

**Analysis of antisense oligonucleotides with the use of ionic liquids as the mobile phase modifiers**

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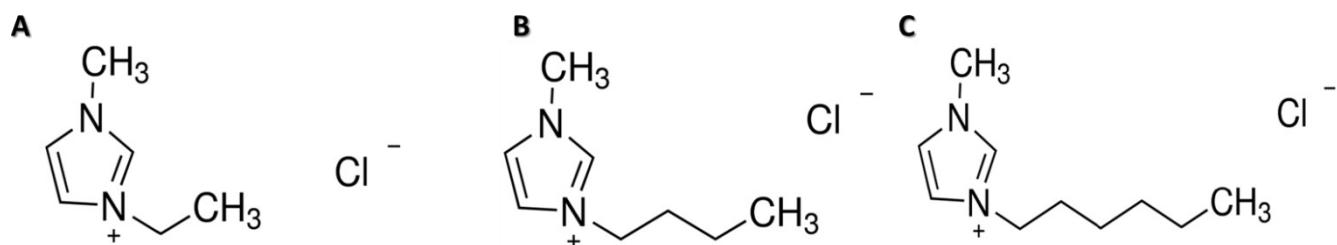


Figure S1. The structures of tested ILs cations: A) [EMIM][Cl], B) [BMIM][Cl], C) [HMIM][Cl].

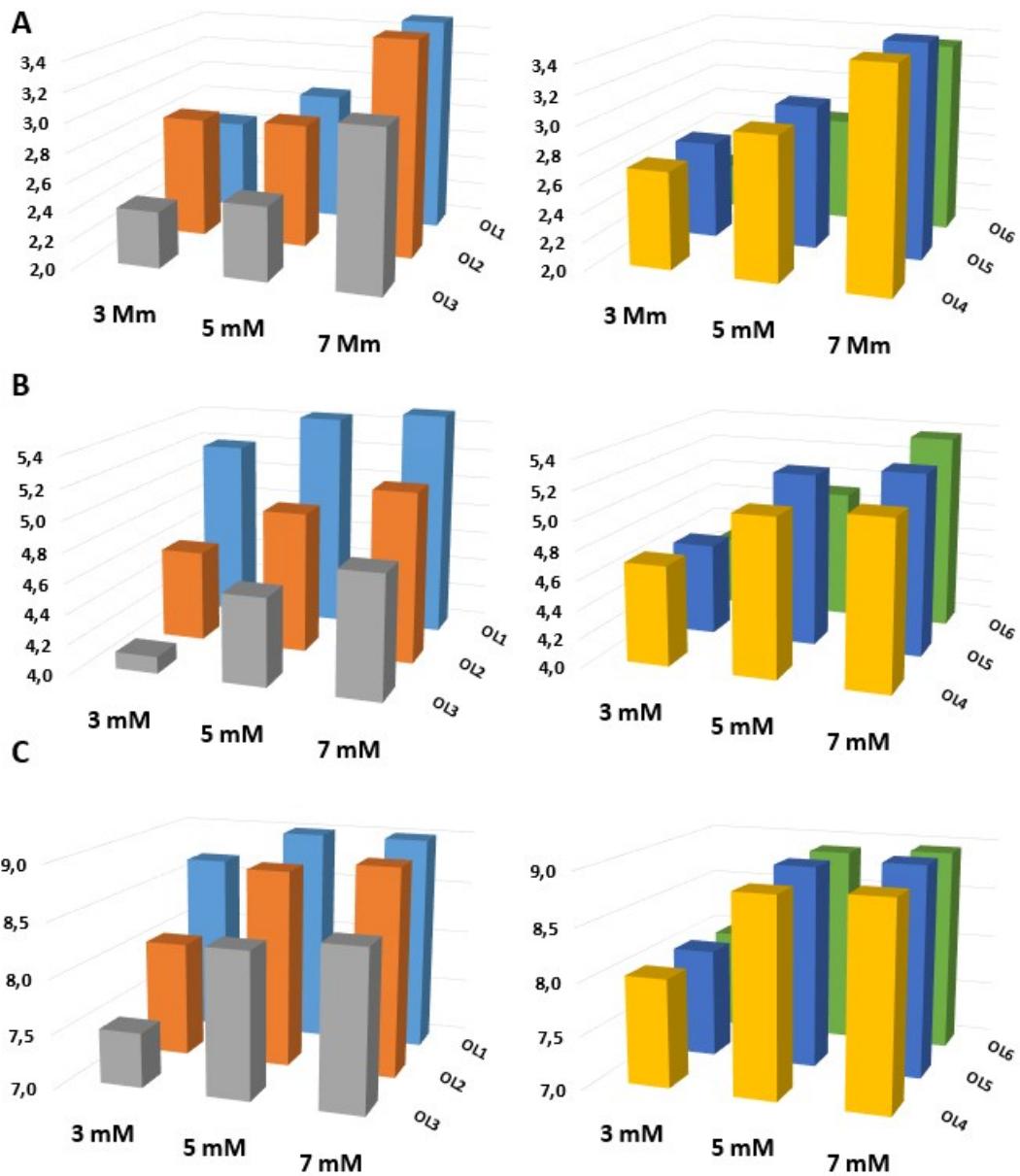


Figure S2. The impact of IL concentration on ASOs retention factors for: A) [EMIM][Cl], B) [BMIM][Cl], C) [HMIM][Cl]. Experimental conditions: Syncronis aQ column, column and autosampler temperature 30°C, mobile phase flow rate 0.3 ml/min; gradient elution program: 25% - 60% v/v MeOH in 15 minutes.

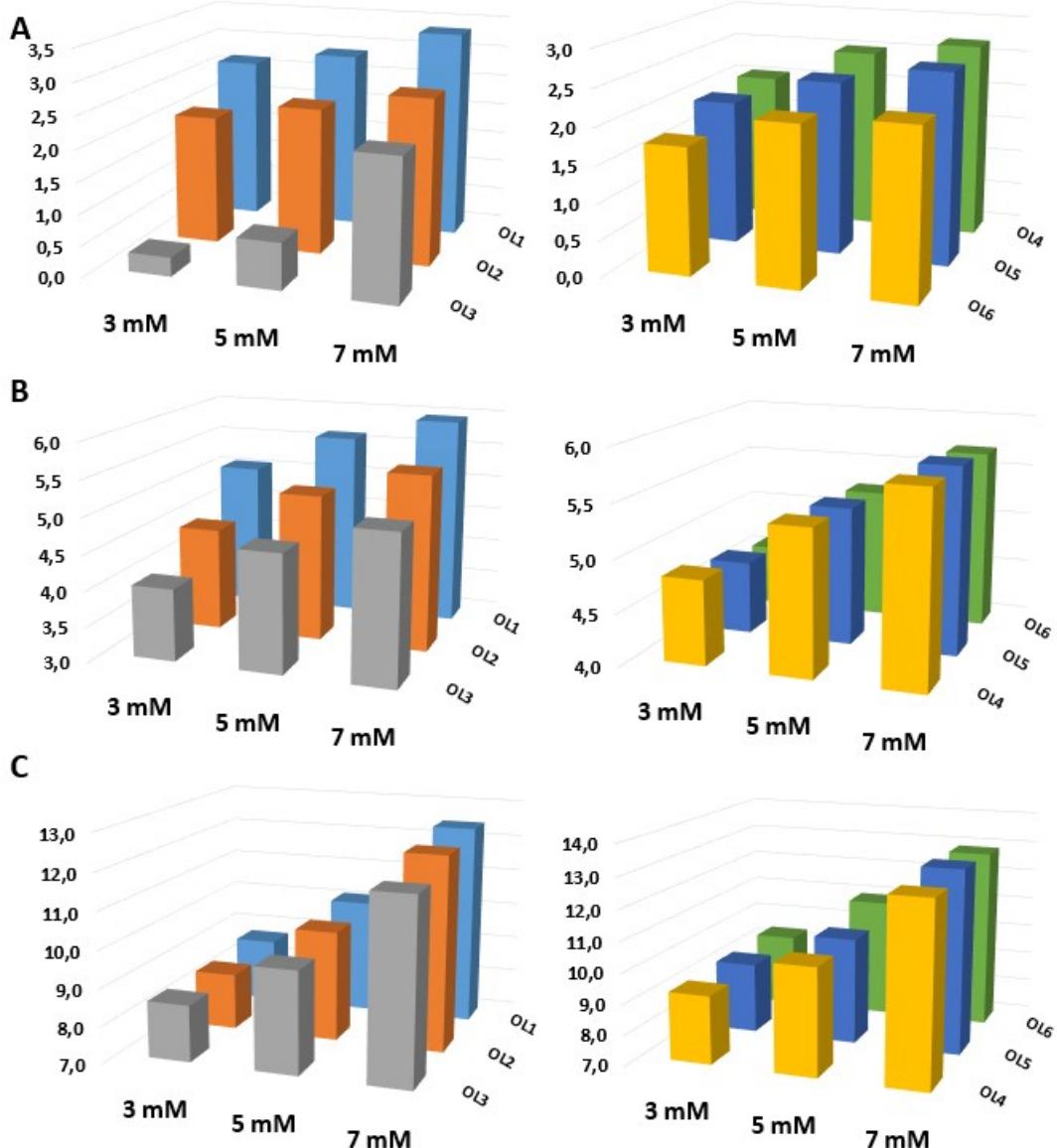


Figure S3. The dependence of the concentration of ionic liquid on ASOs retention coefficients A) [EMIM][Cl], B) [BMIM][Cl], C) [HMIM][Cl]. Experimental conditions: Kinetex C18 column, column and autosampler temperature 30°C, flow rate 0.3 ml/min; gradient elution program: 25% - 60% v/v MeOH in 15 minutes.

Table S1. Asymmetry factors of oligonucleotides for all tested stationary phases. Experimental conditions: column and autosampler temperature 30°C, flow rate 0.3 ml/min for C18 and F5 and 0.2ml/min for aQ; gradient elution program: 25% - 60% v/v MeOH in 15 minutes.

	[EMIM][Cl]			[BMIM][Cl]			[HMIM][Cl]		
<i>Asymmetry factors</i>									
OGN	3 mM	5 mM	7 mM	3 mM	5 mM	7 mM	3 mM	5 mM	7 mM
aQ									
<b>OL1</b>	1.22	1.40	1.56	1.38	1.65	1.97	1.30	1.96	2.20
<b>OL2</b>	1.61	1.21	1.42	1.34	1.59	1.75	1.06	1.65	2.28
<b>OL3</b>	1.88	1.11	1.65	1.67	1.63	1.63	1.59	1.61	2.25
<b>OL4</b>	1.88	1.66	1.83	1.92	2.18	2.54	1.29	1.58	2.33
<b>OL5</b>	1.83	1.90	1.76	1.89	1.95	2.22	1.27	1.79	2.31
<b>OL6</b>	1.61	1.86	1.93	2.13	2.31	2.69	1.20	2.34	2.57
C18									
<b>OL 1</b>	1.71	1.80	1.88	1.95	2.13	1.90	1.00	1.40	1.48
<b>OL 2</b>	0.97	1.22	1.83	1.88	1.99	1.71	1.34	1.52	1.41
<b>OL 3</b>	1.89	2.16	1.13	1.85	1.81	1.79	1.85	1.65	1.67
<b>OL 4</b>	1.14	2.18	2.25	2.40	2.34	2.12	1.41	1.73	1.72
<b>OL 5</b>	1.23	2.02	2.30	2.42	2.40	2.08	1.40	1.54	1.68
<b>OL 6</b>	0.87	1.60	2.39	2.37	2.37	2.12	1.55	1.60	1.92
F5									
<b>OL1</b>	1.44	2.22	2.20	1.56	1.59	1.94	1.24	1.33	1.50
<b>OL2</b>	1.93	2.10	1.96	1.91	1.78	1.90	1.01	1.31	1.71
<b>OL3</b>	1.78	2.0	1.69	2.09	1.79	1.84	1.45	1.99	1.85
<b>OL4</b>	2.63	2.64	2.29	2.45	2.23	2.12	2.30	2.05	2.05
<b>OL5</b>	2.46	2.23	1.95	2.34	2.14	2.17	1.94	2.07	1.92
<b>OL6</b>	2.37	2.07	1.85	2.34	2.01	2.14	2.74	1.77	2.12

Table S2. Mobile phase composition and [BMIM][Cl] retention factors for C18 stationary phase. Experimental conditions: Mobile phase composition H<sub>2</sub>O/MeOH, flow rate 0.3 ml/min, injection volume 5 µl, autosampler and column temperature 30°C.

<i>Percentage of MeOH (% v/v)</i>	<i>k values</i>
30	0
25	0.87
20	1.84
15	3.16
10	4.85
5	7.12