Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry

SUPPORTING INFORMATION

Gadolinium-binding cyclic hexapeptoids: synthesis and relaxometric properties

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List of abbreviations

ACN: acetonitrile

DCM: dichloromethane

DIC: *N*,*N*'-diisopropylcarbodiimide

DIPEA: ethyldiisopropylamine

DMF: *N*,*N*'-dimethylformamide

Fmoc: 9-fluorenylmethoxycarbonyl

HATU: *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate

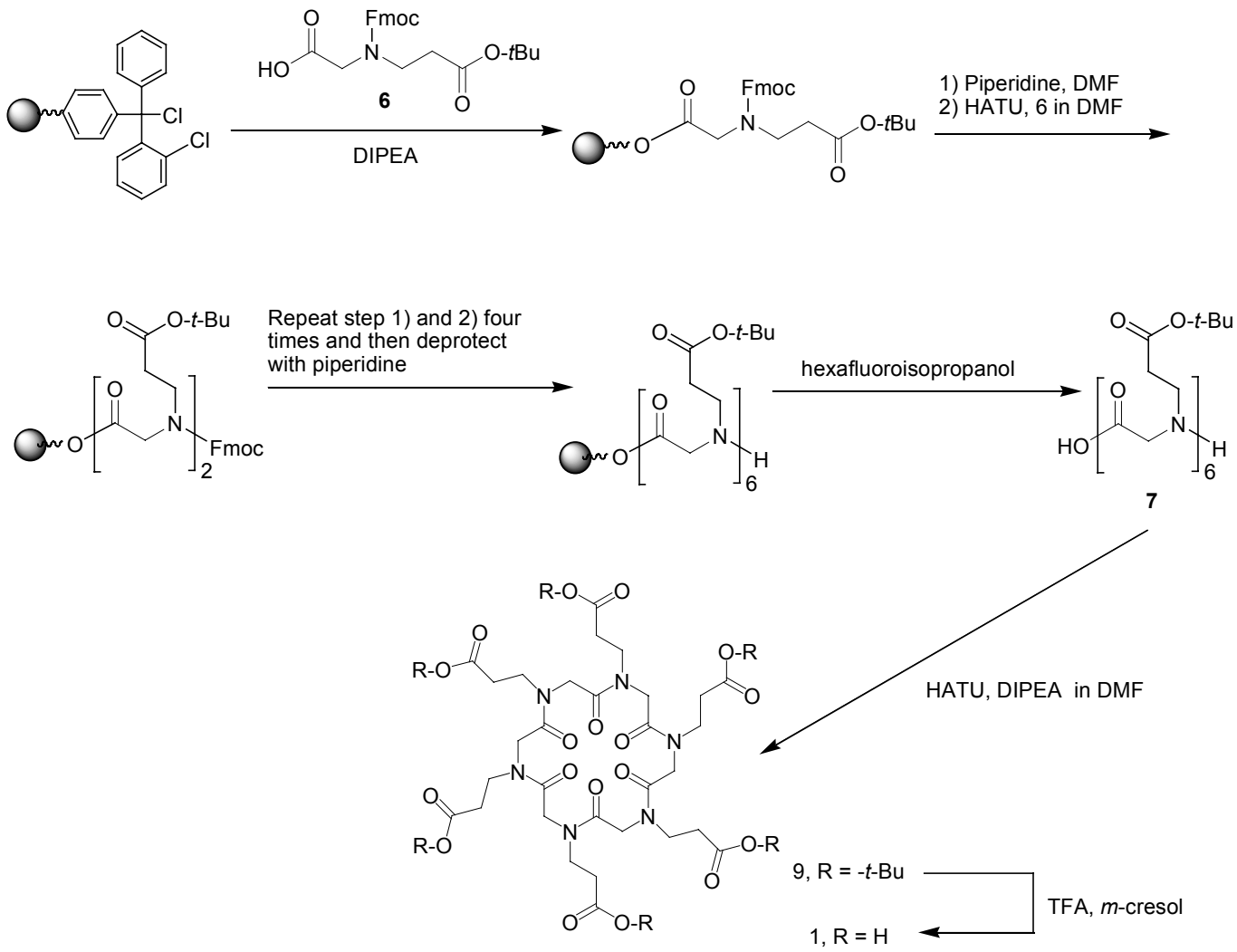
HFIP: hexafluoroisopropanol

RP HPLC: reversed-phase high-performance liquid chromatography

TFA: trifluoroacetic acid

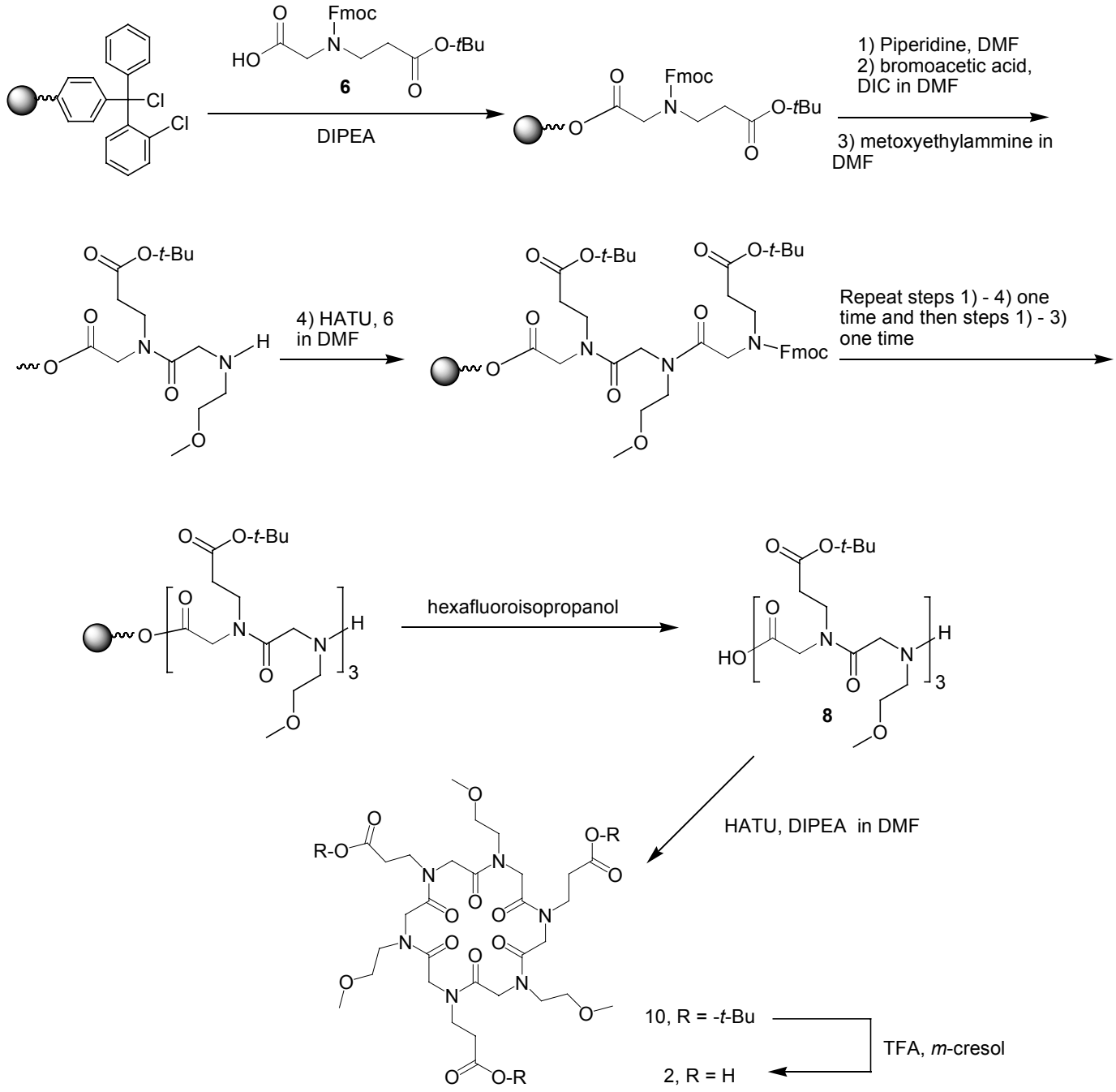
1.0 Synthetic schemes for the solid-phase oligomerization of 1 and 2

1.1 Solid-phase synthesis of 1 (Scheme 1S).



Scheme 1S. Solid-phase synthesis of cyclic peptoid **1**. See experimental section in the paper for details.

1.2 Solid-phase synthesis of 2 (Scheme 2S)



Scheme 2S. Solid-phase synthesis of cyclic peptoid **2**. See experimental section in the paper for details.

1.0 Complexation of cyclic peptoids in the presence of sodium picrate

1.1 Complexation of 9 in the presence of sodium picrate

In an NMR tube, to a 4.0 mM solution of **9** in CD_3CN:$CDCl_3$ 9:1 (0.5 mL), 6.0 equivalents of sodium picrate were added (3.0 mg, 12.0 μmol). After the addition the suspension was vigorously stirred for 5 minutes and the ^{1}H NMR spectrum was recorded.

Na-9: (400.13 MHz, CD_3CN:$CDCl_3$ 9:1, 4.0 mM solution) δ 1.44 (54H, s, C(CH_3)$_3$), 2.56 (12H, t, *J* 6.7 Hz, CH_2CH_2CO), 3.46 (6H, dt, *J* 15.2, 6.7 Hz, $CHHCH_2CO$), 3.64 (6H, dt, *J* 15.2, 6.7 Hz, $CHHCH_2CO$), 3.92 (6H, d, *J* 16.8 Hz, -OCCHHN), 4.61 (6H, d, *J* 16.8 Hz, -OCCHHN), 8.77 (~6H, s, picrate).

^{13}C NMR (100.03 MHz, CD_3CN:$CDCl_3$ 9:1) δ 28.5, 34.8, 45.5, 50.2, 82.3, 127.2 (picrate), 128.2 (picrate), 143.1 (picrate), 163.1 (picrate), 170.1, 171.7.

MS (ES) $[M+Na]^+$, *m*/*z* 1134.

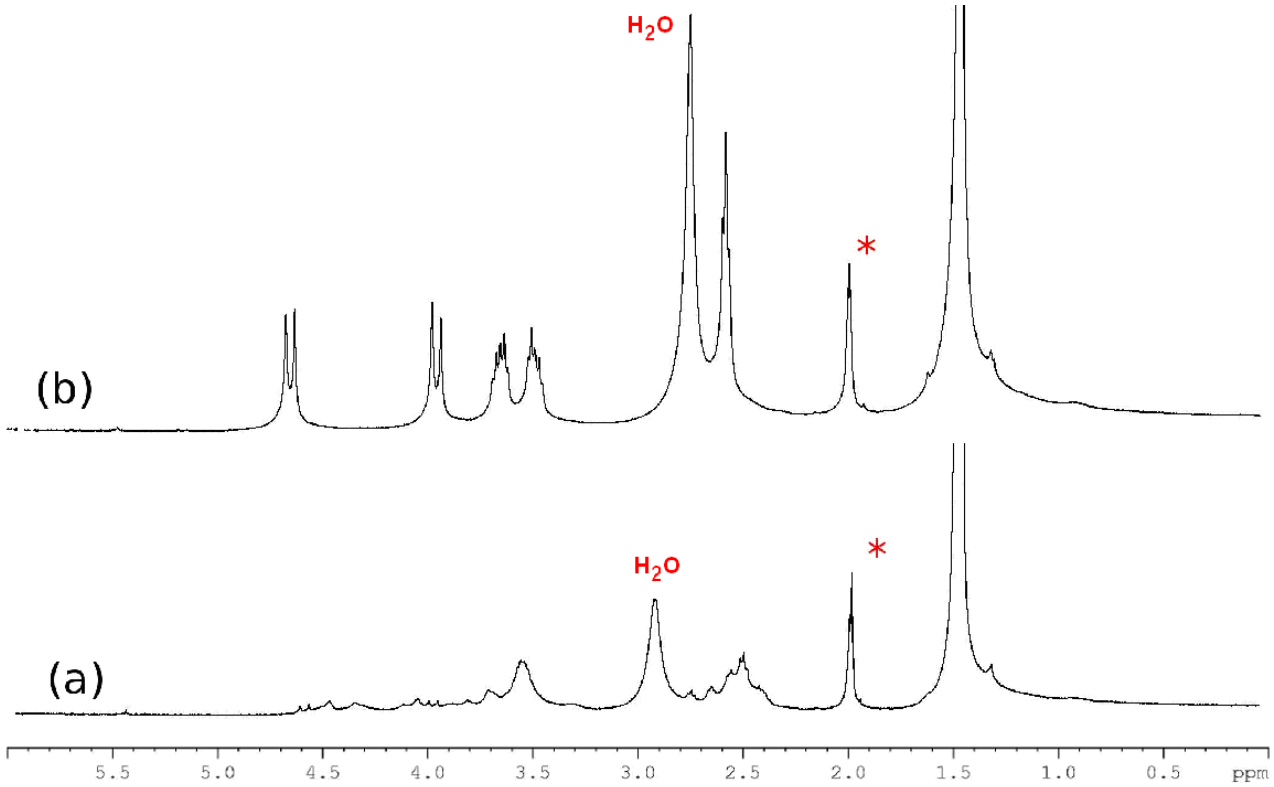


Fig. 1S ^{1}H NMR spectra of free **9** (a) (CD_3CN/$CDCl_3$ 9:1 solution, [**9**] = 4.0 mM, 400.13 MHz) and (b) in the presence of 6.0 eq. of sodium picrate. Residual solvent peaks are labelled with *.

1.2 Complexation of 10 in the presence of sodium picrate

In an NMR tube, to a 4.0 mM solution of **10** in CD_3CN:$CDCl_3$ 9:1 (0.5 mL), 6.0 equivalents of sodium picrate were added (3.0 mg, 12.0 μmol). After the addition the suspension was vigorously stirred for 5 minutes and the ^{1}H NMR spectrum was recorded.

Na-10: (400.13 MHz, CD_3CN:$CDCl_3$ 9:1, 4.0 mM solution) δ 1.44 (27H, s, $C(C\underline{H}_3)_3$), 2.54 (6H, t, *J* 6.0 Hz, $CH_2C\underline{H}_2COO$*t*-Bu), 3.32 (9H, s, $CH_2CH_2OC\underline{H}_3$), 3.47-3.70 (18H, m, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2C\underline{H}_2OCH_3$), 3.81 (3H, d, *J* 17.0 Hz, -OCC<u>H</u>HN), 3.89 (3H, d, *J* 17.0 Hz, -OCC<u>H</u>HN), 4.64 (3H, d, *J* 17.0 Hz, -OCCH<u>H</u>N), 4.68 (3H, d, *J* 17.0 Hz, -OCCH<u>H</u>N), 8.74 (~6H, s, picrate).

^{13}C NMR (75.5 MHz, CD_3CN:$CDCl_3$ 9:1) δ 28.7, 35.2, 45.8, 50.1, 50.6, 50.9, 59.7, 71.4, 82.1, 127.4 (picrate), 128.4 (picrate), 143.0 (picrate), 163.1 (picrate), 170.4, 171.0, 172.1.

MS (ES) $[M+Na]^+$, *m*/*z* 923.

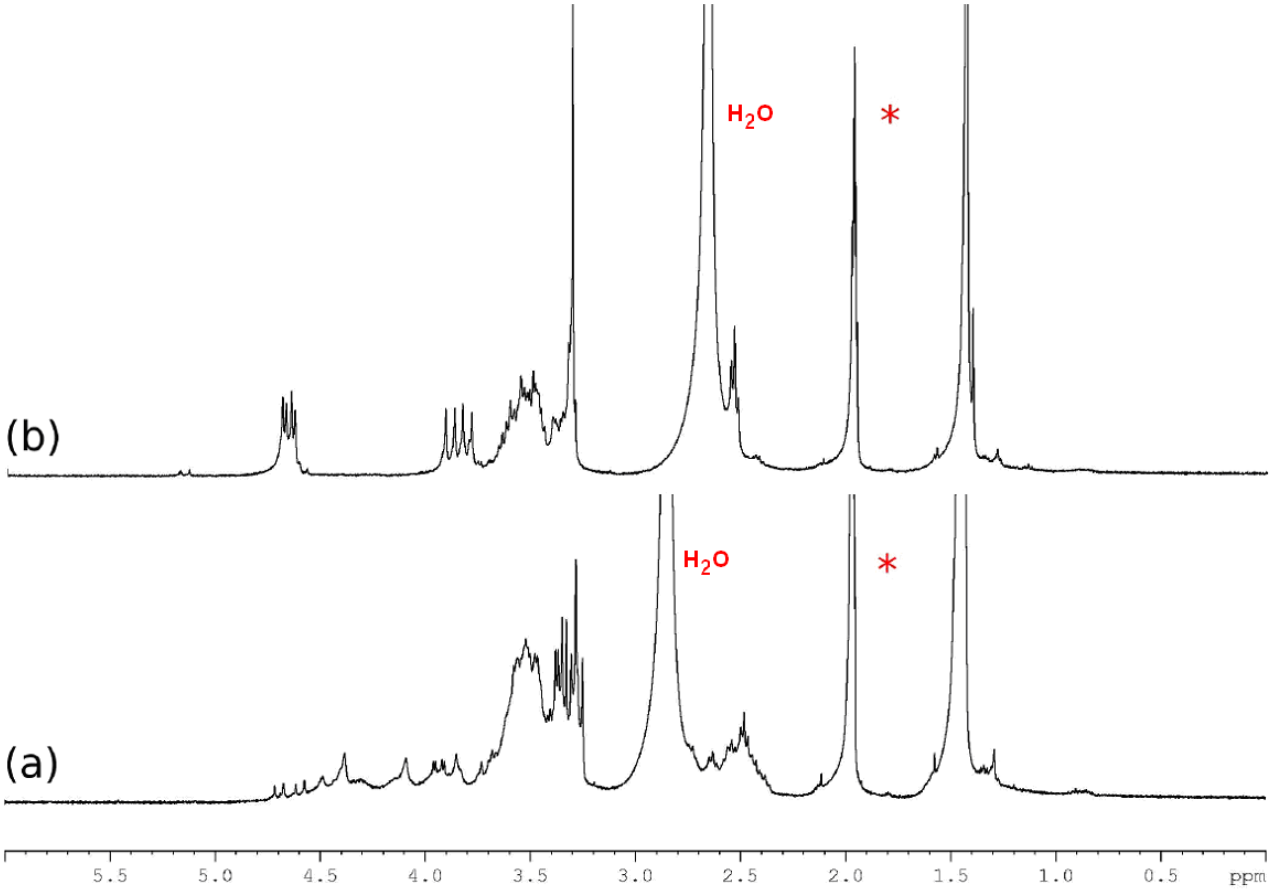


Fig. 2S ^1H NMR spectra of free **10** (a) ($CD_3CN/CDCl_3$ 9:1 solution, [**10**] = 4.0 mM, 400.13 MHz) and (b) in the presence of 6.0 eq. of sodium picrate. Residual solvent peaks are labelled with *.

1.3 Complexation of 1 in the presence of sodium picrate

In an NMR tube, to a 2.0 mM solution of **1** in CD_3CN/D_2O/MeOD (6:2:3, 0.5 mL), 6.0 equivalents of sodium picrate were added (1.5 mg, 6.0 μmol). After the addition the suspension was vigorously stirred for 5 minutes and the ^1H NMR spectrum was recorded.

Na-1: (400.13 MHz, CD_3CN/D_2O/MeOD 6:2:3), δ 2.60 (12H, m, $CH_2C\underline{H}_2COOH$), 3.40-3.60 (12H, m, $C\underline{H}_2CH_2COOH$, overlapped with HOD/CD_3OH signal), 3.85 (6H, d, *J* 17.0 Hz, -OCC<u>H</u>HN), 4.72 (6H, d, *J* 17.0 Hz, -OCCH<u>H</u>N), 8.68 (~6H, s, picrate).

MS (ES) $[M+Na]^+$, *m*/*z* 797.

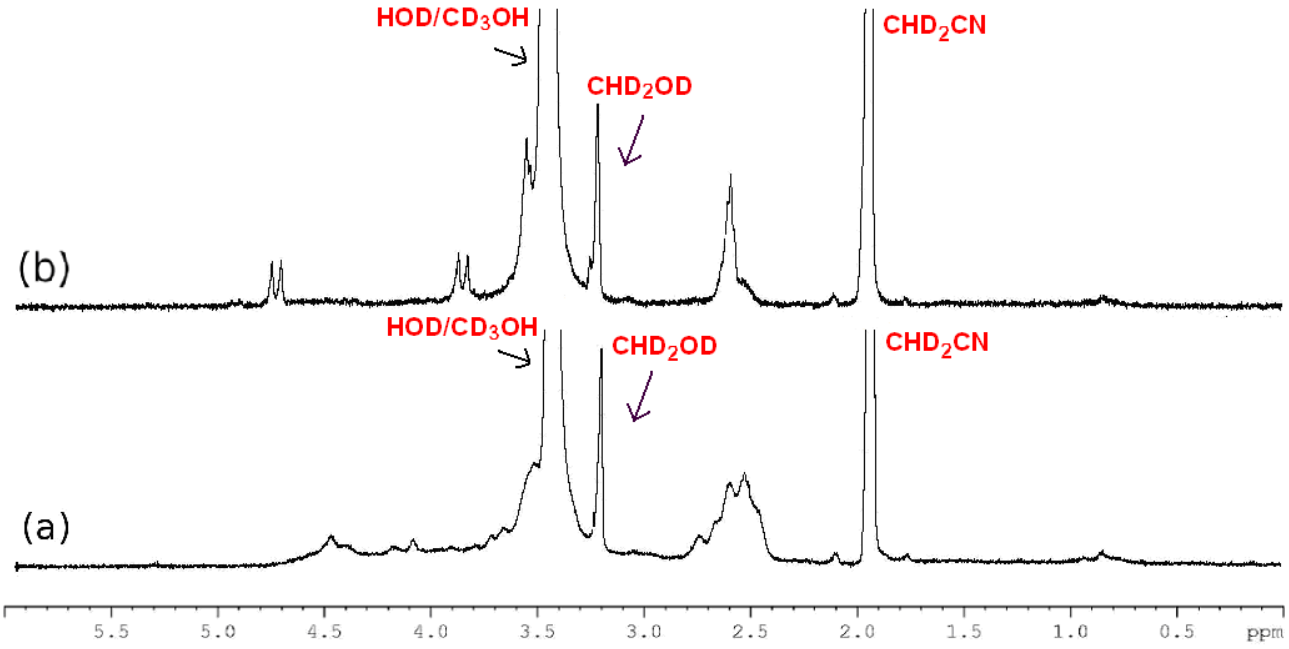


Fig. 3S ^{1}H NMR spectra of free **1** (a) ($CD_3CN/D_2O/CD_3OD$ 6:2:3 solution, [**1**] = 2.0 mM, 400.13 MHz) and (b) in the presence of 6.0 eq. of sodium picrate.

1.4 Complexation of 2 in the presence of sodium picrate

In an NMR tube, to a 2.0 mM solution of **2** in $CD_3CN:CDCl_3$ 9:1 (0.5 mL), 6.0 equivalents of sodium picrate were added (1.5 mg, 6.0 μmol). After the addition the suspension was vigorously stirred for 5 minutes and the ^{1}H NMR spectrum was recorded.

Na-2: (300.1 MHz, $CD_3CN:CDCl_3$ 9:1) δ 2.61 (6H, br s, $CH_2C\underline{H}_2COOH$, overlapped with the water signal), 3.30 (9H, s, $CH_2CH_2OC\underline{H}_3$), 3.50-3.60 (18H, m, $C\underline{H}_2CH_2COOH$ and $C\underline{H}_2C\underline{H}_2OCH_3$), 3.77 (3H, d, *J* 18.0 Hz, -OCC<u>H</u>HN), 3.84 (3H, d, *J* 18.0 Hz, -OCC<u>H</u>HN), 4.64 (3H, d, *J* 18.0 Hz, -OCCH<u>H</u>N), 4.83 (3H, d, *J* 18.0 Hz, -OCCH<u>H</u>N), 8.68 (~6H, s, picrate).

MS (ES) $[M+Na]^+$, *m*/*z* 755.

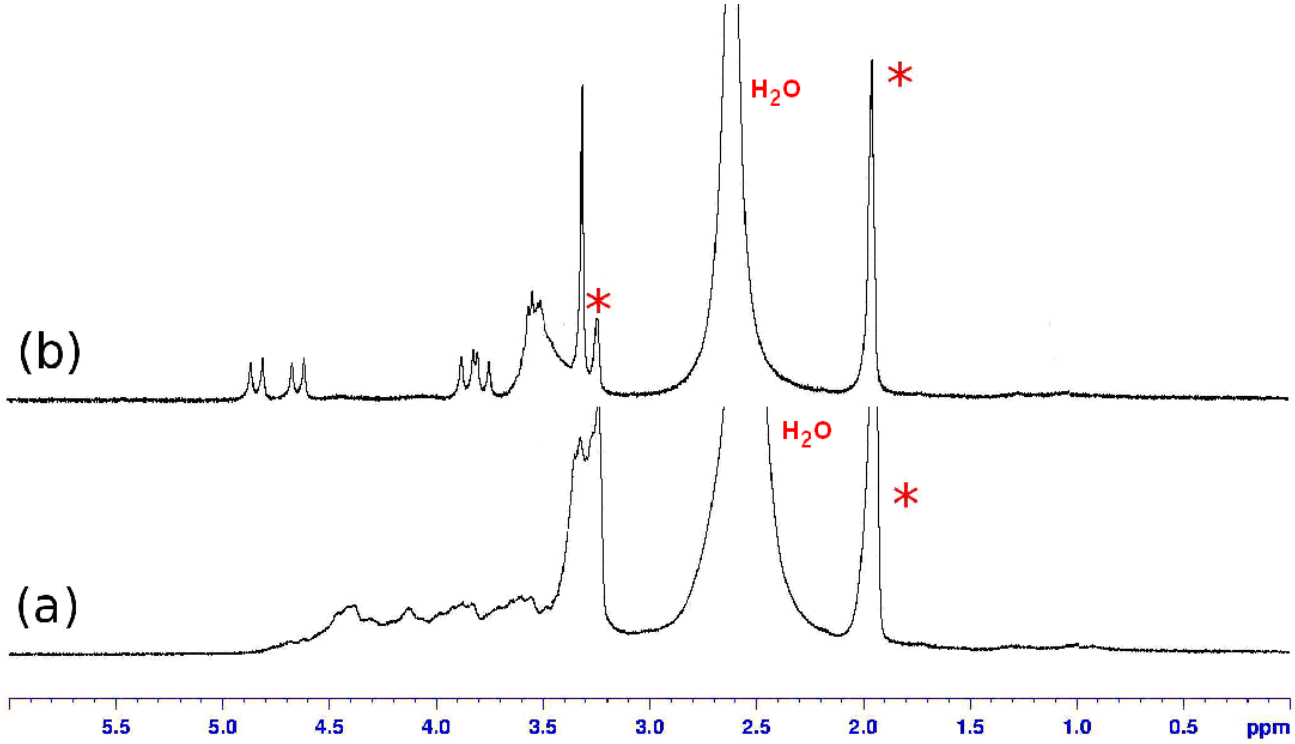


Fig. 4S ^{1}H NMR spectra of free **2** (a) ($CD_3CN/CDCl_3$ 9:1 solution, [**2**] = 2.0 mM, 400.13 MHz) and (b) in the presence of 6.0 eq. of sodium picrate. Residual solvent peaks and impurities (MeOH) are labelled with *.

2.0 Complexation of 1 and 2 with $GdCl_3$

Graphs reporting the complex formation measuring the water proton relaxation rate.

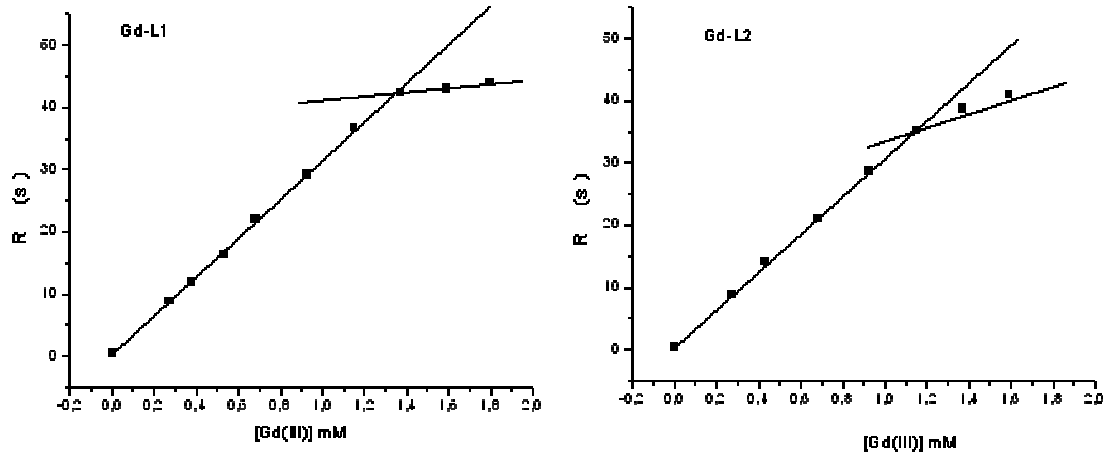


Fig. 5S Relaxometric titration of ligands **1** (left) and **2** (right) with Gd^{3+} ion (20 MHz, 25°C, neutral pH).

3.0 Relaxometric determination of the stability constants of Gd-1 and Gd-2

The observed longitudinal relaxation rates (at 20 MHz and 25°C) of **Gd-1** and **Gd-2** solutions, 0.673 mM and 0.954 mM respectively, were measured as a function of the addition of increasing concentrations of the competing ligand EDTA. The decrease in the observed relaxation rate is due to the transfer of the Gd^{3+} ion from ligand **1** (or **2**) to EDTA ligand. By knowing the stability constant of Gd-EDTA and the relaxivities of the Gd-complexes (**Gd-1** or **Gd-2** and Gd-EDTA) it is possible to fit these experimental data in order to extract the value of the unknown K_f of the investigated Gd-complexes according to the following equations:

The equilibrium is:

GdL1 + EDTA ↔ Gd-EDTA + L1 and the correspondent equilibrium constant: $K = \frac{K_B}{K_A}$

Where $K_A = \frac{[GdL1]}{[Gd][L1]}$ for the equilibrium: Gd + L1 ↔ Gd-L1

and $K_B = \frac{[GdEDTA]}{[Gd][EDTA]}$ for the equilibrium: Gd + EDTA ↔ Gd-EDTA

Given that:

$C_T(Gd) = \text{Total concentration of Gd} = [GdL1] + [GdEDTA]$
$C_T(EDTA) = \text{Total concentration of EDTA} = [EDTA] + [GdEDTA]$

And that the observed relaxation rate is given by:

$$R_{1obs} = r_{1p}^{GdL1}[GdL1] + r_{1p}^{GdEDTA}[GdEDTA] + 0.38$$

Combination of the previous equations gives:

$$R_{1obs} = r_{1p}^{GdL1}\left\{\frac{C_T(Gd) - \left[-(K_E C_T(EDTA) + K_E C_T(Gd)) + \sqrt{\left[(K_E C_T(EDTA) + K_E C_T(Gd))^2 + 4(K_A - K_E)(C_T(Gd)K_E C_T(EDTA))\right]}\right]}{(2(K_A - K_E))}\right\}$$
$$+ r_{1p}^{GdEDTA}\left\{\frac{C_T(Gd) - \left[-(K_E C_T(EDTA) + K_E C_T(Gd)) + \sqrt{\left[(K_E C_T(EDTA) + K_E C_T(Gd))^2 + 4(K_A - K_E)(C_T(Gd)K_E C_T(EDTA))\right]}\right]}{(2(K_A - K_E))}\right\} + 0.38$$