

## Supplementary information

Progress of the selection rounds 4 – 9 with the *full-length library* using a protocol for automated *in vitro* selection.

The first three selection rounds were carried out manually with the *full-length library* at target concentrations of 1  $\mu$ M D-ghrelin (data not shown). The target concentration was kept constant in the subsequent automated rounds (4 and 5) for all three parallel selection strands while the wash volume (wv) was varied; in automated rounds 6-9, the wash volumes were kept constant whereas the target concentrations were adjusted to 900, 300, 100, 33, and 11 nM. The progress of enrichment was determined by quantifying the amount of PCR product using a dsDNA sensitive fluorescent dye. The ratio of fluorescence intensity with / without DNA polymerase is listed under in the respective columns and compared to the void value. The number of PCR cycles are indicated in the first column of the table. The shaded columns indicate the selection conditions that were followed on.

n.a.: not analyzed

<b>R 4</b>	void	5 wv	10 wv	<b>15 wv</b>
7	0,964	1,746	1,934	<b>1,815</b>
10	1,230	3,591	3,759	<b>3,954</b>
13	5,793	-	-	-
16	-	-	-	-
	↓	↓	↓	↓
<b>R 5</b>	void	5 wv	10 wv	<b>15 wv</b>
7	n.a.	n.a.	n.a.	<b>n.a.</b>
10	1,669	2,914	3,168	<b>3,771</b>
13	3,003	-	-	-
16	-	-	-	-
	↓	↓	↓	↓
<b>R 6</b>	void	900 nM	300 nM	<b>100 nM</b>
7	0,994	0,947	0,965	<b>1,037</b>
10	0,914	1,041	0,911	<b>0,866</b>
13	0,992	2,863	1,123	<b>0,969</b>
16	1,150	-	2,147	<b>1,499</b>
	↓	↓	↓	↓
<b>R 7</b>	void	900 nM	300 nM	<b>100 nM</b>
7	1,042	0,947	1,060	<b>1,079</b>
10	0,994	1,007	1,304	<b>1,023</b>
13	0,947	1,408	2,947	<b>1,127</b>
16	1,039	3,385	-	<b>2,452</b>
	↓	↓	↓	↓
<b>R 8</b>	void	300 nM	100 nM	<b>33 nM</b>
10	1,170	6,297	5,652	<b>4,361</b>
13	1,180	-	-	-
16	1,853	-	-	-
19	5,145	-	-	-
	↓	↓	↓	↓
<b>R 9</b>	void	100 nM	<b>33 nM</b>	11 nM
10	1,125	n.a.	<b>3,653</b>	1,226
13	1,295	n.a.	-	2,061
16	1,437	n.a.	-	4,652
19	3,139	n.a.	-	-

wash volume (wv) varied

[D-Ghrelin] 1  $\mu$ M

15 wash volume

[D-Ghrelin] varied