

Table S3. Genotype of 13 kDa *prolamin*-knockout lines (T₁ generation)

sgRNA	T ₁ plants	Transgenic assay ^a	Edited gene	Frequency ^b	Type	Sequencing results
		<i>SpCas9/Hyg^R</i>				
sgRNA-pro13a	WT	-/-			WT	TTGAAACGTAGCTGGTTGCCA...GAA GGG GGTT
	1a-6	-/-	<i>Pro13a.1</i>	99.9%	Ins (1 Nu)	***** A *****
			<i>Pro13a.2</i>	50.0%	Ins (1 Nu)	***** A *****
	1a-8	-/-	<i>Pro13a.1</i>	99.9%	Ins (1 Nu)	***** A *****
			<i>Pro13a.2</i>	99.8%	Ins (1 Nu)	***** A *****
	1a-10	-/-	<i>Pro13a.1</i>	99.9%	Ins (1 Nu)	***** A *****
			<i>Pro13a.2</i>	0.1%	WT	***** ...
	1a-11	-/-	<i>Pro13a.1</i>	99.9%	Ins (1 Nu)	***** A *****
			<i>Pro13a.2</i>	48.9%	Ins (1 Nu)	***** A *****
	2a-1	-/-	<i>Pro13a.1</i>	50.6%	Del (1 Nu)	***** - ...*****
	2a-2	-/-	<i>Pro13a.1</i>	99.8%	Del (1 Nu)	***** - ...*****
	2a-6	-/-	<i>Pro13a.1</i>	0.0%	WT	***** ...
	2a-7	-/-	<i>Pro13a.1</i>	50.8%	Del (1 Nu)	***** - ...*****
	2a-9	-/-	<i>Pro13a.1</i>	52.1%	Del (1 Nu)	***** - ...*****
sgRNA-pro13b	WT	-/-			WT	GTTGAAACGCAGCTGATTGCA...AGA AGG TGCT
	4b-1	+/+	<i>Pro13b.1</i>	49.3%	Ins (1 Nu)	***** A *****

4b-2	+/+	<i>Pro13b.1</i>	50.6%	Ins (1 Nu)	***** *****A*****
4b-3	-/-	<i>Pro13b.1</i>	0.0%	WT	***** *****...
4b-5	+/+	<i>Pro13b.1</i>	50.2%	Ins (1 Nu)	***** *****A*****
4b-7	+/+	<i>Pro13b.1</i>	49.9%	Ins (1 Nu)	***** *****A*****
4b-9	-/-	<i>Pro13b.1</i>	99.4%	Ins (1 Nu)	***** *****A*****
		<i>Pro13b.13</i>	99.8%	Ins (1 Nu)	***** *****A*****
4b-10	+/+	<i>Pro13b.1</i>	49.4%	Ins (1 Nu)	***** *****A*****
8b-3	-/-	<i>Pro13b.3</i>	100.0%	Del (4 Nu)	***** *****- - - - *****...
8b-6	-/-	<i>Pro13b.3</i>	0.1%	WT	***** *****...
8b-11	-/-	<i>Pro13b.3</i>	49.9%	Del (4 Nu)	***** *****- - - - *****...

^aSymbol '+' and '-' indicate that SpCas9/Hyg^R genes are present or absent, respectively

^bIndel (Insertion & Deletion) frequency was calculated from Deep sequencing analysis

Red color dash and letter indicate the deletion and insertion of nucleotide, respectively

Green letters indicate the PAM sequence

Asterisks indicate the nucleotides with no change