

Additional file 4: Oligos used for the cloning of pTARGET and RGEN plasmids and PCR analysis of putative transgenic regenerants.

Primer	Direction	Sequence 5' – 3'	Primer binding site
RGEN-gfp target barley	forward	<u>GATC</u> GTCTTTGCTCAGGCGGACTGGGC	Bp 611-633 of <i>gfp</i> gene, complementary strand
	reverse	<u>AATTG</u> CCCAGTCCGCCCTGAGCAAAGAC	Bp 611-633 of <i>gfp</i> gene
TALEN-gfp target barley/tobacco	forward	<u>GATC</u> TGG TGAACCGCATCGAGCTGAAGGGCATCGACTTCAAGGAGGACGGCAA GTG	Bp 362-407 of <i>gfp</i> gene
	reverse	<u>AATT</u> CAC TTGCCGTCCTCCTTGAAGTCGATGCCCTTCAGCTCGATGCGGTTCA CCA	Bp 362-407 of <i>gfp</i> gene, complementary strand
TALEN-MLO target barley	forward	<u>GATC</u> GCT GGAACACGGCCTCCACAAGCTCGGCCATGTAAGTCCC GTTACCCTAGCTC AG	Bp 100-151 of <i>MLO</i> gene
	reverse	<u>AATT</u> CT GAGCTAGGGTAACGGGACTTACATGGCCGAGCTTGTTGGAGGCCGTGTCC AGC	Bp 100-151 of <i>MLO</i> gene, complementary strand
Sfil-A_f	forward	GTAGGCCCTTAAGGCCATG	Adapter sequence
Sfil-A_r	reverse	CATGGCCTTAAGGCCCTAC	Adapter sequence, complementary strand
Sfil-B_f	forward	GTACGTAAGGCCGCGCCATG	Adapter sequence
Sfil-B_r	reverse	GTACCATTGGCCATGGCGGCTTAC	Adapter sequence, complementary strand
GFP_PP1_f	forward	GGCGTCTTTGCTCAGGGCGGACT	Bp 611-633 of <i>gfp</i> gene
GFP_PP1_r	reverse	AAACAGTCCGCCCTGAGCAAAGA	Bp 611-633 of <i>gfp</i> gene, complementary strand
RGEN-gfp tobacco	forward	<u>ATTG</u> GCTGAAGGGCATCGACTTCA	Bp 375-394 of <i>gfp</i> gene
	reverse	<u>AACT</u> GAAAGTCGATGCCCTTCAGC	Bp 375-394 of <i>gfp</i> gene, complementary strand
Cas9 F2	forward	CAGCTCGTGCAGACCTACAAC	Bp 559-579 of <i>Cas9</i> gene
Cas9 R2	reverse	TGCCTTCTAAGGATAGCGTG	Bp 1258-1277 of <i>Cas9</i> gene, complementary strand
AtU6-26 F1	forward	CAGCTAGAGTCGAAGTAGTG	Bp 365-384 of <i>AtU6</i> promoter tobacco
35S P R1	reverse	GACAGATAGCTGGGCAATGG	Bp 133-152 of <i>CaMV 35S</i> promoter, complementary strand
35S P F1	forward	GACGTAAGGGATGACGCAC	Bp 350-368 of <i>CaMV 35S</i> promoter
BAR R2	reverse	GAGACGTACACGGTCGACTC	Bp 262-281 of <i>BAR</i> gene, complementary strand
GH-GFP R2	forward	TACGGCAAGCTGACCCTGAA	Bp 118-137 of <i>gfp</i> gene
GH-GFP F1	reverse	GGTCACGAACTCCAGCAGGA	Bp 659-678 of <i>gfp</i> gene, complementary strand
FokI F1	forward	ATCGAGATCGCCGGAACAGCACC	Bp 13-36 of <i>FokI</i> gene
FokI R	reverse	ATCATCTCGCCCGCATCAGGAGC	Bp 429-452 of <i>FokI</i> gene, complementary strand
GH-Ubi10 F	forward	CTATTGCTTACCGCCTTAG	Bp 1010-1029 of <i>AtUBIQUITIN-10</i> promoter tobacco
TALEN R2	reverse	TGGCGGCTTGGCGCTGACAG	Bp 32-52 of <i>gfp</i> -specific right TALEN unit Bp 59-79 of <i>gfp</i> -specific left TALEN unit
GH-MLO-F3	forward	ACATCGGTCGTTTCTCACTC	Bp -396-206 of <i>MLO</i> gene
GH-MLO-R1	reverse	ACGCGCACGCCGATTACAAC	Bp -396-206 of <i>MLO</i> gene, complementary strand

Underlined are restriction enzyme recognition sites or respective overhangs and red font are the nucleotides to introduce frameshift in *yfp* gene of target vector