Supplementary Table 2 Potential off-target sites of *WAS* CR-4 identified by *in silico* prediction.

No. of Mismatches	Name	% Indel	Chr	Start	End	Forward primer	Reverse primer	Sequence
0	WAS		chrX	48544569	48544591	TGCGTGCTGATTCTTCCCTG	TGCTCGTCCATCCACATACC	TGGATGGAGGAATGAGGAGTTGG
1	MATN1-AS1	6.27	chr1	30597854	30597876	TGGCACATAGAGGAGCCTGA	GGGGGCTGTTTGTTATGCAC	TGGATGGAGGGATGAGGAGTGGG
2	STK25	6.46	chr2	242451392	242451414	GAGGTCGACAGCAGAGACAC	TGCTAGGAAACCCTGTTGCT	CCCACTCCTCATCCCTCCATCCC
2	GNRH2	7.14	chr20	2989118	2989140	TCAAATTCACAGGGACAGAAAG	TCACCACCATCCGTCTACAG	GGGAGGGAGGAATGAGGAGTGGG
2	POT1	UD	chr3	193793860	193793882	CCACAGGACAGAGTATGGGC	GCAAGTTTCTCAAAAATGGCTTCC	CCCACTCGTCATTCCTCCATCCC
2	POT2	0.60	chr4	1500296	1500318	AGATGAATGGATGGGTGGA	CCATCATCCATCCCCCTAC	TGGATGGAAGAATGAGGGGTGGG
2	POT3	UD	chr14	101313660	101313682	GGCCAAAAAGTCTACATTGCGT	AGTGCCCAGGACTGGATGA	TGGATGGATGAATGAGCAGTGGG
2	POT4	UD	chr7	116536847	116536869	AGAGCTCAGTAAGTATTTGCAT	ACAAAAACAGTAGTTTGGACCAG	TGGATGAAGGAATGATGAGTGGG
3	OSBPL1A	3.33	chr18	21810215	21810237	AGTAGGGTTAGGCCACGACA	TTCCTATGCAACCATCACCA	AGGAGGGAGGAATGGGGAGTTGG
3	CD70	1.24	chr19	6611501	6611523	GGCACTGGTCACCTGTTTGA	TGAGAGGAGTGCTTGATGAAAAGA	CGGACGGAGGAATGGGGAGTGGG
3	Hoxc13	0.58	chr12	54279147	54279169	AGGGAAACCAGGCCAAGAAA	GGCCCACAAACTTGGAGCTA	ACGATGGAGGAATGAGGACTTGG
3	POT5	0.40	chr10	57493261	57493283	CAATCTTAAGGAGGAAAGAATTTGC	GGATGGTTATATGCAGAATGCTCAA	GCGATGGAGGAATGAAGAGTTGG
3	POT6	0.27	chr8	48081086	48081108	TCATTCTCCAGAGCTCCCCA	GGAAGGGCAGTATGGGTCAG	CCCTCTCCTCATTCCTCCATGGA
3			chr7	150636455	150636477			CCTAATCCTCATTCCTCCATGCT
3	POT7	0.04	chr11	128663288	128663310	CCCCGTGTCCCTCCATAAAT	GTGGGGCATTCCAGAGCAAT	TAGATGAAGGAATGAGGAGAAGG
3			chr9	125689354	125689376			CCAACTCCTCACTCCTCCATTCT
3			chr16	3071856	3071878			CCCACTCATCATTCATTCATCCA
3	POT8	UD	chr1	15091127	15091149	GGAGCTCATACCAGGCACTC	ACTGTCCTCAAGGATGGTGAC	TGGATGAAGAAATGAAGAGTTGG
3			chr1	65288700	65288722			GGGCTGGAGGAAGGAGGAGTTGG
3	POT9	UD	chr22	22709609	22709631	AGGCTGTGATCCTAGAATGATGT	GGACAGTCTCCTCTTCAGGGT	TTTATGGAGGAATGAGGAGATGG
3			chrX	79741553	79741575			TCCATGGAGGAATGAGGACTTGG
3			chr2	491190	491212			AGGATGAAGGAATGAGCAGTGGG
3	POT10	UD	chr20	25050236	25050258	CTCCTGCTCCCAGTCTTGTG	CTAGCCCTGGTTGCCCTTG	AGGATGCAGGACTGAGGAGTGGG
3	POT11	UD	chr1	29595076	29595098	GCTGAGGTTGAGAATCCCCG	TCCCTGCAATTGTCTGCATT	TAGATGGAAGAAAGAGGAGTTGG
3			chr11	124481386	124481408			CCGTCTCCTCATTCCACCATCCC
3			chr18	22667995	22668017			GGGATGGAAGACTGAGGAGTGGG
3	POT12	UD	chr5	38143684	38143706	TGTTTGGGTGTTCCCCACTC	ACAGTGGGGCTTGATGGAAG	CCTACTCATCATTCCTCCATACG
3			chr20	39221098	39221120			TGAATGTAGGAATGAGGATTGGG
3	POT13	UD	chr12	55440390	55440412	GACATGAGCCACCCTCCTTC	AATCCAAAGGCACACAGCAA	GGGTTGGTGGAATGAGGAGTAGG
3			chr16	30678115	30678137			TGGTTGTAGGAATGATGAGTAGG
3	POT14	UD	chr8	67261130	67261152	TCAGGGTATTTGGGCCAGAC	TCCAGAGTAGTTTTGCAGGGA	CCTACTCCTCATTCCACCATCTC
3	POT15	UD	chr2	86518341	86518363	TGACACACTGCCTTCCTGAC	TCAGGGCCAGAGTGGAATTT	TGAATGAATGAATGAGGAGTAGG
3			chr10	104817837	104817859			TGGAAGCAGGAGTGAGGAGTAGG
3			chr16	78988298	78988320			TGCATGGATGAATGAGTAGTTGG
3			chr18	834335	834357			TGGCTGGTGGAACGAGGAGTCGG
3			chr12	94153021	94153043			TGGGTGGGGAATGGGGAGTGGG
3	POT16	UD	chr9	130512304	130512326	AATGGCTGTGACCTGAGACC	AAAGCAGCTTGAGGGTGCAA	CCTAATCCTCATTCTTCCAACCA
3			chrY	19152262	19152284			CCTACCCCTCATTCATCCTTCCA
3			chr1	67073705	67073727			CCCACTCCTCATCCACCCATCCA
3			chr13	106193935	106193957			CCCACTTCTCATTTCCCCATCCA
4			chr1	38179295	38179317			CCCACTCCTCATTCATTCCTCCC
4	POT17	UD	chr2	240313168	240313190	GGAGCTTCAGAAAGTCCTGC	TGAGGACACACAGCCCTCTC	CCTACTCCTCATTCCTCCCAAGA

No. of Mismatches	Name	% Indel	Chr	Start	End	Forward primer	Reverse primer	Sequence
4			chr1	72361126	72361148			CCTACTCCTCATTCATCCCTTCC
4	POT18	0.04	chr2	199886050	199886072	AGCTCGAAAACCAGTAAGACCA	ACATTTTACAATGCCCTCTTGTGA	TCTCAGGAGGAATGAGGAGTTGG
4			chr10	103350528	103350550			CCAACTCCTCATTCCCTCCTCCT
4			chr11	93105665	93105687			CCAACTCCTCATTCCCCCACCGT
4	POT19	UD	chr12	5420444	5420466	AGCTCACCAACCGGAAAAGT	TGGTGTTCCCTCCTCTTGAA	GGGAGGAATGAATGAGGAGTTGG
4			chr13	65667276	65667298			GGAATAGAAGAATGAGGAGTGGG
4			chr14	84980632	84980654			CCCACTCCTCATTACACCTCCCA
4	POT20	UD	chr17	47585446	47585468	TGTTACGCTGGGCTAAGGC	CTGATGCCACTTGCCTGTCT	TTGGAGAAGGAATGAGGAGTAGG
4			chr19	42868525	42868547			CCCACTCCTCATTCATACCTCGA
4			chr19	30379572	30379594			CGGCAGGTGGAATGAGGAGTGGG
4	POT21	UD	chr11	109738981	109739003	AACCCCCTGCAACTGTCAAG	CCAGACATGTGGATTGTACTGGT	CCTACTCCTCATTCCTCCCGTCT
4			chr12	3082716	3082738			TGGGACCAGGAATGAGGAGTGGG
4			chr20	30447984	30448006			TTGTTGAATGAATGAGGAGTAGG
4	POT22	UD	chr3	128958553	128958575	GAGCCATCTCTAGATTTTTATGGGT	GCCAGAGGTGTTTCTTTCCCA	AGGAGAGAGAAATGAGGAGTAGG
4			chr5	44714205	44714227			CCAACTCCTCATTCCTTTTTCAA
4			chr5	86010275	86010297			TGGGTTTTGGAATGAGGAGTTGG
4			chr5	27804049	27804071			CCTACTCCTCATTCCTTTCCCCA
4			chr9	135712725	135712747			AGAATGAAAGAATGAGGAGTTGG
6	Mycn	1.18	chr2	15936423	15936444	TTGGGCAGGTGGGAGTCAAT	CCACTCCTGTTGACCTGCTC	AGGATGGGGAAATGAGGAGTAGG

No. of Mismatches denote the number of base mismatches between the WAS CR-4 target site and other genomic POT sites. % Indel denotes the frequency of indel based on high throughput sequencing of the PCR product; Start and End denote the chromosomal coordinates of the IDLV mapped off-target and POT sites; Forward primer and Reverse primer show the primers for PCR ; Sequence denotes the off-target and POT sequence; UD, undetectable.

Supplementary Table 3. Potential off-target sites of *WAS* CR-5 identified by *in silico* prediction.

No. of Mismatches	Name	% Indel	Chr	Start	End	Forward primer	Reverse primer	Sequence
0	WAS		chrX	48544540	48544562	GAGAAGAGGAGGGCTCCCA	TTACGCACCCATCCAACTCC	CCCTGTGTCTCTGGATGGATGGG
3	PABPC5	0.51	chrX	90817726	90817748	CCCCTGAGATGAAGAGGTTTCC	ACAGTGGACGGTCATTCTCAG	CCCTGTGTCTCTGGGTGGAAGAG
3	POT23	UD	chr1	119468927	119468949	AGATCCACTTGATTTTTGCTACCT	GTCAATGGCCTGTGGCAGA	CCATTCTATCCAGAGACACAGGG
3	POT24	UD	chr7	94042192	94042214	ACCTCTAGTGTATCCTTATATCCCT	TCCCTAGTCTGTGGTTTCTGTT	CCCATGCTTCCACAGACACAGGG
3	POT25	UD	chr7	119233024	119233046	GTCAACTGCAGGTTTGCGAC	AGGTCTTCCCAGCAGAGACA	CCCACCCAGCCACAGACACAAGG
3	POT26	UD	chr10	80608251	80608273	GTCAACTGCAGGTTTGCGAC	AGGTCTTCCCAGCAGAGACA	CCCACCCAGCCACAGACACAAGG
3	POT27	UD	chr11	62373613	62373635	GGGCCCCCACTGTACCA	GACCTATGGGATTGTGGCCC	CCCGTCCCTCCGGAGACACAAGG
3	POT28	UD	chr14	42146198	42146220	TGTGGGACCAGAAAATGAAAAC	GATGTGTTCCTGGATTGGCA	CACATGCATCCAGAGACAAAAGG
3	POT29	UD	chr5	81612544	81612566	CCGAAGCCAGGACAAGAGTC	GAGCACTTTGGCATTGTCCC	CCTTGTGTCTATGGAGTGATGGG
3	POT30	UD	chr7	36338348	36338370	GATTCACAGCAGCGCGTTTA	AGGCAGTGAAAACACCCAGA	CCATGTGTGTCTGGATGGACAGG
3	POT31	UD	chr10	72460177	72460199	TTTCTCCCTTCTGCCTGCTG	CACATCTCCTCTCTGCCACC	CCTTGTGACTCTGCCTGGATGGG
3	POT32	UD	chr11	105603133	105603155	GCGTGAACCAGCAGTAGCAT	CTCCCTAAACCATGACGGCT	CCGTGTGTTTCTGGAGGGATGGA
3	POT33	UD	chr13	43743725	43743747	ATGGGTCTCAGCCATCTCCT	TGACTGGAATGTTACAGGATGT	CCTTGTGTCTCTTAATGGGTGGG
3			chrY	4672017	4672039			CCCTGTGTCTCTGGGTGGAAGAG
4	POT34	UD	chr1	25341992	25342014	GCCCCCAGCCTGATTTCTT	AAACCCACAGAGACAGAGGC	CCTCTGCATTCAGAGACACAGGG
4	POT35	UD	chr2	231799753	231799775	GCCAGGACACACATGGGTTA	GAGGGAGAGGAAAAGTGCCC	CCCTTCATGCCAGAGACACACGG
4			chr5	157146235	157146257			CTCCCCCATACAGAGACACAGGG
4	POT36	UD	chr10	98024008	98024030	GAATGGCCCCAAGTCTGTGA	AACCAAGACTCCGTCTCTGC	ATCACCAATCCAGAGACACAGGG
4	POT37	UD	chr11	64017224	64017246	GTGTCTGGGAAGGAAGGAGC	GGCAGCAGCTCCTTGTGG	CCCTGGCCTCCAGAGACACAGGG
4			chr13	113905739	113905761			AGAAGCCATCCAGAGACACAGGG
4			chr15	55794312	55794334			GCCCTTCAGCCAGAGACACAGGG
4	POT38	UD	chr18	55769502	55769524	GGCAGTAATGGTACAGAGGGG	AGTTTCAGGAAAGTGAGCCG	CACATGTCTCCAGAGACACAGGG
4	POT39	UD	chr20	29910751	29910773	AGAGAGAGGTGCAGGAAGGT	AAAAACCGTGGTAATAAGGAACTT	TCCATCCGGACAGAGACACACGG
4			chr2	12490329	12490351			CCATGTGTCTCTGTATGTGTGTG
4	POT40	UD	chr7	152674102	152674124	TCCCTACCCCAAATTTTGTGACT	GCTCCTTATAGCATAAAATGACCTG	CCATGTGTCTCTGTTTGCATGTG
4	POT41	UD	chr9	101674109	101674131	TGGACTTGCTTGTTTATACCCCA	GTTCCCAACCAGCCACTGAC	CCGTGTGTCTCTGGGTGGGTACG
4	POT42	UD	chr10	63590623	63590645	AGAATGTGGCCCAGGAGACT	GAGTCCAGTGACTTGGAGGG	CCATGTGTCTCTGGCTGGTTGTC
4			chr11	65602587	65602609			CCATGTGTCTCTGTGTGTATGTG
4	POT43	UD	chr12	131737388	131737410	CCCCTGCCTGAGTGGTCTTA	CACCAGGGTATGGGCTTTCA	CCCTGTGTCTCTGGGGGGCTGAG
4	POT44	UD	chr20	31491514	31491536	ATGTGTACTGCCATCTGTTCTCT	TCCCAGCCAGTTCCTTCATC	CCTTGTGTCTCTGGACAGATGAT
4			chrX	43259240	43259262			CCATGTGTCTCTGGGTGCTGGGG

See explanation in Supplementary Table 2.

Supplementary Table 4. Genome-wide mapping by IDLV of the off-target sites generated by the *VEGFA* site 3 CRISPR

CRISPR	No. of IS	Chr	Gene	Strand	Distance	Coordinate	No. of Mismatch	Sequence
VEGFA site 3	25	6	*VEGFA*	+	-474	43737471	0	GGTGAGTGAGTGTGTGCGTGTGG
	3	11	*TPCN2*	+	34791	68851140	2	GGTGAGTGAGTGCGTGCGGGTGG
	8	14	*MAX*	-	103	65569159	2	AGTGAGTGAGTGTGTGTGTGGGG
	7	5	*COMDA*	+	13949	115434675	2	TGTGGGTGAGTGTGTGCGTGAGG
	14	22	*CYTH4*	+	-15670	37662824	2	GCTGAGTGAGTGTATGCGTGTGG
	2	14	*TMEM121 (abParts)*	+	36079	106029031	1	GGTGAGTGAGTGTGTGTGTGTGG
	8	5	*MIR3660*	-	-128431	89440968	3	AGAGAGTGAGTGTGTGCATGAGG
	2	12	*LEPREL2*	+	-482	6937055	3	GGTGGATGAGTGTGTGTGTGGGG
	2	3	*LINC00887*	-	36713	193993880	3	AGTGAATGAGTGTGTGTGTGTGG

No. of IS denotes number of IDLV integration sites within ± 60 bp of the CRISPR cleavage site; Chr. denotes the chromosome with the mapped CLIS; Gene denotes the host gene nearest to the CLIS; Strand denotes the orientation of the identified on/off-target sequence relative to that of the host chromosome; Distance denotes the on/off-target sequence to the transcription start site of the nearest host gene in base pairs. "-" indicates that the on/off-target sequence is upstream from the nearest gene; Coordinate denotes the precise chromosomal position of the identified on/off-target sequence; No. of Mismatches denotes the base mismatches from the on-target sequence. Base mismatches are labeled in red.

Supplementary Table 5.   Genome-wide analysis of the WAS CR-4 off-target sites in 15000 pooled puromycin-resistant colonies.

CRISPR	No. of IS	Chr	Gene	Strand	Distance	Coordinate	No. of Mismatch	Sequence
WAS CR-4	14	X	WAS	+	2400	48544585	0	TGGATGGAGGAATGAGGAGTTGG
	14	1	MATN1-AS1	+	-593748	30597870	1	TGGATGGAGGGATGAGGAGTGGG
	21	2	STK25	-	-2411	242451398	2	GGGATGGAGGGATGAGGAGTGGG
	26	20	GNRH2	+	-35133	2989134	2	GGGAGGGAGGAATGAGGAGTGGG
	7	18	OSBPL1A	+	41965	21810231	3	AGGAGGGAGGAATGGGGAGTTGG
	9	19	CD70	+	-20354	6611517	3	CGGACGGAGGAATGGGGAGTGGG
	2	12	HOXC13	+	-53412	54279163	3	ACGATGGAGGAATGAGGACTTGG
	9	2	MYCN	+	-144244	15936438	6	GAGGATGGGGAATGAGGAGTAGG
	3	2	TMEM247	+	18411	46725114	4	CCCATGGAGGAATGAGAAGTAGG
	2	13	RFC3	+	181207	34573412	4	TAAGTGGAGGAAAGAGGAGTAGG
	2	7	KCNH2	-	16457	150636460	3	AGCATGGAGGAATGAGGATTAGG
	2	X	SH2D1A	+	-54884	123425247	13	TGGGATGGAGGAATGAGGAGTGG

No. of IS denotes the number of IDLV integration sites within ± 60 bp of the CRISPR cleavage site; Chr. denotes the chromosome with the mapped CLIS; Gene denotes the host gene nearest to the CLIS; Strand denotes the orientation of the identified on/off-target sequence relative to that of the host chromosome; Distance denotes the on/off-target sequence to the transcription start site of the nearest host gene in base pairs. "-" indicates that the on/off-target sequence is upstream from the nearest gene; Coordinate denotes the precise chromosomal position of the identified on/off-target sequence; No. of Mismatches denotes the base mismatches from the on-target sequence. Base mismatches are labeled in red.

Supplementary Table 6. Top 50 potential off-target sites of WAS CR-4 with NGG or NAG as PAM.

Sequence	Score	Mismatches	Locus	Feature	% Indel
TGGATGGAGGGATGAGGAGTGGG	55.5	1 [11]	chr1:+30597853	MATN1-AS1	6.27
GGGATGGGGGGAATGAGGAGTCAG	7.1	2 [1:8]	chr22:+29583208		
GGGAGGGAGGAATGAGGAGTGGG	6	2 [1:5]	chr20:+2989117	GNRH2	7.14
GGGATGGAGGGATGAGGAGTGGG	4.8	2 [1:11]	chr2:-242451410	STK25	6.46
GGGATGGAGGGAGTGAGGAGTGAG	4.6	2 [1:12]	chr6:-166303465		
TGGAGGGAGGAATGAGGACTAAG	3.8	2 [5:19]	chr8:+98430874		
GGGTTGGTGGAATGAGGAGTAGG	2.6	3 [1:4:8]	chr12:+55440389	POT8	UD
GAGATGGTGGAATGAGGAGTAGG	2.6	3 [1:2:8]	chr8:-67261148	POT6	UD
AGGAAGGGGGAATGAGGAGTTAG	2.6	3 [1:5:8]	chrY:+14062575		
GGGAGGGAGCAATGAGGAGTAAG	2.5	3 [1:5:10]	chr2:+223365089		
GGGAGGGAGAAATGAGGAGTTAG	2.5	3 [1:5:10]	chr1:-185065591		
TGGATGGATGAATGAGGAGAGAG	2.4	2 [9:20]	chr19:-51192769		
GGGATGGAGGAATGACGAGTGGG	2.3	2 [1:16]	chr3:-193793878	POT3	UD
TGGATGGATGAATGAGTAGTGAG	1.8	2 [9:17]	chr5:+178869818		
TGGATGGATGAATGAGCAGTGGG	1.8	2 [9:17]	chr14:+101313659	POT1	UD
AGGAGGCAGGAATGAGGAGTGAG	1.7	3 [1:5:7]	chr1:+225646474		
TTGAGGGAAGAATGAGGAGTGAG	1.6	3 [2:5:9]	chr4:+146157572		
GGGAGGGAGGAATGAGGAGGGAG	1.5	3 [1:5:20]	chr19:-12964869		
GGGATGGTGGAATGAGGAGACGG	1.5	3 [1:8:20]	chr11:-124481404		
TGGGTGGCAGAATGAGGAGTCAG	1.5	3 [4:8:9]	chr5:-16965291		
AGGAGTGAGGAATGAGGAGTGAG	1.5	3 [1:5:6]	chr4:+3680374		
AGGAGTGAGGAATGAGGAGTGAG	1.5	3 [1:5:6]	chr4:+3680132		
AGGAGTGAGGAATGAGGAGTGAG	1.5	3 [1:5:6]	chr4:+3680058		
AGGAGTGAGGAATGAGGAGTGAG	1.5	3 [1:5:6]	chr4:+3679910		
TTTATGGAGGAATGAGGAGATGG	1.5	3 [2:3:20]	chr22:+22709608	POT9	UD
TCCATGGAGGAATGAGGAGAGGG	1.5	3 [2:3:20]	chr8:-48081104	POT10	0.27
TGGTAGGAGGTATGAGGAGTCAG	1.4	3 [4:5:11]	chr8:+27619085		
GGGAGGGAGGAGTGAGGAGTGAG	1.4	3 [1:5:12]	chr6:-166303487		
AGAATGGAGGAGTGAGGAGTTGG	1.4	3 [1:3:12]	chr9:-125689372		
CGGCAGGTGGAATGAGGAGTGGG	1.4	4 [1:4:5:8]	chr19:+30379571		
GGGGAGGGGGAATGAGGAGTGAG	1.4	4 [1:4:5:8]	chr9:+134697289		
ACGGTGGGGGAATGAGGAGTTGG	1.4	4 [1:2:4:8]	chr11:-93105683		
GGGTAGGTGGAATGAGGAGTCAG	1.4	4 [1:4:5:8]	chr5:+65269522		
TGGGTGGAGAGATGAGGAGTTAG	1.3	3 [4:10:11]	chr6:-98167734		
ATGGGGGAGGAATGAGGAGTTAG	1.3	4 [1:2:4:5]	chr7:+95650322		
CAGATGGGGCAATGAGGAGTTAG	1.3	4 [1:2:8:10]	chr10:+17647092		
GGGTGGGAGAAATGAGGAGTGAG	1.3	4 [1:4:5:10]	chr10:+1114984		
AGGAAGGGGAAATGAGGAGTTAG	1.3	4 [1:5:8:10]	chr13:-38303999		
GAAAGGGAGGAATGAGGAGTTAG	1.3	4 [1:2:3:5]	chr11:+114744341		
CTTAAGGAGGAATGAGGAGTTAG	1.3	4 [1:2:3:5]	chr9:-1087222		
AGACGGGAGGAATGAGGAGTAGG	1.3	4 [1:3:4:5]	chr11:-109738999	POT17	UD
GGTATGGGGAAATGAGGAGTTAG	1.3	4 [1:3:8:10]	chr8:+79906546		
GCTATGGAGAAATGAGGAGTCAG	1.3	4 [1:2:3:10]	chr6:+98157546		
TCTTGGGAGGAATGAGGAGTAGG	1.3	4 [2:3:4:5]	chr2:-240313186	POT21	UD
TCTCAGGAGGAATGAGGAGTTGG	1.3	4 [2:3:4:5]	chr2:+199886049	POT20	0.04
TAAATGGGGAAATGAGGAGTCAG	1.3	4 [2:3:8:10]	chr2:-53843685		
TGGGAGGTGTAATGAGGAGTGGG	1.3	4 [4:5:8:10]	chr14:-84980650		
TGGTGGGGGAAATGAGGAGTTAG	1.3	4 [4:5:8:10]	chr11:-31445159		
GGGCTGGAGGAAGGAGGAGTTGG	1.2	3 [1:4:13]	chr1:+65288699		

Sequence	Score	Mismatches	Locus	Feature	% Indel

Sequence denotes the POT site with NGG or NAG as PAM. NAG is labeled in red. The table is ranked by scores generated with CRISPR Design Tool at http://crispr.mit.edu/; Mismatches denote the base mismatches between the WAS CR-4 target site and each POT site. Numbers in the bracket indicate the positions of the mismatches with position 1 at the distal end from PAM; Locus denotes the chromosome and the coordinate of each POT site. "+" and "-" refer to the orientation of each POT site relative to that of the chromosome; Feature indicates those off-target sites confirmed by deep sequencing; %indel denotes the frequency of indel determined by deep sequencing; UD, undetectable.

Supplementary Table 7. Primers used for the Surveyor assay

CRISPR or TALEN	Gene	Forward Primer	Reverse Primer	*Expected length (bp)	**Peak 1 (bp)	**Peak 2 (bp)
WAS CR-4	*WAS*	AAAGGAAGTTGGGCAGAGGTGAGT	CCATCGATTGTGTGTTGGATGGTCATGGAGGT	838	500	338
WAS CR-5	*WAS*	AAAGGAAGTTGGGCAGAGGTGAGT	CCATCGATTGTGTGTTGGATGGTCATGGAGGT	838	539	299
WAS CR-3	*WAS*	AAAGGAAGTTGGGCAGAGGTGAGT	CCATCGATTGTGTGTTGGATGGTCATGGAGGT	838	534	304
TAT CR-4	*TAT*	TGGGGACACTACTGAGGGGCTG	TCCCGAGACCCGGTTCCCAA	414	309	105
TAT CR-1	*TAT*	TGGGGACACTACTGAGGGGCTG	TCCCGAGACCCGGTTCCCAA	414	276	138
TAT CR-6	*TAT*	TGGGGACACTACTGAGGGGCTG	TCCCGAGACCCGGTTCCCAA	414	251	163
TAT CR-1	*PHACTR1*	TCCTGGCTTCATGCCAAAGT	TGAAGAAACGGACTGCTCCC	689	400	289
WAS CR-4	*MATN1-AS1*	GGACTTCCCCAAATCTGGCA	GGGCCTTACACTCCCCTTTC	670	420	250
WAS CR-4	*OSBPL1A*	AGCATGAAGGGAAGAGGTGC	TAACGACAGGCTGCCATTGT	523	140	383
WAS CR-4	*MYCN*	TGTCCAGTTTCTGCTCCCAC	ACCCAGCTCAAAGCCTCTTC	635	420	215
WAS CR-4	*Stk25*	GCAGGTGAGTGCCACAAATG	GAATCCAAGAGCCTGCGGTA	653	464	189
WAS CR-4	*GNRH2*	GCTGGCAAAAGCAAGTCTGA	ACACTAATCTCTCCATCAGTGTCC	899	383	516
WAS CR-4	*HOXC13*	AGGCCTGAGGTTTTGCATGA	GGGGATACAGGGTGAGGACT	615	366	249
WAS CR-4	*CD70*	TGGAACTCCGATGCTCCAAG	GGGCATATCTCAGAAAGTCAGC	624	387	237
WAS CR-5	*PABPC5*	AGTCAGCATGCAGATTCTGGT	ATGGTGTGGCCCAGAAAGTC	622	350	272
WAS TALEN	*WAS*	AAAGGAAGTTGGGCAGAGGTGAGT	CCATCGATTGTGTGTTGGATGGTCATGGAGGT	838	519	319
miR-9-2 TALEN	*miR-9-2*	TCCTGGACGACCACTCTTCGGT	GCAGCTGCAACAACCCCTCTCA	373	221	152
TAT TALEN	*TAT*	TGGGGACACTACTGAGGGGCTG	TCCCGAGACCCGGTTCCCAA	414	290	124
ZRSR2 TALEN	*ZRSR2*	TTGTAGCCGCTGATCGTCGTGTAT	ACTACGTGTGCAAGGACTGTGCTA	423	310	113