes

Europium complexes	Terbium complexes		
9a : 0.87 ms	10a : 0.28 ms		
9b : 0.84 ms 9c : 0.85 ms	10b : 0.04 ms 10c · no signal		
9d : 1.12 ms	10d : 0.67 ms		

and THPE) time resolved emission spectra are consistent with the characteristic optical features of Eu^{3+} and Tb^{3+} ions embedded in **9b** and **10d** complexes respectively (see ESI[†]).

The TPE and THPE processes are unambiguously confirmed by the quadratic and cubic dependence of the ${}^{5}D_{0} \rightarrow {}^{7}F_{2}$ (616 nm) and ${}^{5}D_{4} \rightarrow {}^{7}F_{5}$ (554 nm) emissions with the laser power (TPE: $\lambda_{exc} = 760$ nm; THPE: $\lambda_{exc} = 957$ nm) as shown in Fig. 6 for compound **9b**. The same quadratic and cubic dependences are observed for these anti-Stokes emissions under the experimental conditions employed for both samples. The TPE and THPE maximum wavelengths fit quite perfectly with those recorded by OPE, indicating that the excited states reached by such processes are the same and the ATE mechanism occurs. Such results are very promising for future biphotonic or multiphotonic applications.¹² Table 4Maximum excitation wavelengths and constants of decay timesrecorded from samples 9b and 10d upon OPE, TPE and THPE conditions

	9b ($\lambda_{\text{em.}} = 616 \text{ nm}$)	Decay/ms	10d ($\lambda_{\text{em.}} = 554 \text{ nm}$)	Decay/ms
1 photon	374 nm	0.84 ^a	340 nm	0.67ª
2 photons	762 nm	1.09 ^b	757 nm	0.58
3 photons	990 nm	N.D. ^c	996 nm	N.D. ^c
" Excitation	n at 337 nm. ^{<i>b</i>} Excit	ation at 762	nm. ^e Not determin	ed.

The decay curves of the Tb³⁺ ${}^{5}D_{4} \rightarrow {}^{7}F_{5}$ transition (547 nm) indicate an approximately single exponential profile with lifetime around 0.6 ms under OPE or TPE regimes. To the contrary the decay curve resulting from the Eu³⁺ ${}^{5}D_{0} \rightarrow {}^{7}F_{2}$ transition is bi-exponential in OPE conditions while exhibiting a single exponential shape with a time constant of 0.97 ms in the case of TPE conditions (see ESI†). These results are presently not totally explained but may be due to differences in the selection rules between OPE and TPE regimes and the possible existence of two sites for the Eu³⁺ ion.¹³ All the data are gathered in Table 4.

Grafting on solid support

The first application of these new lanthanide complexes consisted of the preparation of luminescent films, covalently fixed on glass slides. For this purpose, functionalizable ligand **7d** was treated with isocyanate **11** to afford the carbamate **12** (Scheme 3).



Scheme 3 Method for glass surface coating.

To our pleasure, deposition from an ethanol solution of (3:1)12: Ln³⁺ onto a glass substrate by the spin-coating method led to transparent, smooth and homogeneous films with a thickness close to half a micrometre as demonstrated by scanning electron microscopy and profilometry analyses, respectively (see ESI†). In Fig. 7, pictures of the resulting films are shown without and with UV excitation (365 nm). The strong luminescence resulting from these samples emphasizes the outstanding capabilities of these rare earth ion doped hybrids to be used as advanced phosphors in biolabelling applications. Detailed emission spectra recorded at room temperature upon 365 nm excitation are presented in the ESI†.



Fig. 6 (a) quadratic and (b) cubic dependence of the ${}^{5}D_{0} \rightarrow {}^{7}F_{2}$ (616 nm) emission with the laser power (TPE: $\lambda_{exc} = 760$ nm; THPE: $\lambda_{exc} = 957$ nm) in compound **9b**. The straight line corresponds to the linear fit of the experimental data.



Fig. 7 Pictures of coatings achieved from compounds 13a (terbium complex) and 13b (europium complex) without (left) and with (right) excitation by UV light at 365 nm displaying respectively green and red emission.

Conclusion

In this report, we have disclosed an elegant, practical and straightforward synthesis of adjustable antennae for DPA lanthanidebased probes. Complexes exhibit one, two and three photon excitation properties that are of great interest for biomedical applications. Finally, coating these luminescent complexes on glass support using a sol-gel process paves the way for future applications. For this purpose the grafting of modified "clicked" lanthanide complexes onto silica nanoparticles is currently under study. Resulting core-shell type luminescent nanohybrids will be then evaluated as new bio-imaging sensors.

Experimental

Chemistry

NMR spectra were recorded in Fourier Transform mode with a Bruker AVANCE 500 spectrometer (¹H at 500 MHz) or a Bruker AVANCE 400 spectrometer (¹H at 400 MHz, ¹³C at 100 MHz), at 298 K. Data are reported as chemical shifts (δ) in ppm. Residual solvent signals were used as internal references (¹H, ¹³C). Electrospray (positive mode) high-resolution mass spectra were recorded on a Q-TOF micro spectrometer (Waters), using an internal lock mass (H₃PO₄) and an external lock mass (leucineenkephalin [M + H]⁺: m/z = 556.2766). IR spectra were recorded on a Shimadzu Fourier Transform Infrared Spectrophotometer FTIR-8400S.

Synthesis of dimethyl 4-azidopyridine-2,6-dicarboxylate (4a)

A mixture of dimethyl 4-chloropyridine-2,6-dicarboxylate (2.2 g, 9.56 mmol, prepared according to reference 8) and sodium azide (6 g, 10 eq.) in DMF (30 mL) is heated at 50 °C overnight. The yellow reaction mixture is cooled down to room temperature and poured onto 80 mL of cold water under vigourous stirring. Compound **4a** precipitates immediately and a filtration affords 2.0 g (90 %) of a pale yellow solid. Mp: 160 °C. R_f : 0.35 (AcOEt–Cyclohexane = 1 : 1). ¹H NMR (400 MHz, CDCl₃) δ (ppm): 7.93 (2H, s), 4.03 (6H, s). ¹³C NMR (100 MHz, CDCl₃) δ (ppm): 164.5, 151.7, 149.9, 118.0, 53.4. IR (KBr) ν/cm^{-1} : 3075, 2960, 2122, 1753, 1718, 1594, 1445, 1351, 1273, 1250, 1190, 1158. Anal. Calcd for C₉H₈N₄O₄: C, 45.77; H, 3.41; N, 23.72. Found: C, 45.19; H, 3.41; N, 23.75.

General procedure for 7a-d (CuAAC reactions)

Method A (Cu(1)/THPTA). Azide 4a (236 mg, 1 mmol, 1.0 eq.) is suspended in 20 mL of methanol and the appropriate alkyne (1.5 mmol, 1.5 eq.) is added. Then, 200 μ L of copper complex (0.1 M in aqueous stock solution, 0.02 mmol, 2 mol %) followed by 600 μ L of ascorbic acid (0.1 M in aqueous solution, 0.06 mmol, 6 mol %) are added. The solution turns from green to yellow. After 10–20 min the solution returns to green and, if necessary, an additional 600 μ L of ascorbic acid is added. In the cases of **7a–c** the product deposits during the reaction. In these cases the products are obtained pure after a simple filtration followed by washing with cold methanol and ether. In the case of compound **7d**, the reaction is evaporated to 1/3 of the volume. 20 mL of water is added and the solution is extracted with 3×30 mL of CHCl₃. Combined organic extracts are dried over MgSO₄, filtered and then evaporated to furnish pure compound.

Method B [CuCl(SIMes)(4,7-dichloro-1,10-phenanthroline)]. Azide 4a (236 mg, 1 mmol, 1.0 eq.) is suspended in 20 mL of methanol and the appropriate alkyne (1.5 mmol, 1.5 eq.) is added. Then, copper complex (13 mg, 2 mol %) is added in one portion. In cases of 7a–c the product deposits during the reaction. In these cases the products are obtained pure after a simple filtration followed by washing with cold methanol and ether. In the case of compound 7d, the reaction is evaporated to 1/3 of the volume. 20 mL of water are added and the solution is extracted with 3×30 mL of CHCl₃. Combined organic extracts are dried over MgSO₄, filtered and then evaporated to furnish pure compound.

General procedure for 4b, 8a-d

To a 5 mL suspension of diester (1 mmol) in water is added 4 mL of aqueous NaOH (1 M). The reaction mixture is heated at 50 °C until total disappearance of the starting material after which the temperature is maintained for an additional 2 h. After cooling to room temperature, HCl (3 M) is added dropwise until pH = 3. The desired diacid precipitates, and is isolated after simple filtration followed by washing with water and drying under vacuum.

Dimethyl 4-(4-phenyl-1*H*-1,2,3-triazol-1-yl)pyridine-2,6dicarboxylate (7a)

Pale yellow solid. Mp: 240 °C, R_f : 0.35 (ethyl acetate–cyclohexane: 1 : 1).¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 9.84 (1H, s), 8.81 (2H, s), 7.99 (2H, d, J = 8 Hz), 7.53 (2H, dd, J = 7 and 8 Hz), 7.43 (1H, t, J = 7 Hz), 3.99 (6H, s).¹³C NMR (100 MHz, DMSO-d₆) δ (ppm): 163.8, 149.6, 148.0, 144.7, 129.4, 129.0, 128.6, 125.3, 119.9, 116.8, 52.9. IR (KBr) ν/cm^{-1} : 3125, 2957, 1750, 1717, 1599, 1446, 1251, 1223, 1153, 1119, 1047, 1019, 987, 947, 892, 782, 756, 692. HR-ESI-MS calculated for $C_{17}H_{14}N_4O_4Na$ [M + Na]⁺: 361.0913, found: 361.0899.

Dimethyl 4-[4-(4-methoxyphenyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6-dicarboxylate (7b)

Pale yellow solid. Mp: 260.4 °C. R_f : 0.21 (ethyl acetatecyclohexane: 1:1). ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 9.73 (1H, s), 8.80 (2H, s), 7.91 (2H, d, J = 9 Hz), 7.10 (2H, d, J = 9 Hz), 3.99 (6H, s), 3.82 (3H, s). ¹³C NMR (100 MHz, DMSOd₆) δ (ppm): 163.8, 159.5, 147.9, 144.7, 126.8, 121.9, 118.9, 116.8, 114.4, 55.1, 52.9. IR (KBr) ν /cm⁻¹: 3143, 2954, 1748, 1716, 1597, 1479, 1438, 1360, 1323, 1249, 1186, 1156, 1124, 1050, 1030, 956, 846, 752. HR-ESI-MS calculated for C₁₈H₁₆N₄O₅Na [M + Na]⁺: 391.1018, found: 391.1000.

Dimethyl 4-[4-(4-nitrophenyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6dicarboxylate (7c)

Pale yellow solid. Mp: 263.7 °C. R_f : 0.19 (ethyl acetate). ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 10.11 (1H, s), 8.83 (2H, s), 8.43 (2H, d, J = 8 Hz), 8.25 (2H, d, J = 8 Hz), 4.00 (6H, s). ¹³C NMR: compound not soluble enough in common NMR solvents. IR (KBr) ν/cm^{-1} : 3142, 3103, 3062, 2956, 1748, 1711, 1597, 1519, 1476, 1444, 1339, 1259, 1205, 1160, 1124, 1048, 1008, 864, 756. HR-ESI-MS calculated for $C_{17}H_{13}N_5O_6Na$ [M + Na]⁺: 406.0764, found: 406.0745.

Dimethyl 4-[4-(hydroxymethyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6-dicarboxylate (7d)

White solid. Mp: 188 °C. R_f : 0.22 (ethyl acetate–cyclohexane: 1 : 1). ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 9.23 (1H, s), 8.79 (2H, s), 5.54 (1H, t, J = 5.5 Hz), 4.65 (2H, d, J = 5.5 Hz), 3.98 (6H, s). ¹³C NMR (400 MHz, DMSO-d₆) δ (ppm): 163.9, 151.0, 149.7, 144.9, 121.6, 117.0, 54.8, 53.0. IR (KBr) ν/cm^{-1} : 3310, 3144, 3080, 2961, 1720, 1599, 1473, 1437, 1425, 1363, 1317, 1288, 1250, 1234, 1213, 1198, 1151, 1122, 1064, 1024, 1010, 989, 924, 891, 835, 821, 785, 767, 744, 734. HR-ESI-MS calculated for $C_{12}H_{12}N_4O_5Na$ [M + Na]⁺: 315.0705, found: 315.0696.

4-(4-Phenyl)-1*H*-1,2,3-triazol-1-yl)pyridine-2,6-dicarboxylic acid (8a)

White solid. Mp: >290 °C. ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 9.82 (1H, s), 8.76 (2H, s), 7.99 (2H, d, J = 8 Hz), 7.52 (2H, d, J = 8 Hz), 7.41 (1H, t, J = 8 Hz). ¹³C NMR: compound not soluble enough in common NMR solvents. IR (KBr) ν/cm^{-1} : 3144, 3062, 1728, 1647, 1603, 1480, 1450, 1359, 1279, 1238, 1216, 1202, 1170, 1128, 1050, 1011, 907, 762, 690. HR-ESI-MS calculated for C₁₄H₉N₄O₂ [M - CO₂]⁺: 265.0726, found: 265.0717.

4-[4-(4-Methoxyphenyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6dicarboxylic acid (8b)

Pale yellow solid. Mp: 286 °C. ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 9.73 (1H, s), 8.76 (2H, s), 7.92 (2H, d, J = 9 Hz), 7.08 (2H, d, J = 9 Hz), 3.82 (3H, s). ¹³C NMR: compound not soluble enough in common NMR solvents. IR (KBr) ν/cm^{-1} : 3140, 3080, 2359, 1727, 1646, 1603, 1481, 1359, 1306, 1253, 1213, 1170, 1128, 1052, 1016, 906, 798. HR-ESI-MS calculated for C₁₅H₁₁N₄O₃ [M – CO₂]⁺: 295.0831, found: 295.0842.

4-[4-(4-Nitrophenyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6-dicarboxylic acid (8c)

Pale yellow solid. Mp: 272 °C (dec.). ¹H NMR (400 MHz, DMSOd₆) δ (ppm): 10.09 (1H, s), 8.28 (2H, s), 8.42 (2H, d, J = 9 Hz), 8.25 (2H, d, J = 9 Hz). ¹³C NMR: compound not soluble enough in common NMR solvents. IR (KBr) ν/cm^{-1} : 3581, 3504, 3098, 1710, 1596, 1504, 1476, 1434, 1339, 1275, 1242, 1106, 1048, 1016, 910, 872, 783. HR-ESI-MS calculated for C₁₄H₈N₅O₄ [M – CO₂]⁺: 310.0576, found: 310.0572.

4-[4-(Hydroxymethyl)-1*H*-1,2,3-triazol-1-yl]pyridine-2,6-dicarboxylic acid (8d)

White solid. Mp: 225 °C. ¹H NMR (400 MHz, D_2O) δ (ppm): 9.20 (1H, s), 8.73 (2H, s), 4.63 (2H, s). ¹³C NMR (400 MHz, D_2O + NaOH): 171.2, 155.1, 150.7, 144.8, 121.4, 115.3, 54.9. IR (KBr) ν/cm^{-1} : 3575, 3160, 1726, 1599, 1482, 1430, 1149, 1230, 1166, 1048, 1007, 964, 904, 823, 759, 723, 677. HR-ESI-MS calculated for $C_9H_7N_4O_3$ [M – CO_2]⁺: 219.0518, found: 219.0524.

Preparation of lanthanide complexes 9-10

To a suspension of 0.2 mmol (3 equivalents) of diacid hydrochloride **4b** or **8a–d** (prepared as described above) in 5 mL of water is added, under stirring, 4.5 equivalents of guanidinium carbonate. To the resulting solution is added 1 equivalent of europium(III) chloride hexahydrate or 1 equivalent of terbium(III) nitrate hexahydrate. The resulting complex **9** or **10** precipitates almost immediately. After 1 h of stirring, the lanthanide complex is isolated by filtration, washed with water and finally dried *in vacuo*. LnL₃ stoichiometry of complexes prepared was checked through elemental analysis of **10a**, calculated (%) for C₄₈H₄₂N₂₁O₁₂Tb·6H₂O: C 42.02, N 21.44, Tb 11.58; found C 42.10, N 21.52, Tb 11.78.

Complex **9d**: ¹H NMR (500 MHz, D₂O) δ (ppm): 7.44 (1H, br s), 4.37 (2H, br s), last signal (2 aromatic H) masked by solvent residual peak. Complex **10d**: ¹H NMR (500 MHz, D₂O) δ (ppm): 41.45 (2H, br s), 24.38 (1H, br s), 10.94 (2H, br s). Other complexes were insoluble in common NMR solvents.

Synthesis and analytical data for compound 12

Triethylamine (50 µL, 0.35 mmol, 1.0 eq.) and 3-(triethoxysilyl)propyl isocyanate (130 µL, 0.53 mmol, 1.5 eq.) were added under argon to a solution of **7d** (102 mg, 0.35 mmol) in anhydrous CHCl₃ (18 mL). The mixture was heated under reflux for 24 h. After cooling to room temperature, the mixture was added dropwise into petroleum ether under stirring by glass rod. The resulting white precipitate was isolated by filtration, washed for 4 times with petroleum ether and dried *in vacuo* for 10 h, to give cleanly and quantitatively trialkoxysilane **12**. ¹H NMR (400 MHz, CDCl₃) δ (ppm): 8.72 (2H, s), 8.35 (1H, s), 5.29 (2H, s), 5.09 (1H, br s), 4.08 (6H, s), 3.80 (6H, q, J = 7 Hz), 3.20 (2H, q, J = 7 Hz), 1.63 (2H, m), 1.21 (9H, t, J = 7 Hz), 0.62 (2H, t, J = 7.5 Hz). This compound was immediately engaged in the next step, without further purification.

Coatings preparation

Coatings were prepared with Laurell WS-400-6NPP-LITE spincoater. The hybrid films were prepared from the solution of $Ln(12)_3$ (Ln = Eu, Tb) in ethanol. Typically, the detailed procedure for the spin-coating is as follows: compound 12 (152 mg, 0.28 mmol, 3 eq.) and $Ln(NO_3)_3 \cdot 6H_2O$ (0.094 mmol, 1 eq.) were dissolved, under argon and at room temperature, in 3 mL of anhydrous ethanol. After stirring for 10 h, the solution was carefully dropped into the middle of the clean glass substrate. The transparent films were obtained with a rotation speed of 5000 rpm for 2 min, and after drying at 80 °C in an oven for 2 h. Micrographs were recorded by means of a ZEISS Supra 55VP scanning electron microscope operating in high vacuum at 1 kV, using secondary electron detector (Everhart–Thornley detector). Thickness of coatings was measured by using a KLA-Tencor Alpha-Step IQ Profilometer.

Luminescence

The cw photoluminescence spectra were collected using, as excitation source, a 450 W xenon lamp continuous wave monochromatized (TRIAX 180 from Jobin-Yvon/Horriba) and analyzed by a TRIAX 550 Jobin-Yvon/Horriba monochromator equipped with either a R928 Hamamatsu photomultiplier or a nitrogencooled CCD camera as detector. Cw excitation and emission spectra were obtained by monitoring the detector response while scanning the appropriate monochromator. One Photon Excited (OPE) luminescence intensity dependence and decays were obtained by pulsed excitation using a Jobin-Yvon LA-04 Nitrogen Laser (337.1 nm, 8 ns pulse, 25 Hz) and analyzed through a Jobin-Yvon HR 1000 monochromator (focal length: 1 m, 1200 groove mm⁻¹ grating and band-pass of 8 Å mm⁻¹ slits). The detector was a R1104 Hamamatsu photomultiplier tube. Fluorescence decays were measured with a Lecroy 9310A - 400 MHz digital oscilloscope. The Two Photon Excited (TPE) luminescence measurements were performed using a pulsed dye laser (Continuum ND60) pumped by a Continuum Surelite I-SL10 doubled Nd:YAG laser (10 ns pulse, 0.1 cm⁻¹ band width, 10 Hz repetition rate). The dye laser was equipped with a hydrogen Raman-shifter cell. Mixing of Rhodamines 590 and 610 used as dye solution provides energy up to 4 mJ in the selected infrared region. The Stokes-1 Raman shifted beam (shift of 4155 cm⁻¹) is spatially isolated from the other Stokes and anti-Stokes beams generated in the H₂-Raman cell by two Pellin-Brocca prisms associated with an iris diaphragm. In addition a set of IR Schott filter RG 750 and green-red transmission coloured filters are used to blocks any parasitic laser radiation and to select the overall visible fluorescence of Tb³⁺ and Eu³⁺ embedded in the complexes. Thanks to the high efficiency of the TPE processes, the emitted photons are collected with the previous experimental set-up consisting of the HR-1000 monochromator and R1104 photomultiplier associated with an EG&G PAR Boxcar Averager Model 162/164 ensuring a good signal/noise ratio. The TPE excitation spectra were recorded from the whole spectral response of the dye and corrected for its intensity distribution. The emission intensity dependence on the excitation beam power is analyzed by interposition of a set of neutral density filters (transmission varying between 2% and 100%) on the optical pathway. Moreover, using LDS 690 as the dye solution allows tuning the laser excitation in the 0.9-1.1 µm IR range, by using the Stokes-1 Raman shifted beam, which is consistent with a Three Photon Excitation (THPE) induced luminescence process. Images of the luminescent films are obtained with a digital camera after excitation provided by the 350 nm selected-radiation of a Hg lamp.

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