

SUPPLEMENTARY MATERIALS

Analysis of NHEJ-based DNA repair after CRISPR-mediated DNA cleavage

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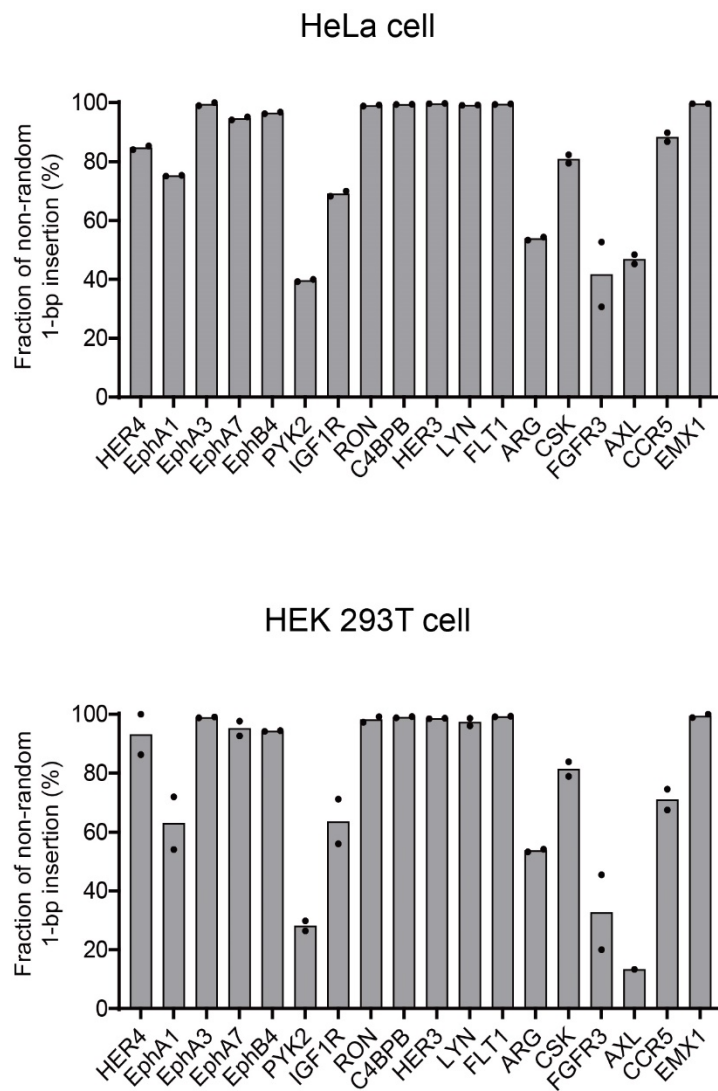
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Supplementary Figure S1. The fraction of non-random 1-bp insertion. The fraction of 1-bp insertion which is identical to -4bp upstream of the PAM is diverse across the genes.



Supplementary Table S1. Sequence of dsODN and target sites of each gene.

dsODN		5' - P-G*T*TAAATTGAGTTGTCATATGTTAATAACGGT*A*T - 3' 3' - C*A*AATTAACCAACAGTATACAATTATTGCCA*T*A - P - 5'		
Gene		sgRNA	Targeted sequence	Strand
<i>HER4</i>		1	CTTGGATTAAAGAACTTGACAGG	-
		2	TTATGAGGATCGATATGCCTTGG	-
		3	TCGTGGGACAAAACCTTTATGAGG	-
		4	ATAATGCGTAAATTCTCCAGAGG	+
		5	AGTCACAGGCTACGTGTTAGTGG	-
		6	CAATTCTTTAGAATATGATATGG	-
<i>EphA1</i>		1	AAGGTCTCCTTGACGCCAGAGG	+
		2	GAAACTCTTGCACTCCCGCACGG	+
		3	ACTGCAGCTCCACGTGGACGCGG	+
		4	GCTCCAATTGGATCTACCGCGGG	-
		5	GACGCAGAGACACTGACCACTGG	-
		6	TACCAGGACTGCCCAATGCAAGG	-
<i>EphB4</i>		1	GTGGGTTGCCCTAATTTGATGGG	-
		2	AAACACGGACAGTATCTCATCGG	-
		3	GCAGAATATTCGGACAAACACGG	-
		4	GCTCTGCTTCCTGTAGCCGATGG	+
		5	CTGAGTCACACATCTTTATGAGG	+
		6	TTATGAGGCACACACACCAAGG	+
<i>EMX1</i>		1	GACTGAGGCTACATAGGGTTAGG	-
		2	GCCGTTTGTACTTTGTCTCCGG	-
		3	GAGTCCGAGCAGAAGAAGAAGGG	+
		4	AGGGCTCCCATCACATCAACCGG	+
		5	CACGAAGCAGGCCAATGGGGAGG	+
		6	GTCACCTCCAATGACTAGGGTGG	+
Duplicated sequence	Chr3:75395213	1	GCAGCTCTATGGAGGCCTGGNGG	+
	Chr5:28926841	2		+
	Chr1:144720581	3		+
	Chr13:25017428	4		+
	Chr9:40883645	5		+

* represents phosphorothioate internucleotide linkages.

Supplementary Table S2. NHEJ accuracy at the two DSB ends in each gene.

sgRNA	<i>HER4</i>		<i>EphA1</i>		<i>EphB4</i>		<i>EMX1</i>	
	PAM-distal	PAM-proximal	PAM-distal	PAM-proximal	PAM-distal	PAM-proximal	PAM-distal	PAM-proximal
1	56.1%	35.8%	83.9%	46.3%	51.8%	82.9%	42.5%	39.1%
2	59.1%	60.8%	88.0%	2.5%	79.5%	12.0%	84.5%	40.6%
3	52.7%	31.4%	22.7%	10.2%	5.3%	7.5%	47.3%	2.6%
4	95.4%	51.2%	60.7%	92.5%	79.2%	63.0%	89.8%	70.3%
5	35.0%	87.5%	64.3%	90.0%	84.5%	52.7%	81.5%	54.7%
6	69.7%	57.8%	25.0%	0.0%	78.0%	34.6%	75.8%	52.6%
Avg. NHEJ accuracy	61.3 ±8.2%	54.1 ±8.2%	57.4 ±11.5%	40.3 ±17.5%	63.0 ±12.5%	42.1 ±12.1%	70.2 ±8.2%	43.3 ±9.4%

Supplementary Table S3. Targeted sequence of each gene.

Gene	Targeted sequence	Strand	Location
<i>HER4</i>	ATAATGCGTAAATTCTCCAGAGG	+	Chr2
<i>EphA1</i>	GCTCCAATTGGATCTACCGCGGG	-	Chr7
<i>EphA3</i>	TTGTCGACCAGGTTTCTACAAGG	+	Chr3
<i>EphA7</i>	CACCTGGTATGTTCGTATCGGGG	+	Chr6
<i>EphB4</i>	GCAGAATATTCGGACAAACACGG	-	Chr7
<i>PYK2</i>	GGTCCTGAATCGTATTCTTGGGG	+	Chr8
<i>IGF1R</i>	TCAGTACGCCGTTTACGTCAAGG	+	Chr15
<i>RON</i>	GTCATCGGGCCGGTTATGGTGGG	+	Chr3
<i>C4BPB</i>	AATGACCACTACATCCTCAAGGG	+	Chr1
<i>HER3</i>	ACCATTGCCCAACCTCCGCGTGG	+	Chr12
<i>LYN</i>	TTAATCATGTCGCTGATACAGGG	-	Chr8
<i>FLT1</i>	AAAATAAGAGAGCTTCCGTAAGG	-	Chr13
<i>ARG</i>	TCCATCTCGCTCAGGTACGAGGG	-	Chr1
<i>CSK</i>	CTGACCGACCCCTAGACCGCAGG	-	Chr15
<i>FGFR3</i>	CGGCAACTACACCTGCGTCGTGG	+	Chr4
<i>AXL</i>	GTCCCGTGTCGAAAGCTGCAGG	-	Chr19
<i>CCR5</i>	TGACATCAATTATTATACATCGG	+	Chr3
<i>EMX1</i>	GAGTCCGAGCAGAAGAAGAAGGG	+	Chr2

Supplementary Table S4. NHEJ accuracy in each gene.

Gene	HeLa cell		HEK 293T cell	
	PAM-distal	PAM-proximal	PAM-distal	PAM-proximal
<i>HER4</i>	95.32%	47.75%	94.98%	55.11%
<i>EphA1</i>	55.22%	91.92%	54.23%	93.19%
<i>EphA3</i>	68.19%	66.76%	89.43%	8.47%
<i>EphA7</i>	81.02%	12.68%	86.04%	17.08%
<i>EphB4</i>	5.32%	6.97%	34.70%	17.59%
<i>PYK2</i>	87.99%	21.10%	91.38%	22.09%
<i>IGF1R</i>	33.50%	12.46%	80.40%	22.56%
<i>RON</i>	73.84%	67.68%	88.47%	32.74%
<i>C4BPB</i>	84.74%	47.69%	94.07%	18.84%
<i>HER3</i>	93.11%	0.65%	96.52%	0.77%
<i>LYN</i>	49.76%	28.78%	50.94%	15.30%
<i>FLT1</i>	64.04%	59.18%	74.00%	23.99%
<i>ARG</i>	36.73%	85.41%	35.91%	91.61%
<i>CSK</i>	53.58%	89.17%	49.61%	90.95%
<i>FGFR3</i>	71.38%	16.68%	95.79%	38.11%
<i>AXL</i>	51.22%	89.58%	49.82%	91.24%
<i>CCR5</i>	92.09%	57.98%	96.16%	51.21%
<i>EMX1</i>	44.86%	3.20%	72.89%	4.32%
Avg. NHEJ accuracy	63.44±5.75%	44.76±7.68%	74.18±5.22%	38.62±7.64%

Supplementary Table S5. The fraction of predictable 1-bp insertion among the total 1-bp insertion in the two human cell lines.

Gene	HeLa cell	HEK 293T cell
<i>HER4</i>	84.70%	93.16%
<i>EphA1</i>	75.21%	63.01%
<i>EphA3</i>	99.50%	98.95%
<i>EphA7</i>	94.65%	95.18%
<i>EphB4</i>	96.53%	94.35%
<i>PYK2</i>	39.59%	28.09%
<i>IGF1R</i>	69.07%	63.56%
<i>RON</i>	99.04%	98.22%
<i>C4BPB</i>	99.45%	99.03%
<i>HER3</i>	99.72%	98.63%
<i>LYN</i>	99.20%	97.39%
<i>FLT1</i>	99.52%	99.26%
<i>ARG</i>	53.86%	53.71%
<i>CSK</i>	80.88%	81.41%
<i>FGFR3</i>	41.65%	32.73%
<i>AXL</i>	46.81%	13.33%
<i>CCR5</i>	88.31%	71.02%
<i>EMX1</i>	99.68%	99.45%