Supporting Information

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Table S1. Recognition helices for ZFN10, ZFN11, ZFN-H2a, and ZFN-H2b

	Target (5′ → 3′)	Recognition helix I	Recognition helix II	Recognition helix III
ZFN10	ATGGATGCA	QSGDLRR (1)	TSGNLVR (1)	RRDELNV (2)
ZFN11	TTGTGGGAA	QSSNLQK (3)	RSDHLTT (4)	RSDSLTK (5)
ZFN-H2a	GTTGGTGCT	QSSDLTR (6)	TSGHLVR (1)	TSGSLVR (1)
ZFN-H2b	GCTGCTGTC	DRSALAR (6)	QSSDLTR (6)	QSSDLTR (6)

References are in parentheses.

1. Dreier B, Segal DJ, Barbas CF, III (2000) Insights into the molecular recognition of the 5′-GNN-3′ family of DNA sequences by zinc finger domains. *J Mol Biol* 303:489–502.
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3. Kim YG, Shi Y, Berg JM, Chandrasegaran S (1997) Site-specific cleavage of DNA-RNA hybrids by zinc finger/FokI cleavage domain fusions. *Gene* 203:43–49.
4. Segal DJ, Dreier B, Beerli RR, Barbas CF, III (1999) Toward controlling gene expression at will: selection and design of zinc finger domains recognizing each of the 5′-GNN-3′ DNA target sequences. *Proc Natl Acad Sci USA* 96:2758–2763.
5. Mani M, Kandavelou K, Dy FJ, Durai S, Chandrasegaran S (2005) Design, engineering, and characterization of zinc finger nucleases. *Biochem Biophys Res Commun* 335:447–457.
6. Liu Q, Xia Z, Zhong X, Case CC (2002) Validated zinc finger protein designs for all 16 GNN DNA triplet targets. *J Biol Chem* 277:3850–3856.

Table S2. Primers used for the assembly of ZFN10, ZFN11, ZFN-H2a, and ZFN-H2b

Name	Use	Sequence (5′→3′)
BBO1	Backbone primer 1	GAAAAACCTTACAAGTGTCCTGAATGTGGAAAGTCTTTTTCT
BBO2	Backbone primer 2	CAGCGAACACACACAGGTGAGAAGCCATATAAATGCCCAGAATGTGGTAAATCATTCAG
BBO3	Backbone primer 3	CAACGGACCCACAfCCGGGGAGAAGCCATTTAAATGCCCTGAGTGCGGGAAGAGTTTTT
ZFP10-SDO1	Helix primer I in ZFP10	ACCTGTGTGTGTTCGCTGGTGACGACGCAAATCTCCAGACTGAGAAAAAAGACTTTCCACA
ZFP10-SDO2	Helix primer II in ZFP10	CCCGGTGTGGGTCCGTTGGTGACGAACCAAATTTCCAGAAGTACTGAATGATTTACCACA
ZFP10-SDO3	Helix primer III in ZFP10	TCCAGTATGAGTACGTTGATGAACATTCAATTCATCACGACGTGAAAAAACTCTTCCCGCAC
ZFP11-SDO1	Helix primer I in ZFP11	ACCTGTGTGTGTTCGCTGGTGCTTCTGAAGGTTGCTAGACTGAGAAAAAAGACTTTCCACA
ZFP11-SDO2	Helix primer II in ZFP11	CCCGGTGTGGGTCCGTTGGTGAGTAGTAAGATGATCGGAACGACTGAATGATTTACCACA
ZFP11-SDO3	Helix primer III in ZFP11	TCCAGTATGAGTACGTTGATGCTTAGTTAGTGAATCGGAACGTGAAAAAACTCTTCCCGCAC
ZFN-H2a-SDO1	Helix primer I in ZFN-H2a	ACCTGTGTGTGTTCGCTGGTGACGAGTAAGATCAGAAGACTGAGAAAAAAGACTTTCCACA
ZFN-H2a-SDO2	Helix primer II in ZFN-H2a	CCCGGTGTGGGTCCGTTGGTGACGAACAAGATGTCCAGAAGTACTGAATGATTTACCACA
ZFN-H2a-SDO3	Helix primer III in ZFN-H2a	TCCAGTATGAGTACGTTGATGACGAACAAGAGATCCAGAAGTTGAAAAAACTCTTCCCGCAC
ZFN-H2b-SDO1	Helix primer I in ZFN-H2b	ACCTGTGTGTGTTCGCTGGTGACGAGCAAGAGCAGAACGATCAGAAAAAAGACTTTCCACA
ZFN-H2b-SDO2	Helix primer II in ZFN-H2b	CCCGGTGTGGGTCCGTTGGTGACGAGTAAGATCAGAAGACTGACTGAATGATTTACCACA
ZFN-H2b-SDO3	Helix primer III in ZFN-H2b	TCCAGTATGAGTACGTTGATGACGAGTAAGATCAGAAGACTGTGAAAAAACTCTTCCCGCAC

Underlined letters indicate complementary regions between BBO and the corresponding SDO primers.

Table S3. Primers used for the construction of pSAT10, pSAT11, pSAT12.1, and pRCS11

Name	Used in assembly of	Sequence (5′ → 3′)
SAT10-F	pSAT10	ATAAGAATGCGGCCGCTGCATCCATGTAAGTATGGATGCAGTAATCATGGTCATAGCTGTTTCC
SAT10-R-V2	pSAT10	GACGCACCGGTTGCATCCATACTTACATGGATGCAGGCACTGGCCGTCGTTTTACAACG
SAT11-F	pSAT11	ATAAGAATGCGGCCGCTTCCCACAAGTAAGTTTGTGGGAAGTAAGTATGGATGCAGTAATCATGGTCATAGCTGTTTCC
SAT11-R	pSAT11	GACGCACCGGTTTCCCACAAACTTACTTGTGGGAAGGCACTGGCCGTCGTTTTACAACG
SAT12-F	pSAT12.1	ATAAGAATGCGGCCGCAGCACCAACGTAAGTGTTGGTGCTGTAAGTATGGATGCAGTAATCATGGTCATAGCTGTTTCC
SATI2-R	pSAT12.1	GACGCACCGGTAGCACCAACACTTACGTTGGTGCTGGCACTGGCCGTCGTTTTACAACG
RCS11-UP-1	pRCS11	TCCCCCGGGTTCCCACAAACTTACTTGTGGGAAAGCACCAACACTTACGTTGGTGCTCCCGGGGGA
RCS11-DW-1	pRCS11	TCCCCCGGGAGCACCAACGTAAGTGTTGGTGCTTTCCCACAAGTAAGTTTGTGGGAACCCGGGGGA
RCS11-UP-2	pRCS11	GGGGTACCTGCATCCATACTTACATGGATGCAGGTACCCC
RCS11-DW-2	pRCS11	GGGGTACCTGCATCCATACTTACATGGATGCAGGTACCCC

Underlined letters indicate ZFN target sites.