Experiment

Cleavage of dinucleotides

Dinucleotides (ApA, CpC, GpG, UpU) were purchased from Invitrogen. The 0.10 mM NpN and 0.05 mM [Tb(THP)]$(NO_3)_3 \cdot H_2O$ were dissolved in DEPC water. The reaction solution was incubated at 37 °C for 16 h, and then studied by ESI-MS.

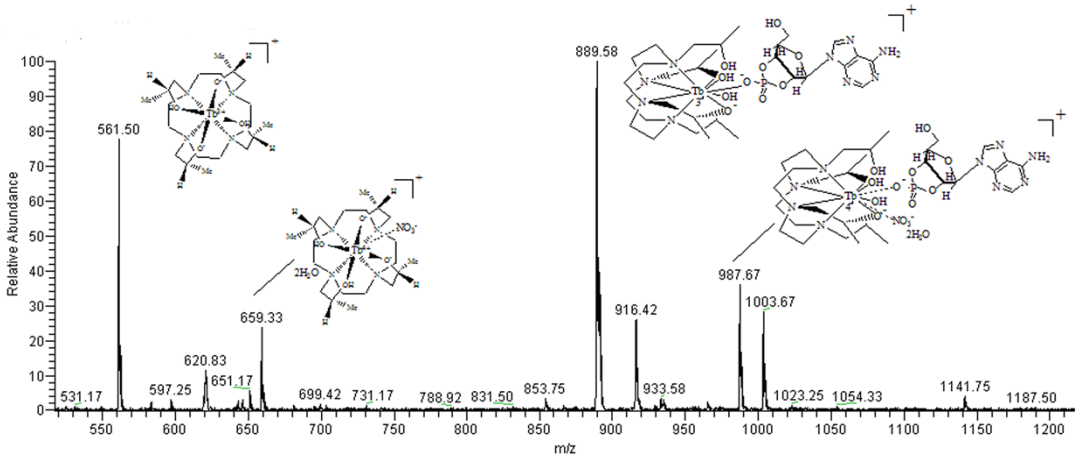


Fig.S1 ESI-MS spectrum of reaction of ApA and THPTb after 16 h.

Cleavage of RNA Oligonucleotides

The 5'-FAM end-labeled RNA oligonucleotides were purchased from Invitrogen. Cleavage experiments were carried out in a mixture (400 μL in total) containing 1 μM RNA, 50 mM MOPS buffer (pH 7.2, 25 °C), 100 mM $NaClO_4$, and various concentrations of [Tb(THP)]$(NO_3)_3 \cdot H_2O$]. The resulting solutions were incubated at 37 °C, from which 20 μL aliquots were periodically taken (up to 6 h), quenched with 20 μL of loading buffer (98% formamide, 2% 5×TBE and 0.05% w/v bromophenol blue), and cooled to -20 °C. The solutions were stored at this temperature until all samples were collected. Twenty microliters of these stored solutions were separated on 7 M urea/20% polyacrylamide gels. The developed gel was visualized by a Gel Doc XR (BioRad), and quantification analysis was performed with Quantity One software (version 4.6.2).

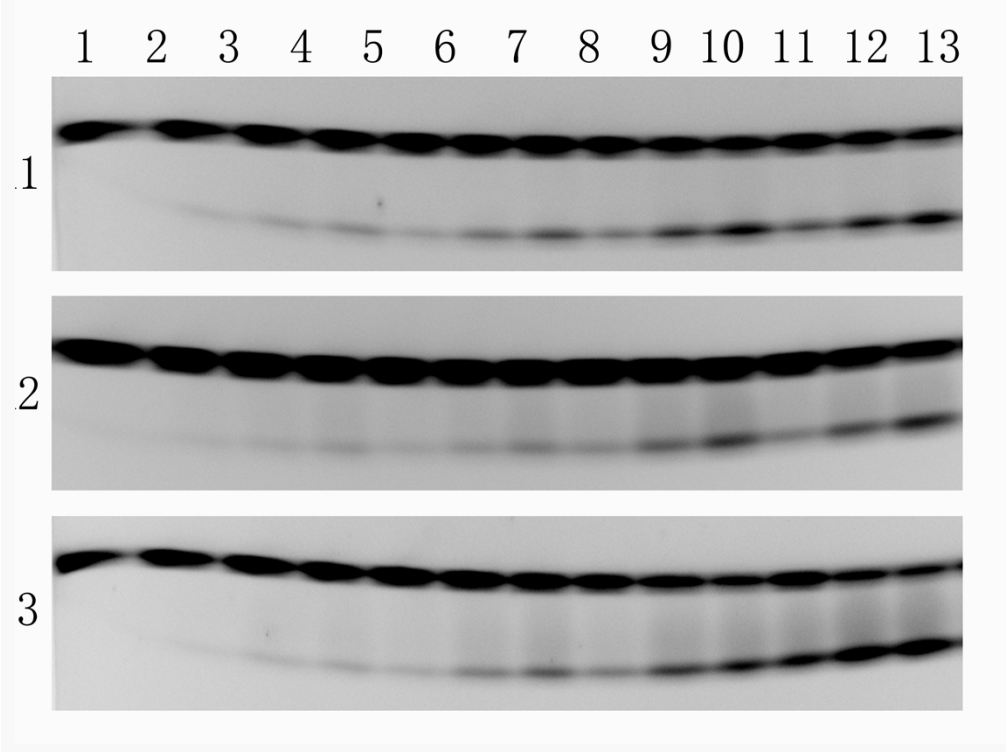


Fig.S2 Electrophoretograms of denaturing PAGE gel showing time and concentration dependences in the cleavage of oligonucleotides R1-3 R1(1) R2(2) R3(3) by THPTb. Lane 1: RNA only in DEPC-treated water, 0 h; lanes 2-4: 5 μM THPTb, 2, 4 and 6 h; lanes 5-7: 10 μM THPTb, 2, 4 and 6 h; lanes 8-10: 25 μM THPTb, 2, 4 and 6 h; lanes 11-13, 50 μM THPTb, 2, 4 and 6 h.

MALDI-TOF/TOF-MS

The 5'-FAM end-labeled RNA oligonucleotides were purchased from Invitrogen. Cleavage experiments were carried out as described in section, "Cleavage of RNA Oligonucleotides". Two hundred microliters aliquots were periodically taken from the reaction mixture at 0, 2, 4 h and dried in vacuum. The precipitates were then re-dissolved in 10 μL of DEPC water, loaded on a C18 resin column, and then desalted with 0.2% TFA. The results were collected on a MALDI-TOF/TOF mass spectrometer.

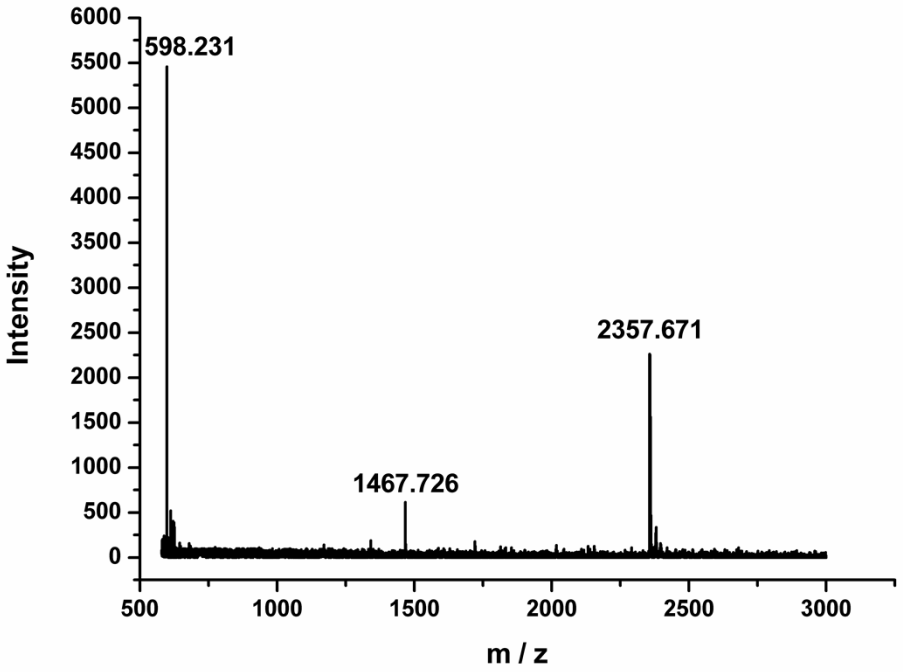


Fig.S3 Mass spectra of cleavage of the oligoribonucleotide R4 5'-(FAM)CCAAUC-3'. The peaks at *m/z* 598.231 and 1467.726 correspond to cleavage at ApU and ApA, respectively.

HPNP Cleavage Kinetics.

A solution (total reaction volume=2.5 mL) including 200 μM Tb(III) complex, 50 mM MOPS (pH 7.2, 25 °C), and 100 mM $NaClO_4$ was equilibrated at room temperature for 10 min in a quartz cuvette (1 cm). We then added 10 μL of 5 mM HPNP to give a 20 μM solution. The NP^- release was quantified by measuring absorbance at 400 nm using a first order modeling process, and then fitting the absorbance versus time data to the equation $Abs=A+Be^{-kobst}$, where A and B are constants.

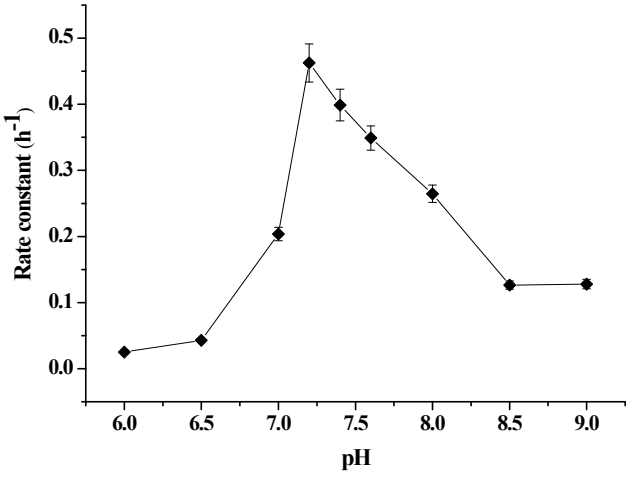


Fig.S4 The effect of pH on cleavage reactions of HPNP. The pH–rate profile was plotted for the hydrolysis of 20

μM HPNP at 25°C by 200 μM THPTb. 50 mM MES buffer for pH 6.0 and 6.5, 50 mM MOPS buffer for pH 7.0 and 7.2, 50 mM Tris buffer for pH 7.4, 7.6, 8.0, 8.5, and 9.0. The Data were presented as mean ± S.D. of three independent experiments.

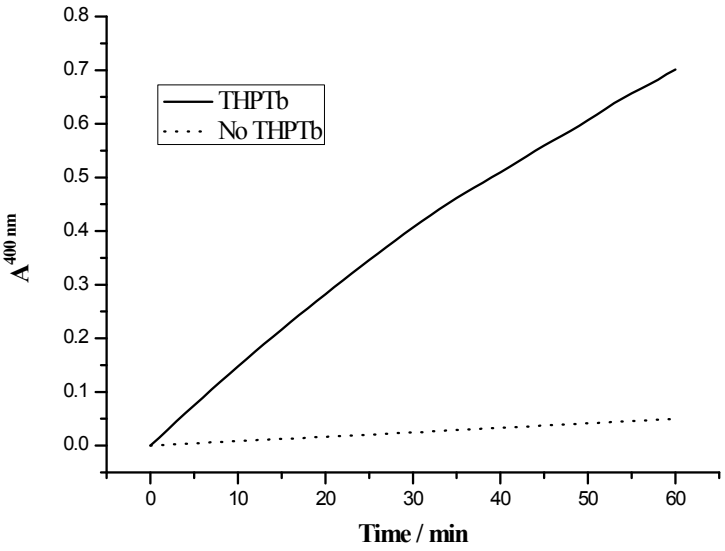
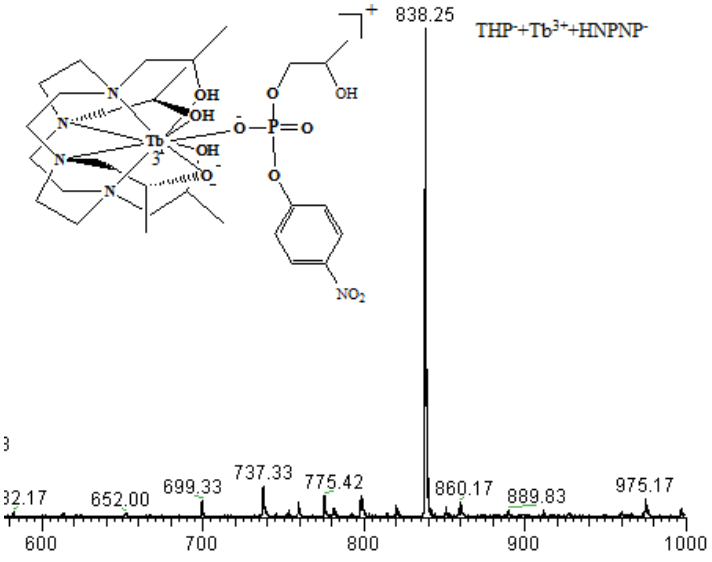
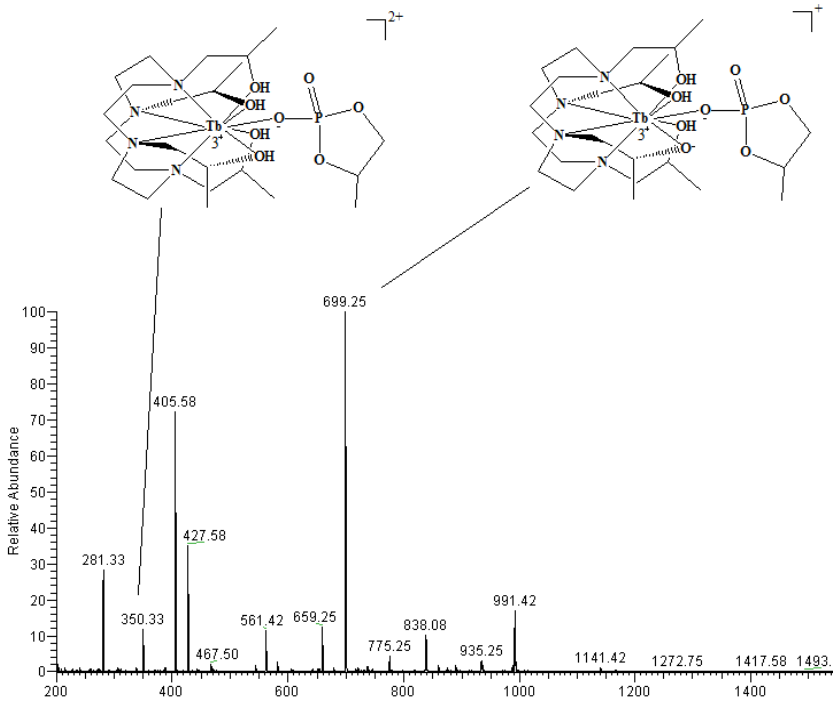


Fig.S5 Hydrolysis of 2-hydroxypropyl-p-nitrophenyl phosphate (HPNP=20 μM) with or without 200 μM THPTb.

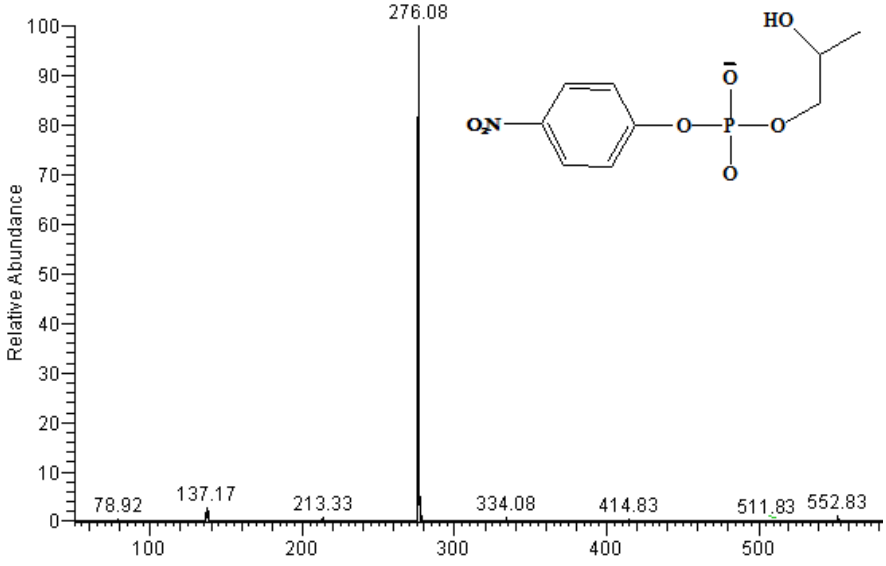
a



b


Relative Abundance
699.25
405.58
427.58
281.33
350.33
561.42
659.25
467.50
775.25
838.08
935.25
991.42
1141.42
1272.75
1417.58
1493.

c


Relative Abundance
276.08
78.92
137.17
213.33
334.08
414.83
511.83
552.83
HO
O2N

d

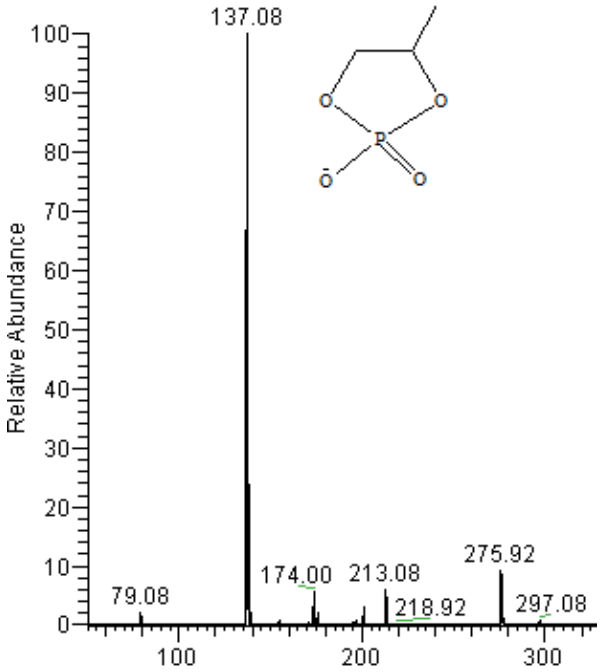


Fig.S6 ESI-MS of HPNP cleaved by THPTb. Panel a, b positive-ion mode spectra for 2 min, 24 h; Panel c, d negative-ion mode spectra for 2 min, 24 h..

X-Ray crystallographic studies

Syntheses

(1) 1,4,7,10-Tetrakis(2-hydroxypropyl)-cyclen (THP)

Cyclen (1 mmol, 0.17 g) was dissolved in triethylamine and ethanol at room temperature, to which was dropwise added 50% excess liquid epoxy isopropyl (~5 mL). The mixture was stirred at room temperature for 2 d and evaporated in vacuum as a rod-like white powder that were recrystallized in boiling n-hexane, filtered and dried as a white powder (0.16 g, yield: 40.0%). ESI-MS *m/z*: 405.42 $[M+1]^+$.

(2) 1,4,7,10-tetrakis(carbamoylmethyl)-cyclen (DOTAM)

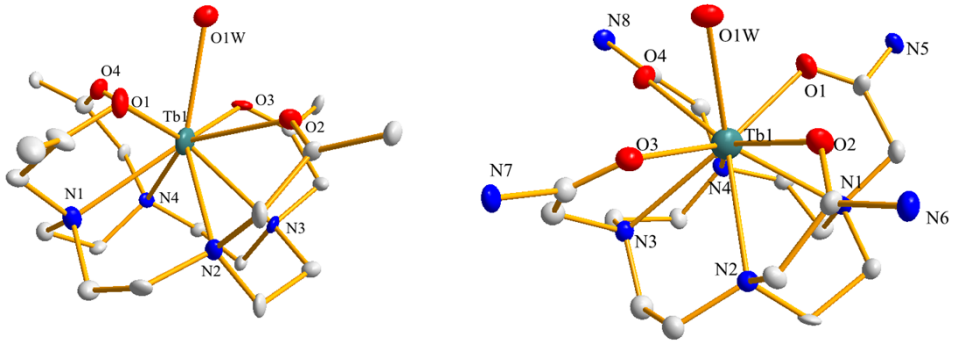
Cyclen (2.02 g, 0.012 mol), chloroacetamide (4.52 g, 0.048 mol) and triethylamine (5.01 g, 0.049 mol) were dissolved in 40.0 mL of ethanol and refluxed for 4 h. After cooling, the precipitated white powders were filtered, washed twice with 5.0 mL of ethanol, and dried in vacuum at 100 °C, yielding white powder (1.38 g, yield: 28.1%). ESI-MS *m/z*: 401.32 $[M+1]^+$.

(3) Preparation of $[Tb(THP)](NO_3)_3 \cdot H_2O$

The THP Ligand (0.041 g) and $Tb(NO_3)_2·6H_2O$ (0.045 g) (0.10 mmol, 1:1) were dissolved in methanol, magnetically stirred under reflux for 30 min and cooled, yielding white microcrystals (0.023 g, yield: 30.1%) that were then dissolved in 10.0 mL of CH_3OH. Colorless crystals were obtained after 1 week of slow ethyl ether diffusion (0.016 g, yield: 69.5%). ESI-MS m/z: 281.50(100%) $[Tb^{3+}+H_3L^{-}]^{2+}/2$, 561.42 (68%) $[THP^{2-}+Tb^{3+}]^{+}$, 659.17 (18%) $[Tb^{4+}+H_2L^{2-}+NO_3^{-}+2H_2O]^{+}$.

(4) Preparation of $[Tb(DOTAM)](NO_3)_3·H_2O$

The DOTAM Ligand (0.040 g) and $Tb(NO_3)_2·6H_2O$ (0.045 g) (0.10 mmol, 1:1) were dissolved in methanol, magnetically stirred while refluxing for 30 min and cooled until white microcrystals precipitated (0.040 g, yield: 53.2%). The products were then dissolved in 10.0 mL of CH_3OH with ethyl ether slowly diffusing, yielding colorless crystals after one week (0.018 g, yield: 45%). MALDI-TOF-MS *m/z*: 557.35 $[DOTAM^{2-}+Tb^{3+}]^{+}$.



(a) $[Tb(THP)](NO_3)_3·H_2O$ (b) $[Tb(DOTAM)](NO_3)_3·H_2O$

Fig.S7 Cationic structures

Table S1 Main bond lengths (Å) and bond angles (°) of $[Tb(THP)](NO_3)_3·H_2O$

O1–Tb1	2.356(5)	O2–Tb1	2.356(4)
O3–Tb1	2.356(4)	O4–Tb1	2.354(4)
O1W–Tb1	2.427(5)	N1–Tb1	2.653(6)
N2–Tb1	2.678(5)	N3–Tb1	2.614(5)
N4–Tb1	2.628(5)	O2–Tb1–O1	82.96(16)
O2–Tb1–O3	87.96(15)	O4–Tb1–O3	83.03(15)
O4–Tb1–O1	84.04(16)	N1–Tb1–N2	68.25(16)
N3–Tb1–N2	67.72(17)	N3–Tb1–N4	69.19(16)
N4–Tb1–N1	68.42(17)		

Table S2 Main bond lengths (Å) and bond angles (°) of $[Tb(DOTAM)](NO_3)_3 \cdot H_2O$

O1–Tb1	2.368(4)	O2–Tb1	2.363(4)
O3–Tb1	2.354(4)	O4–Tb1	2.358(4)
O1W–Tb1	2.451(4)	N1–Tb1	2.622(5)
N2–Tb1	2.644(5)	N3–Tb1	2.657(5)
N4–Tb1	2.622(5)	O2–Tb1–O1	82.80(15)
O3–Tb1–O2	87.26(14)	O3–Tb1–O4	83.82(15)
O4–Tb1–O1	84.40(15)	N1–Tb1–N2	68.39(15)
N2–Tb1–N3	68.32(15)	N4–Tb1–N3	67.54(14)
N1–Tb1–N4	68.95(14)		