Electronic Supplementary Information 8

Non-identical Electronic Characters of the Internucleotidic Phosphates in RNA Modulate the Chemical Reactivity of the Phosphodiester Bonds

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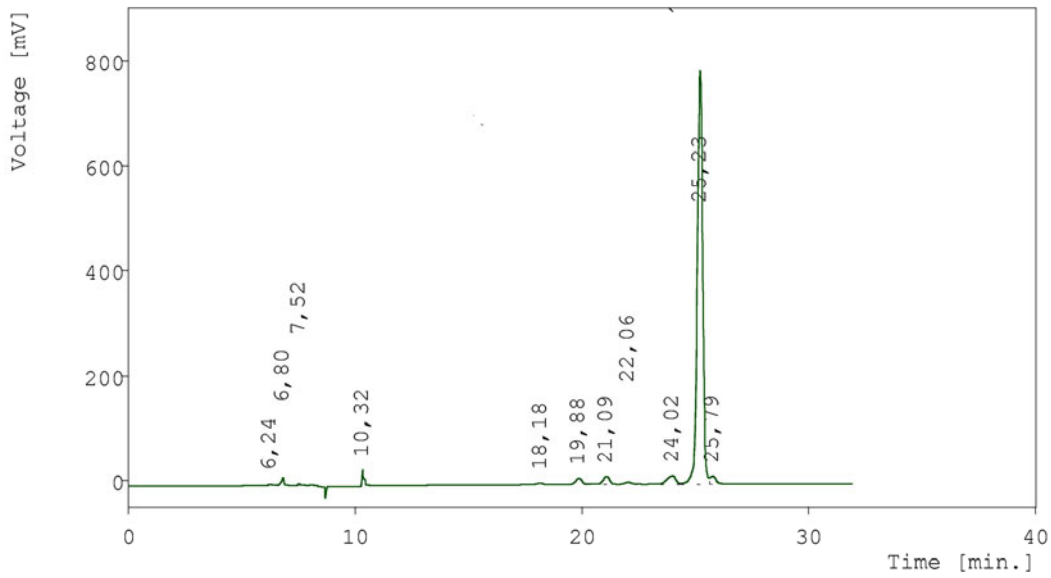
Content:

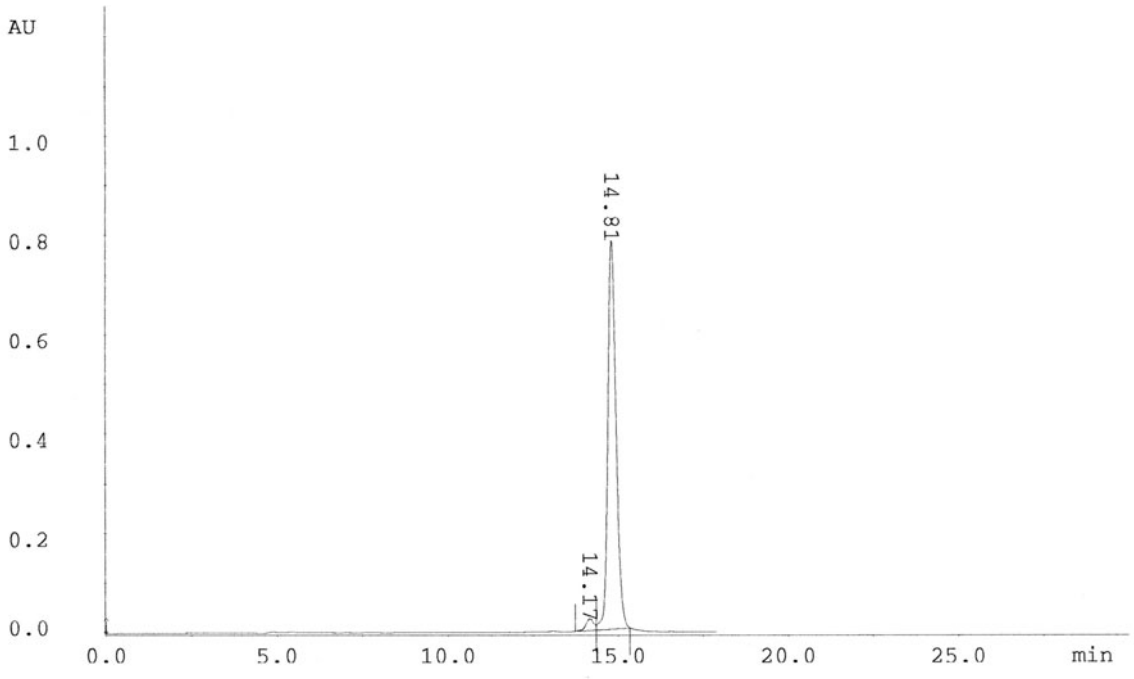
Figure S15D



Result Table - Calculation Method Uncal

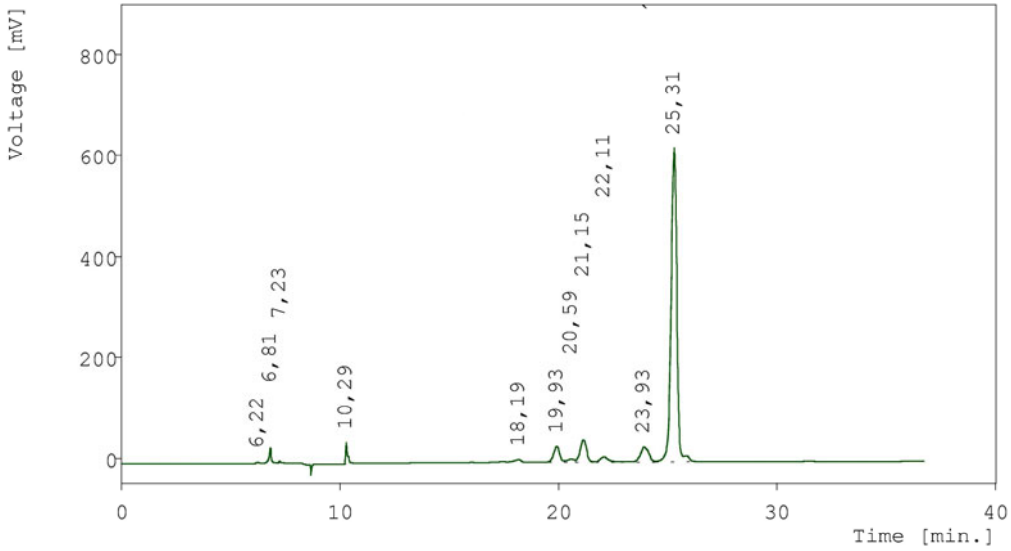
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,240	34,6353	2,379	0,300	0,233	0,263
2	6,800	148,8392	15,265	0,113	1,001	1,687
3	7,520	25,1110	3,668	0,087	0,169	0,405
4	10,320	231,6464	31,432	0,073	1,558	3,474
5	18,180	42,7957	2,533	0,273	0,288	0,280
6	19,880	200,7339	11,230	0,287	1,350	1,241
7	21,093	297,6109	14,256	0,313	2,002	1,576
8	22,060	79,1519	3,621	0,320	0,532	0,400
9	24,020	432,4915	15,658	0,427	2,909	1,731
10	25,233	13092,3190	790,019	0,260	88,054	87,324
11	25,787	283,1023	14,643	0,307	1,904	1,619
-	Total	14868,4371	904,702			

Figure S15(d1): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **0.5h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.17	13.75	14.36	0.62	6.2961	23.585
2	14.81	14.36	15.35	1.00	218.6636	783.298

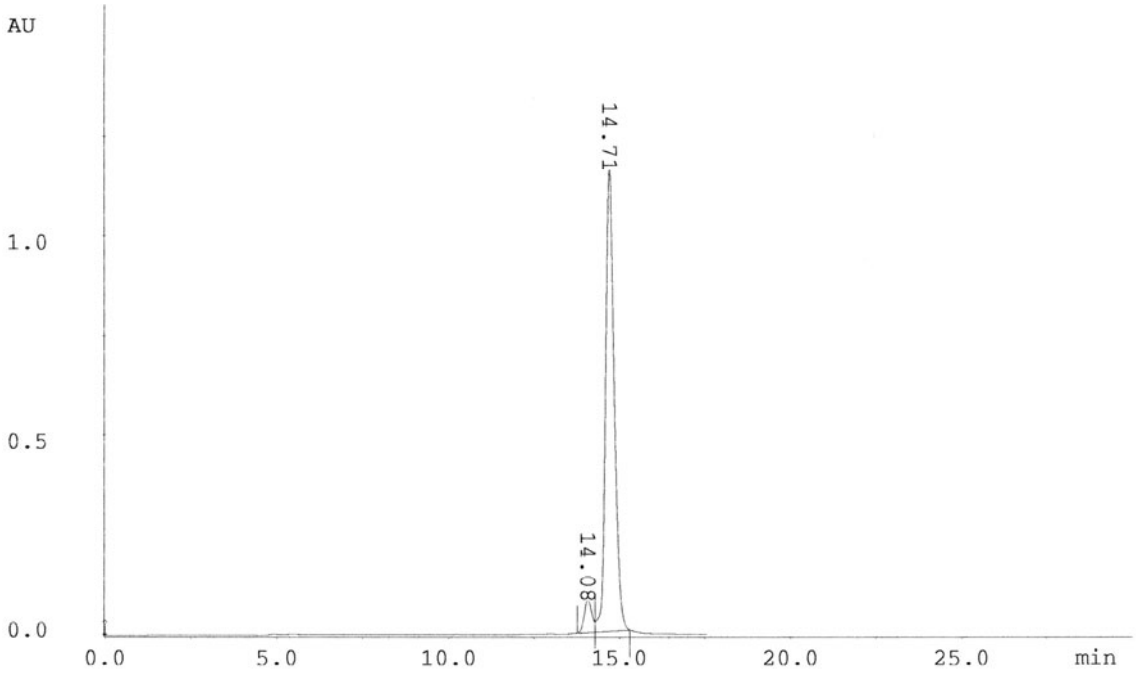
Figure S15(d1i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 25.23 min** in **Figure S15(d1)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.17 min from the heptamer peak eluting at R_T = 14.81 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

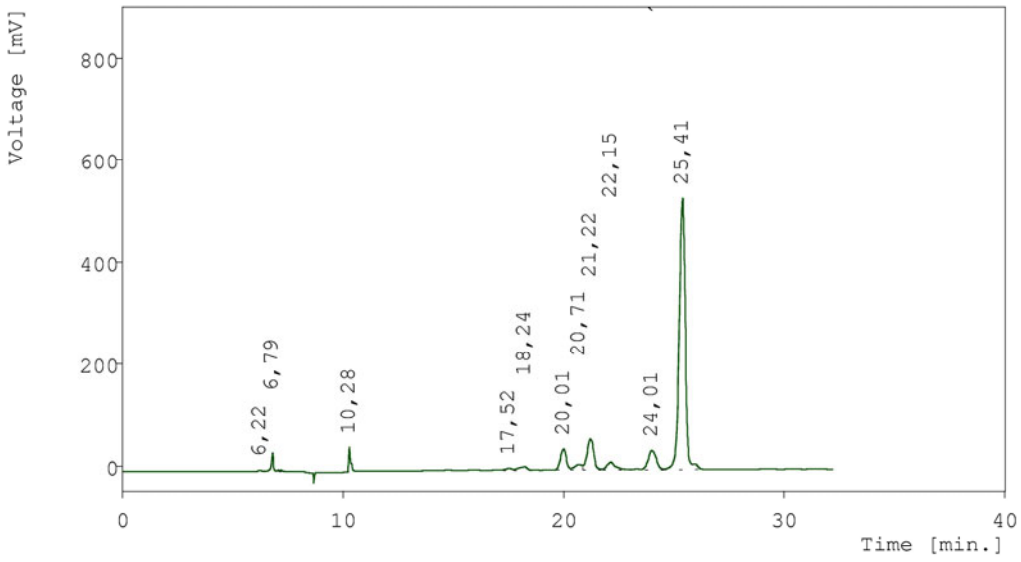
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,220	22,0697	2,178	0,173	0,148	0,264
2	6,807	227,7752	30,105	0,093	1,530	3,643
3	7,233	27,7786	3,792	0,087	0,187	0,459
4	10,287	289,7271	42,111	0,067	1,946	5,096
5	18,187	102,7126	5,091	0,333	0,690	0,616
6	19,933	593,1911	30,883	0,300	3,985	3,737
7	20,593	139,8911	5,994	0,427	0,940	0,725
8	21,147	910,3779	43,346	0,327	6,116	5,245
9	22,107	268,1316	10,531	0,373	1,801	1,274
10	23,927	807,7912	29,711	0,420	5,427	3,595
11	25,313	11495,9047	622,679	0,280	77,230	75,346
-	Total	14885,3509	826,421			

Figure S15(d2): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **2h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.08	13.78	14.30	0.53	21.0067	80.840
2	14.71	14.30	15.31	1.02	339.3989	1155.572

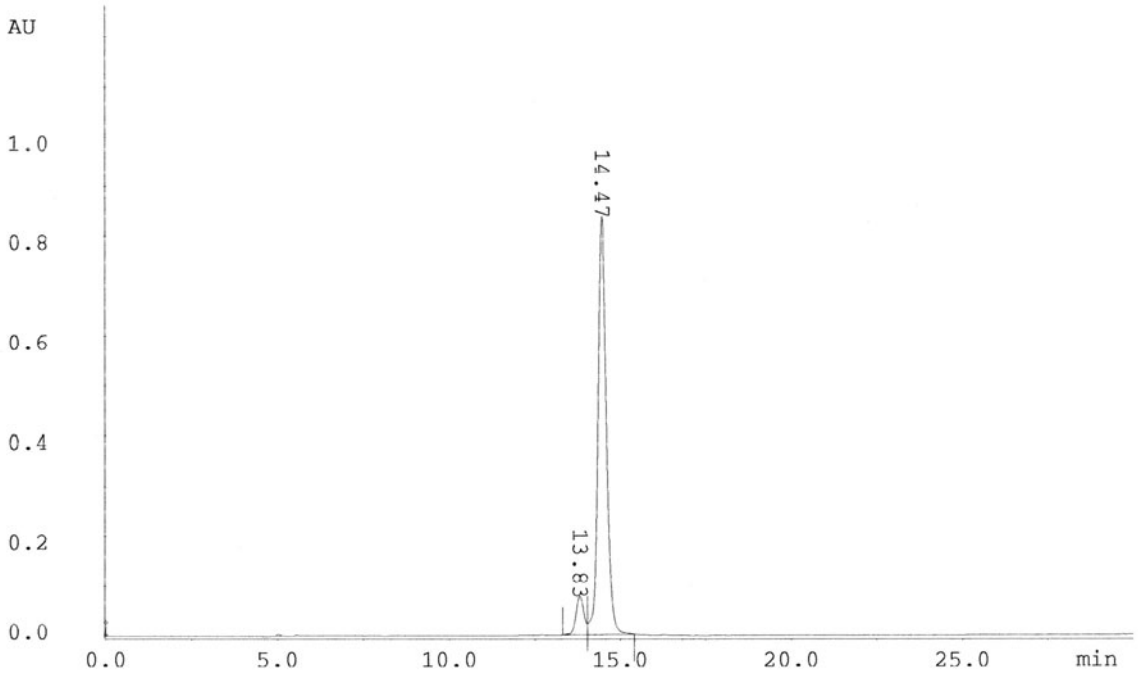
Figure S15(d2i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 25.31 min** in **Figure S15(d2)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.08 min from the heptamer peak eluting at R_T = 14.71 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

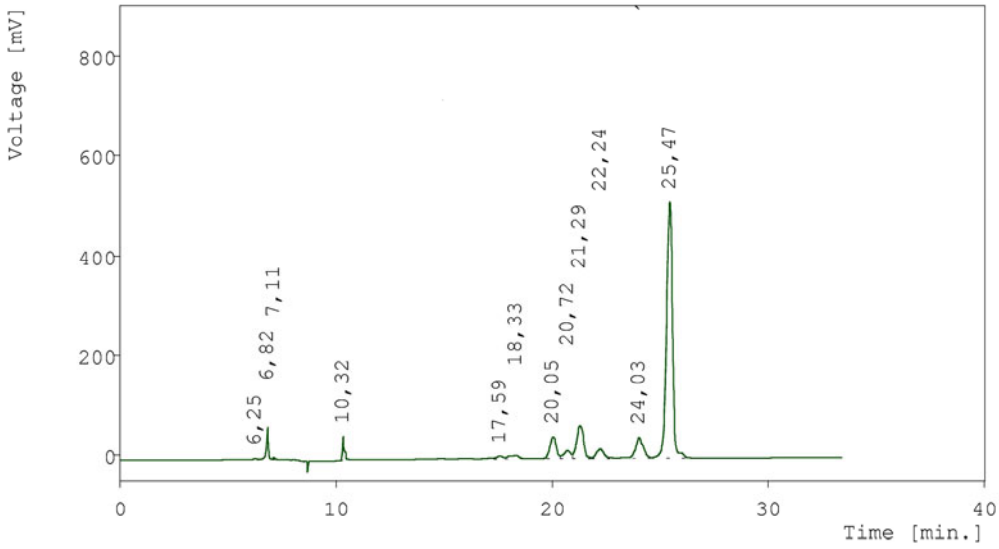
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,220	25,0516	2,299	0,180	0,167	0,288
2	6,793	272,5776	36,317	0,087	1,813	4,549
3	10,280	320,6310	49,144	0,060	2,132	6,155
4	17,520	103,9352	3,956	0,393	0,691	0,495
5	18,240	198,5188	6,966	0,467	1,320	0,872
6	20,013	773,0186	41,683	0,287	5,141	5,221
7	20,713	258,5709	10,933	0,400	1,720	1,369
8	21,220	1318,2367	60,889	0,333	8,767	7,626
9	22,153	415,0343	15,192	0,373	2,760	1,903
10	24,007	1003,1072	38,115	0,407	6,671	4,774
11	25,413	10347,9218	532,927	0,300	68,818	66,748
-	Total	15036,6038	798,421			

Figure S15(d3): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **3h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	13.83	13.32	14.05	0.75	20.1624	75.654
2	14.47	14.05	15.41	1.37	239.8648	836.393

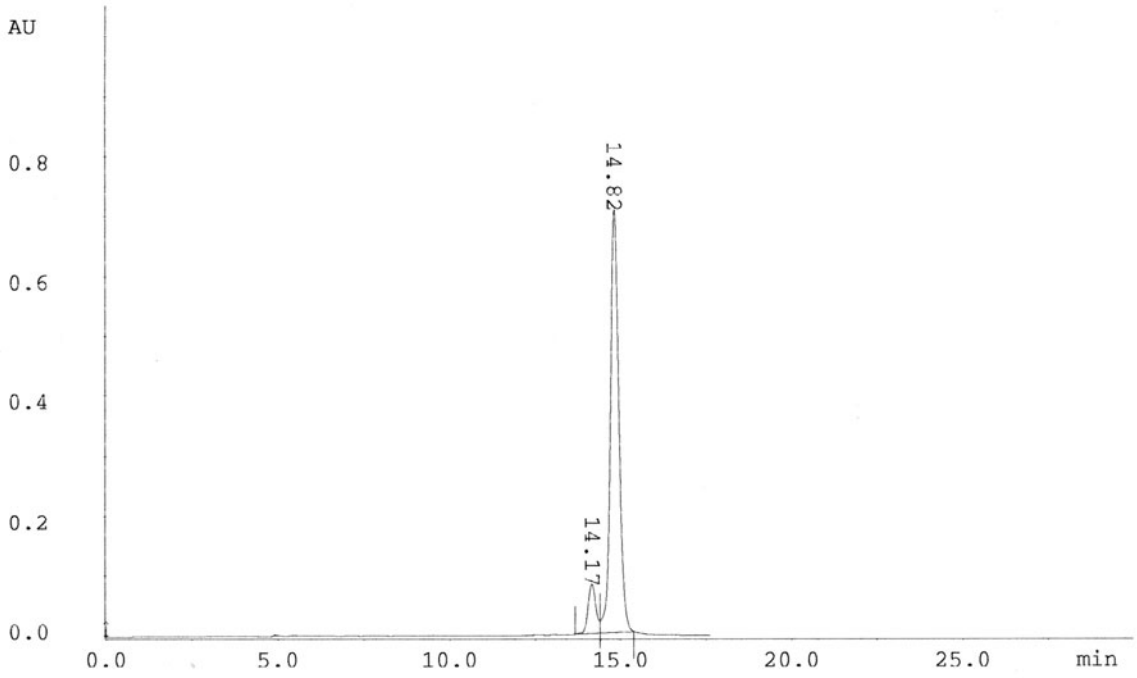
Figure S15(d3): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 25.41 min** in **Figure S15(d3i)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 13.83 min from the heptamer peak eluting at R_T = 14.47 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

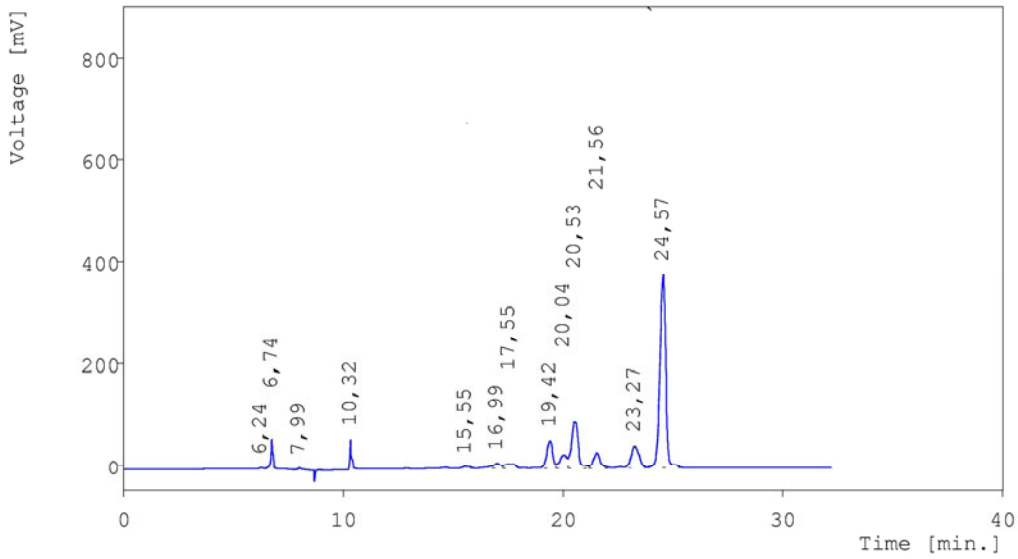
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,247	23,7498	2,233	0,187	0,152	0,266
2	6,820	412,2376	66,272	0,093	2,634	7,881
3	7,113	38,6585	4,881	0,100	0,247	0,580
4	10,320	344,0521	49,937	0,067	2,198	5,938
5	17,587	157,1746	5,851	0,360	1,004	0,696
6	18,333	248,3604	7,178	0,567	1,587	0,854
7	20,047	929,8444	45,040	0,313	5,940	5,356
8	20,720	366,8859	16,246	0,400	2,344	1,932
9	21,293	1527,5843	67,324	0,347	9,759	8,006
10	22,240	487,4480	19,143	0,393	3,114	2,276
11	24,033	1085,5755	43,154	0,373	6,935	5,132
12	25,467	10031,2120	513,658	0,300	64,086	61,083
-	Total	15652,7830	840,917			

Figure S15(d4): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **4h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.17	13.68	14.40	0.73	21.4810	81.122
2	14.82	14.40	15.39	1.00	201.7550	705.156

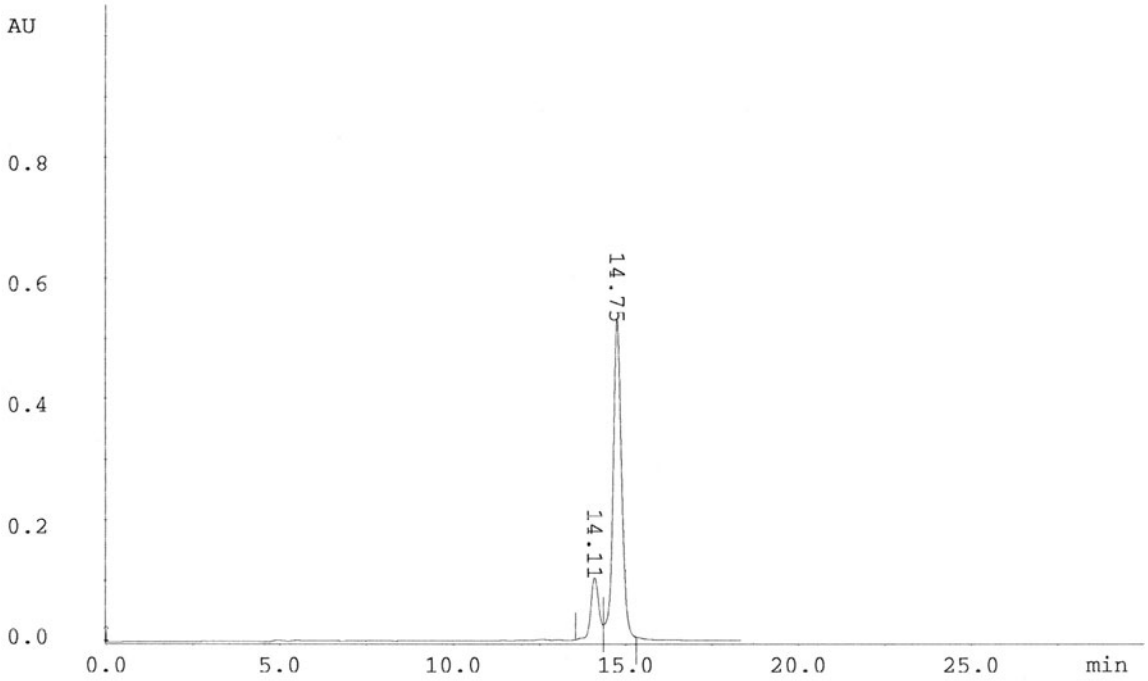
Figure S15(d4i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 25.47 min** in **Figure S15(d4)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.17 min from the heptamer peak eluting at R_T = 14.82 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

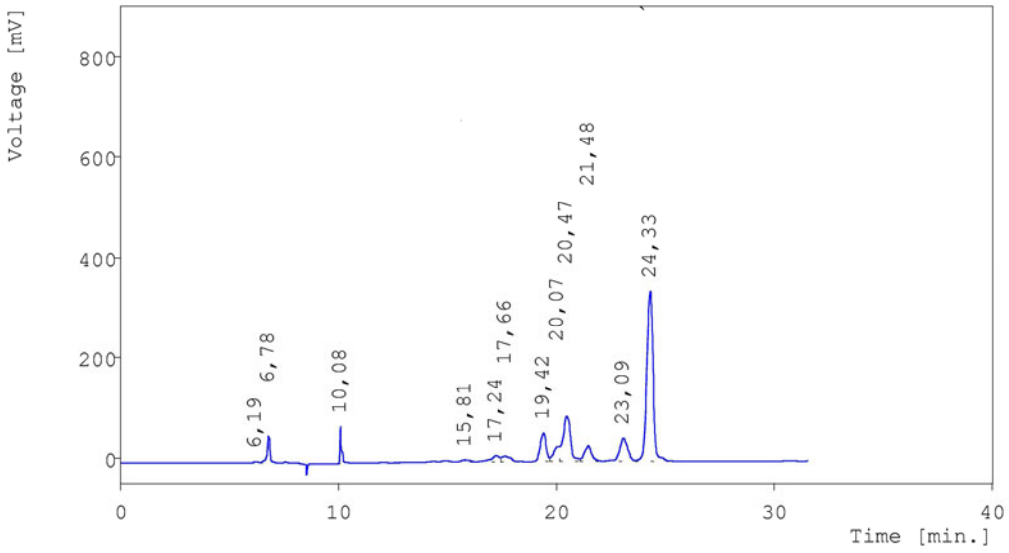
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,240	24,9078	2,396	0,180	0,172	0,305
2	6,740	409,2157	59,713	0,100	2,823	7,603
3	7,987	61,5969	3,813	0,160	0,425	0,486
4	10,320	394,4316	61,191	0,067	2,721	7,791
5	15,553	103,8139	3,984	0,407	0,716	0,507
6	16,993	312,9535	11,472	0,393	2,159	1,461
7	17,547	332,6045	9,395	0,607	2,295	1,196
8	19,420	1076,4575	55,485	0,300	7,427	7,065
9	20,040	634,8081	27,977	0,387	4,380	3,562
10	20,533	2060,3287	92,169	0,333	14,215	11,736
11	21,560	711,2660	32,112	0,333	4,907	4,089
12	23,273	1212,2345	45,463	0,413	8,364	5,789
13	24,573	7159,1170	380,198	0,293	49,396	48,410
-	Total	14493,7357	785,369			

Figure S15(d5): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **8h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.11	13.55	14.36	0.81	27.3930	101.489
2	14.75	14.36	15.31	0.96	148.9190	525.251

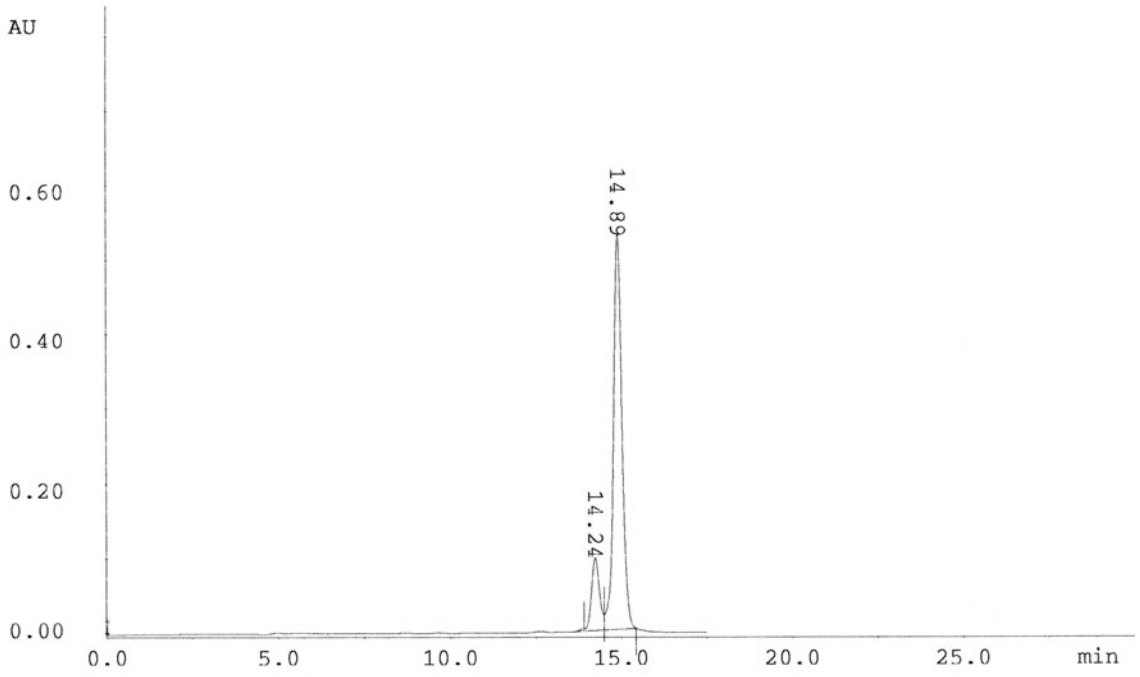
Figure S15(d5i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 24.57 min** in **Figure S15(d5)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.11 min from the heptamer peak eluting at R_T = 14.75 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

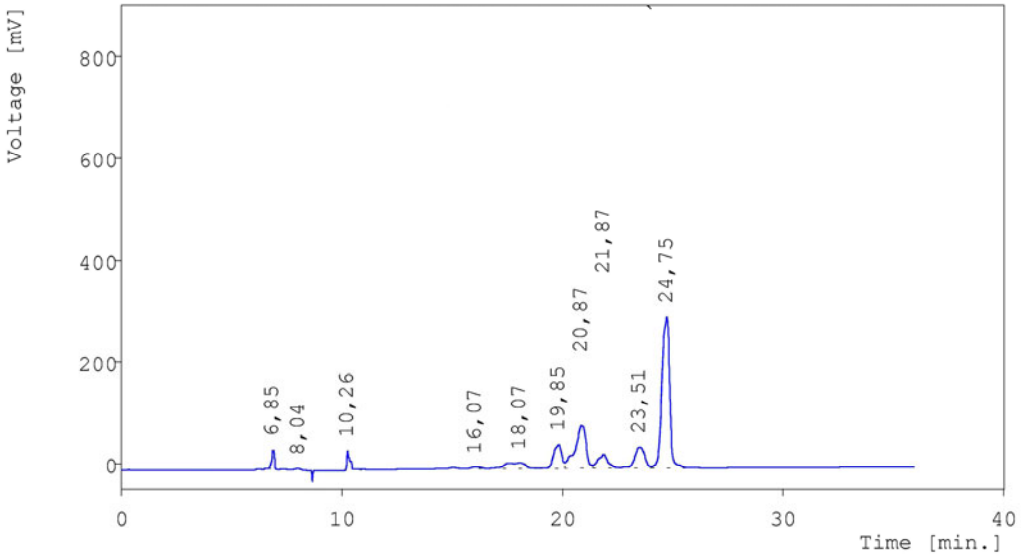
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,193	27,9882	2,271	0,233	0,195	0,302
2	6,780	490,2544	53,191	0,120	3,423	7,079
3	10,080	405,8633	73,556	0,067	2,834	9,789
4	15,807	95,6755	3,592	0,453	0,668	0,478
5	17,240	317,3561	12,105	0,387	2,216	1,611
6	17,660	318,1391	10,872	0,520	2,221	1,447
7	19,420	1087,1037	57,066	0,307	7,591	7,594
8	20,067	551,1103	30,993	0,280	3,848	4,124
9	20,473	2240,4717	90,338	0,373	15,645	12,022
10	21,480	739,5728	31,873	0,353	5,164	4,241
11	23,087	1206,1877	46,468	0,400	8,422	6,184
12	24,327	6841,4214	339,122	0,313	47,773	45,129
-	Total	14321,1441	751,448			

Figure S15(d6): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **15h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.24	13.91	14.49	0.59	25.8350	96.857
2	14.89	14.49	15.43	0.94	151.0045	528.236

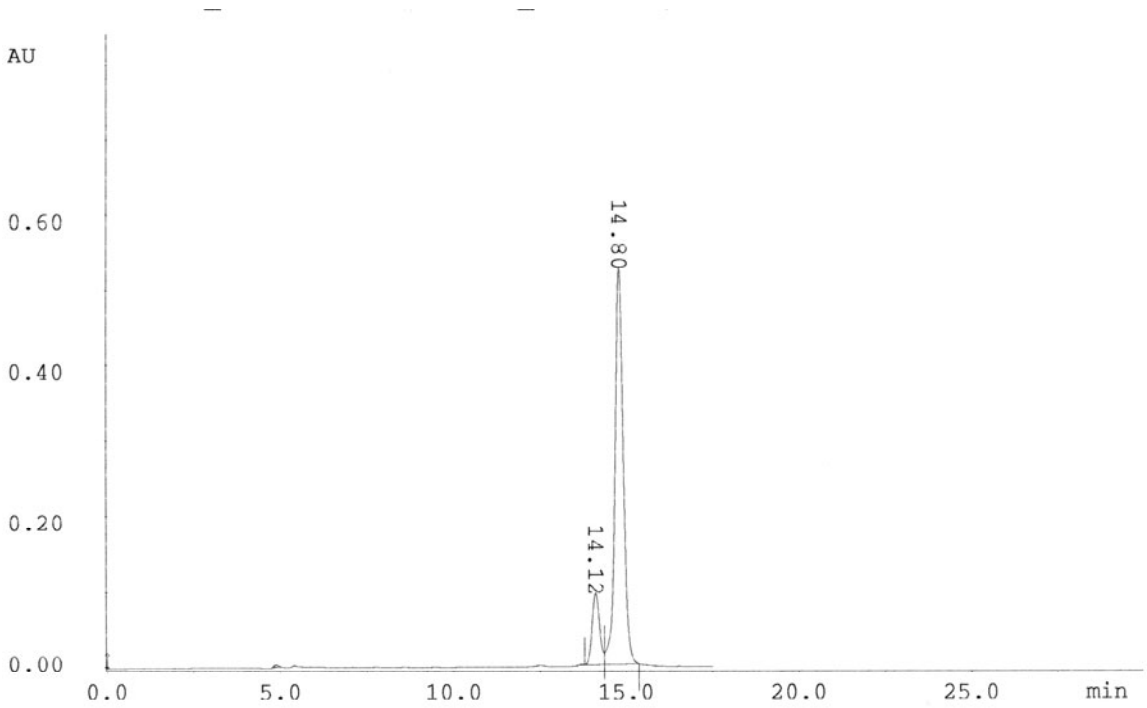
Figure S15(d6i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 24.33 min** in **Figure S15(d6)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.24 min from the heptamer peak eluting at R_T = 14.89 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

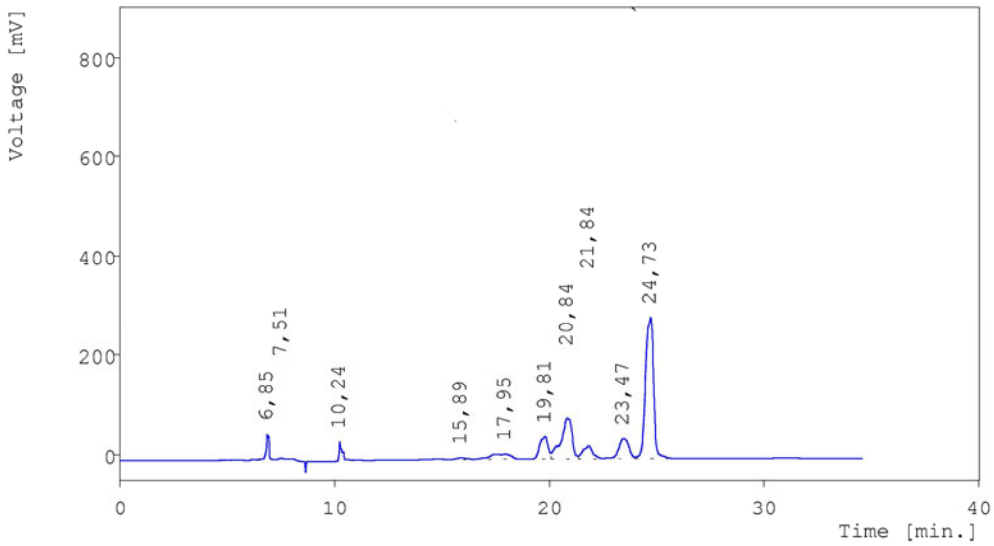
Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,847	323,3811	37,307	0,133	2,120	6,297
2	8,040	64,5276	3,172	0,340	0,423	0,535
3	10,260	432,8494	38,952	0,140	2,838	6,575
4	16,067	118,7310	3,726	0,527	0,778	0,629
5	18,067	738,8041	11,289	1,020	4,844	1,906
6	19,847	1189,0125	47,472	0,413	7,796	8,013
7	20,867	2931,1495	84,146	0,480	19,218	14,203
8	21,873	852,2121	27,169	0,480	5,588	4,586
9	23,513	1304,8674	41,542	0,493	8,555	7,012
10	24,747	7296,5717	297,665	0,393	47,840	50,244
-	Total	15252,1064	592,440			

Figure S15(d7): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **27h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.12	13.80	14.37	0.58	23.5829	94.429
2	14.80	14.37	15.37	1.01	146.0077	526.685

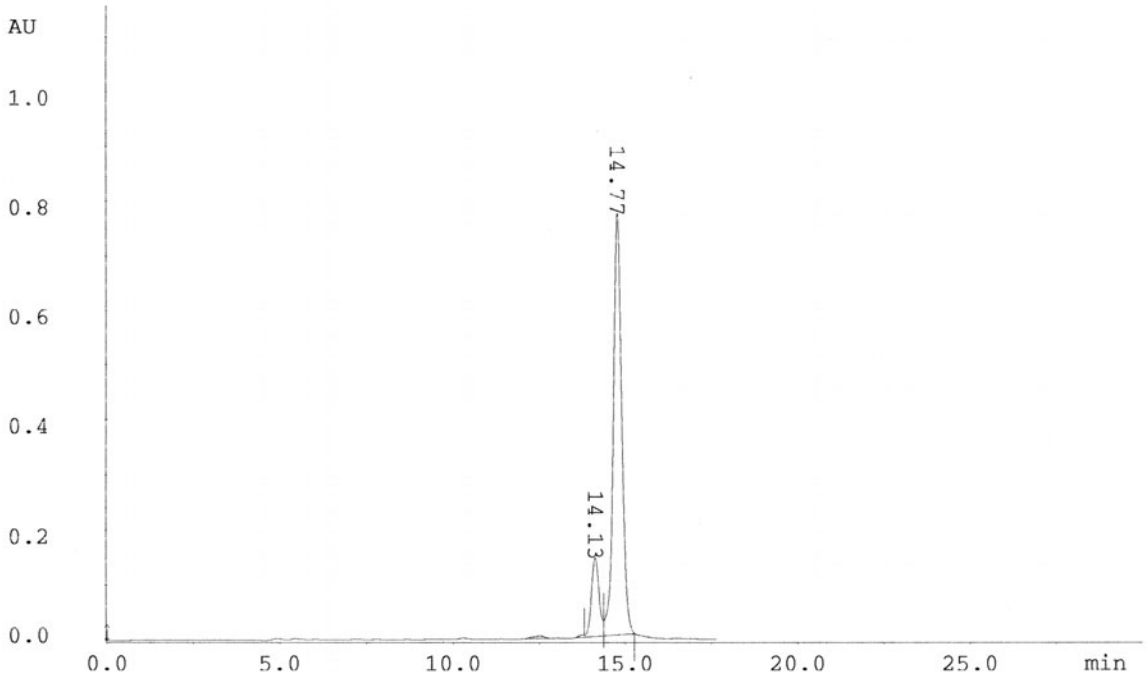
Figure S15(d7i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 24.75 min** in **Figure S15(d7)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.12 min from the heptamer peak eluting at R_T = 14.80 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.



Result Table - Calculation Method Uncal

Peak No.	Reten. time	Area [mV.s]	Height [mV]	W05 [min.]	Area [%]	Height [%]
1	6,853	476,9008	50,038	0,140	3,092	8,611
2	7,507	31,8363	2,331	0,233	0,206	0,401
3	10,240	433,6931	38,329	0,147	2,812	6,596
4	15,893	105,6168	3,137	0,587	0,685	0,540
5	17,953	717,1339	10,464	1,073	4,650	1,801
6	19,813	1179,9994	44,733	0,440	7,650	7,698
7	20,840	2944,9173	81,971	0,487	19,093	14,106
8	21,840	832,3735	25,998	0,507	5,397	4,474
9	23,473	1307,3139	40,074	0,520	8,476	6,896
10	24,733	7394,0575	284,014	0,420	47,939	48,877
-	Total	15423,8424	581,089			

Figure S15(d8): RP-Hplc analysis of alkaline Hydrolysis products of **5′-r(CACGCAC)-3′ (8b)** [after digestion for **48h** at pH 12.5 using 0.03N NaOH/ 20°C, followed by quenching with 0.03 N aq. acetic acid]. For Hplc conditions see the experimental section in the text.



No	Ret (min)	Peak start (min)	Peak end (min)	Dur (min)	Area (min*mAU)	Height (mAU)
1	14.13	13.81	14.37	0.57	36.6624	143.892
2	14.77	14.37	15.27	0.91	217.9755	769.115

Figure S15(d8i): SMART™ RP-Hplc analysis of the heptameric peak at **$R_{T=}$ 24.73 min** in **Figure S15(d8)** for **(8b)**. Note the separation of the hexamer (5'-ACGCAC-3') peak at R_T = 14.13 min from the heptamer peak eluting at R_T = 14.77 min. Hplc conditions: Jupiter 5µm C18 300Å column with 150 x 2 mm dimension. Gradient: linear gradient starting from 0% B Buffer (50% CH_3CN in 0.1M TEAA) + 100% A Buffer (5% CH_3CN in 0.1M TEAA) to 20% B Buffer + 80% A Buffer in 45 minutes with a constant flow rate of 100 µl min^{-1}.