SUPPLEMENTAL TABLES AND FIGURES

Table S1. gRNA targeting sequences to introns 1 and 6. The score indicated was calculated by the Zhang Lab algorithm for cutting efficiency and offsite targeting [1].

Target Location	Sequence	Score
Intron 1	AAAACATAGCAGGGATCCGC	84
Intron 1	ACAGAAGGGGTCTTTCAATC	78
Intron 1	CCAGTGCTCAGCAATTATGG	76
Intron 6	TAGCCGTGCTTCGTATTTCA	85
Intron 6	ATTCTTTTTAGGAGGCACGT	80
Intron 6	ACTCCTTGAAATACGAAGCA	79

Table S2. Genotyping and sequencing primers

Primer	Sequence	PCR fragment length, bp
Forward 1, common	TGCAGGCTAACGTTTTCTCA	
Reverse 1, *Col1a2*^{GFP}	AGTCGTGCTGCTTCATGTGG	648
Reverse 2, *Col1a2*	CCTCCCCAGAAATGACACC	517
Reverse 3, *Col1a2* indels	CCCGGATTCTCCTGTTATGA	1611 (GFP), 4932 (uncut), ~470 (exon 2-6 excision)
*Forward 2 for Reverse 3	TTGGGGGAAAGGGCTAAGTC	4641 (GFP), 7962 (uncut), ~2500 (exon 2-6 excision)

*Forward 2 primer was used in early genotyping experiments (Fig. S1b)

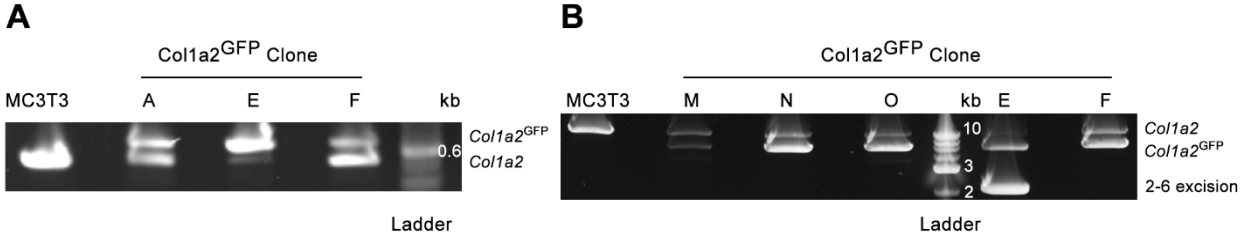

Fig.S1. Sample genotyping gels. **a** 2:1:1 mix of Forward-1:Reverse-1:Reverse-2 primers. **b** 1:1 mix of Forward-2:Reverse-3 primers. Clone M did not survive freezing; clones N and O did not mineralize and were discarded. Bottom band in clone E results from exon 2-6 excision.

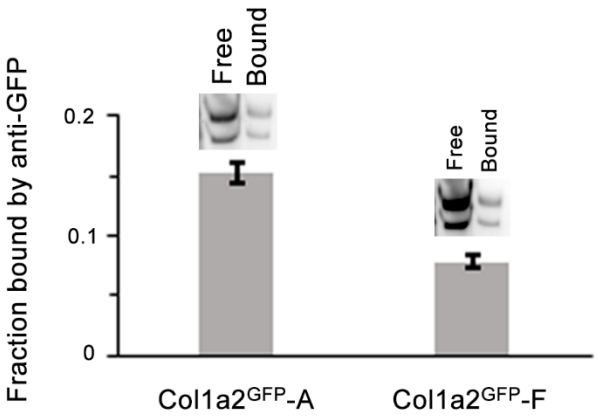

Fig. S2. Fraction of secreted procollagen containing the GFP-proα2(I) chain. Secreted procollagen was precipitated with 0.178 mg/ml ammonium sulfate, resuspended in bead buffer (10 mM Tris/Cl pH 7.5, 0.5 M NaCl, 0.5 mM EDTA) and incubated with anti-GFP agarose beads (Chromotek). After separating the free and bound fractions, collagen triple helices were isolated by pepsin treatment and selective NaCl precipitation, labeled by Cy5 and quantified by SDS-PAGE (insets). Error bars represent standard error of the mean in 3 biological replicates.

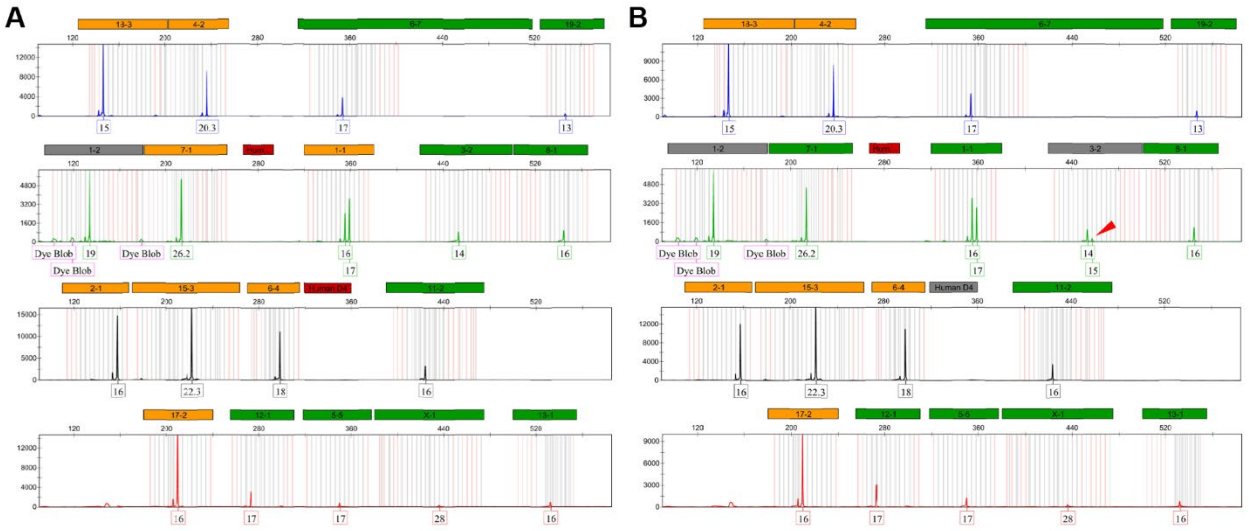

Fig. S3. STR profile reports from ATCC. **a** Col1a2^{GFP}-A, 100% identical to MC3T3-E1 (subclone 4). **b** Col1a2^{GFP}-F, 97% identical to MC3T3-E1 (subclone 4) and clone A. Red arrowhead shows a small satellite STR allele peak at locus 3-2, which distingishes this clone.

REFERENCES

1. Liu G, Zhang Y, Zhang T (2020) Computational approaches for effective CRISPR guide RNA design and evaluation. Computational and Structural Biotechnology Journal 18:35-44. https://doi.org/10.1016/j.csbj.2019.11.006