Supplementary Material

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Fig. S1. Photographs of tadpoles and frogs that were raised from TALEN-mRNA-injected embryos. Embryos injected with $\Delta N\Delta C$ -ELD/KKR-Tyr I mRNAs were reared to albino tadpoles (A) and frogs (B). The white arrowheads indicate a wild-type tadpole (A) and a wild-type frog (B) that were bred from uninjected embryos. The inset shows the higher magnification of a right eye of an albino frog (B).



Fig. S2. Gel electrophoresis images of genomic PCR products from TALEN-mRNA-injected embryos after restriction enzyme digestion. Embryos were injected with 400 pg or 80 pg of TALEN-Tyr I mRNAs (A) and 400 pg (B) or 80 pg (C) of TALEN-Tyr II mRNAs. Genomic DNA from each embryo was prepared and subjected to PCR using a specific primer set to amplify a DNA fragment containing the target sites. The PCR products were digested using HaeIII (A) or PfIMI (B,C). The injected TALEN scaffolds and doses of mRNA are shown at the top and bottom of the gel images, respectively. These images show the result from a portion of the embryos analyzed in Fig. 4.

Α	Tyr I Left	HaellI GGCC	Tyr I Right	
	CTGTTCTTCTTCCTCCATGT CTGTTCTTCTTCTCCTCCATGT CTGTTCTTCTTCCTCCATGT CTGTTCTTCTTCTCCTCCATGT CTGTTCTTCTTCTCCTCCATGT CTGTTCTTCTTCTTCCTCCATGT	TTGCAGGGGCCAGTT TTGCA - GGCCAGTT TTGCA - GGCCAGTT TTGC - CAGTT TTG - CAGTT TTG-CAG	CCCAAGGGCA TGTAGCACCG CCCAAGGGCA TGTAGCACCG CCCAAGGGCA TGTAGCACCG CCCAAGGGCA TGTAGCACCG CCCAAGGGCA TGTAGCACCG GGGCA TGTAGCACCG - CCAAGGGCA TGTAGCACCG	$\begin{array}{ccc} WT & x \ 3 \\ \Delta 1 & x \ 2 \\ \Delta 2 & x \ 3 \\ \Delta 6 & x \ 1 \\ \Delta 7 & x \ 4 \\ \Delta 14 & x \ 1 \\ \Delta 107 & x \ 1 \end{array}$

	PfIMI			
Tyr II Left	CCANNNNNTGG	Tyr II Right		
ACTGGCCCTCAGTT	TCCATTCACTGGG	GTTGACGATAGAGAGAACTGGCCAAC	WT	
ACTGGCCCTCAGTT	TCCATTCTCT	TGACGATAGAGAGAACTGGCCAAC	Δ5	X 1
ACTGGCCCTCAGTT	TCCATTCTCTGG-	CGATAGAGAGAACTGGCCAAC	$\Delta 6$	x 1
ACTGGCCCTCAGTT	TCCATTCTCTG	ACGATAGAGAGAACTGGCCAAC	Δ6	x 3
ACTGGCCCTCAGTT	TCCATT	GACGATAGAGAGAACTGGCCAAC	Δ10 Δ11	x 1
ACTGGCCCTCAGTT	TCCATTCTCTGGG	GAGAACTGGCCAAC	Δ12	x 1
ACTGGCCCTCAGTT	ТССА	CGATAGAGAGAACTGGCCAAC	Δ14	x 1
ACTOCCCCTCACTT		ACGATAGAGAGAACTGGCCAAC	Δ96 Δ407	x 1
ACTGGCCCTCAGTT	TCCATTCT	- A TGACGA TA GAGAGAAC TGGCCAAC	Δ7+1	x1
ACTGGCCCTCAGTT	TCCATTCTCTGGA	ATAGAGAGAACTGGCCAAC	∆8+1	x 1
ACTGGCCCTCAGTT	TC <mark>AG</mark>	TAGAGAGAACTGGCCAAC	∆19 +2	x 1

Fig. S3. Mutated target sequences in TALEN-mRNAinjected embryos. Target DNA sequences were determined using genomic DNA purified from an NF-stage 35/36 embryo that had been injected with 400 pg of Δ N Δ C-ELD/KKR-Tyr I mRNAs (A) or -Tyr II mRNAs (B). The wild-type sequence is shown at the top. The black bars indicate the Tyr I (A) and Tyr II (B) target sites. Gaps resulting from deletion are denoted as dashes. Inserted nucleotides are indicated as red characters. The HaeIII (A) and PfIMI (B) recognition sequences are indicated as blue characters. The mutation types and frequencies are indicated on the right.

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