

Title: Generation of a conditional Flpo/FRT mouse model expressing constitutively active TGF β in fibroblasts

Authors:

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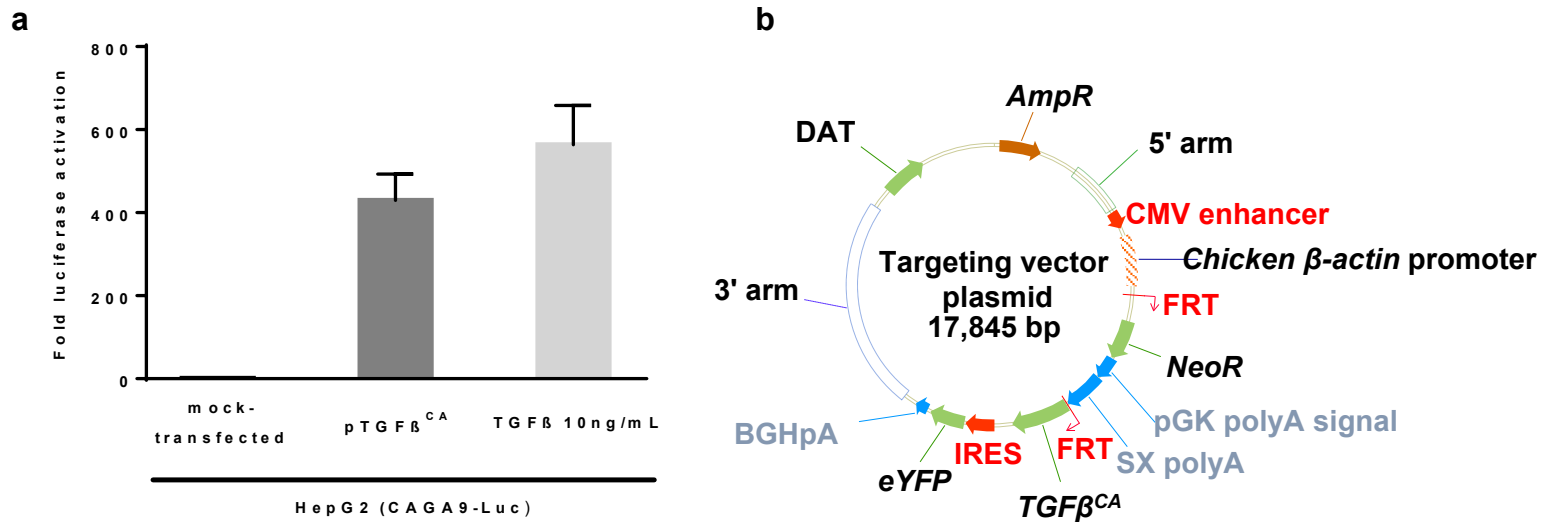


Fig. S1: Construction of the ^{FSF}TGF β^{CA} targeting homologous recombination vector.

a) Luciferase assay in HepG2 cells transiently transfected with a TGF β -sensitive reporter (CAGA9-luc) in the presence or not of TGF β (positive control), and, a-co-transfected with a plasmid expressing TGF β^{CA} (pTGF β^{CA}).

b) Circular map of the ^{FSF}TGF β^{CA} transgenesis vector. DAT, Dopamine Transporter ; AmpR, ampicillin antibiotic resistance cassette; CMV enhancer, human cytomegalovirus enhancer and chicken β -actin promoter; NeoR, neomycin antibiotic resistance cassette; pGK, (phosphoglycerate kinase I) polyadenylation signal; TGF β^{CA} sequence; IRES, internal ribosome entry site; eYFP, enhanced yellow fluorescent protein; bGH polyA, bovine growth hormone polyadenylation signal.

5' homology arm (1.1 kb)

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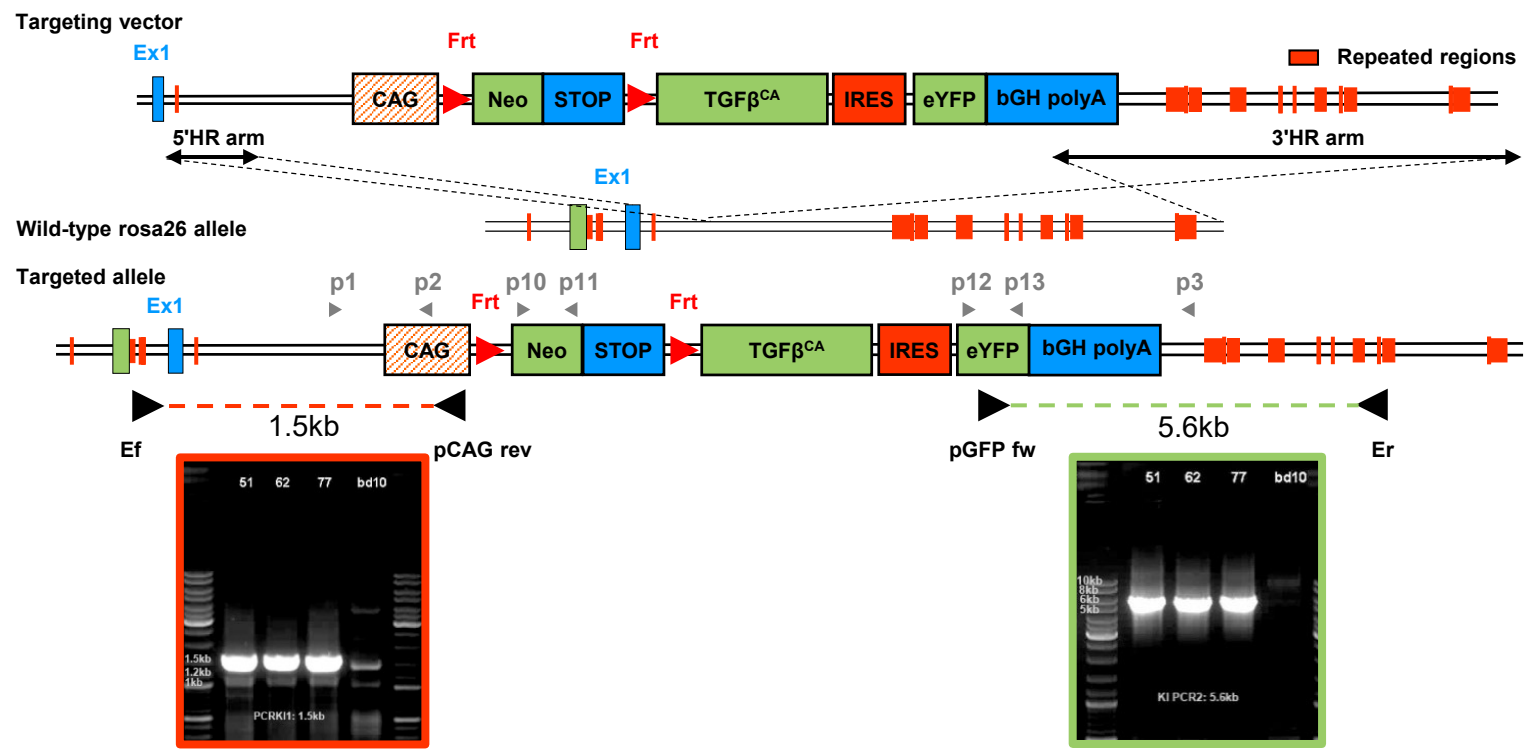
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 cgcgcgtaatacagactactataggcgcaattggagctcc

Fig. S2: Verified sequence of the *FSFTGFβ*^{CA} transgene cloned inside the recombination vector.**

Result of sequencing. 5' homology arm - CMV (human cytomegalovirus) Enhancer/Chicken β-actin promoter - FRT-
 neomycin antibiotic resistance cassette-FRT- *TGFβ*^{CA} sequence/IRES (internal ribosome entry site) - eYFP (enhanced
 yellow fluorescent protein) - BGHpA (bovine growth hormone polyadenylation signal) - 3' Homology arm, Vector
 backbone sequence. Primers sequences used for PCR and RT-PCR are underlined.

a

PCR screening strategy



b

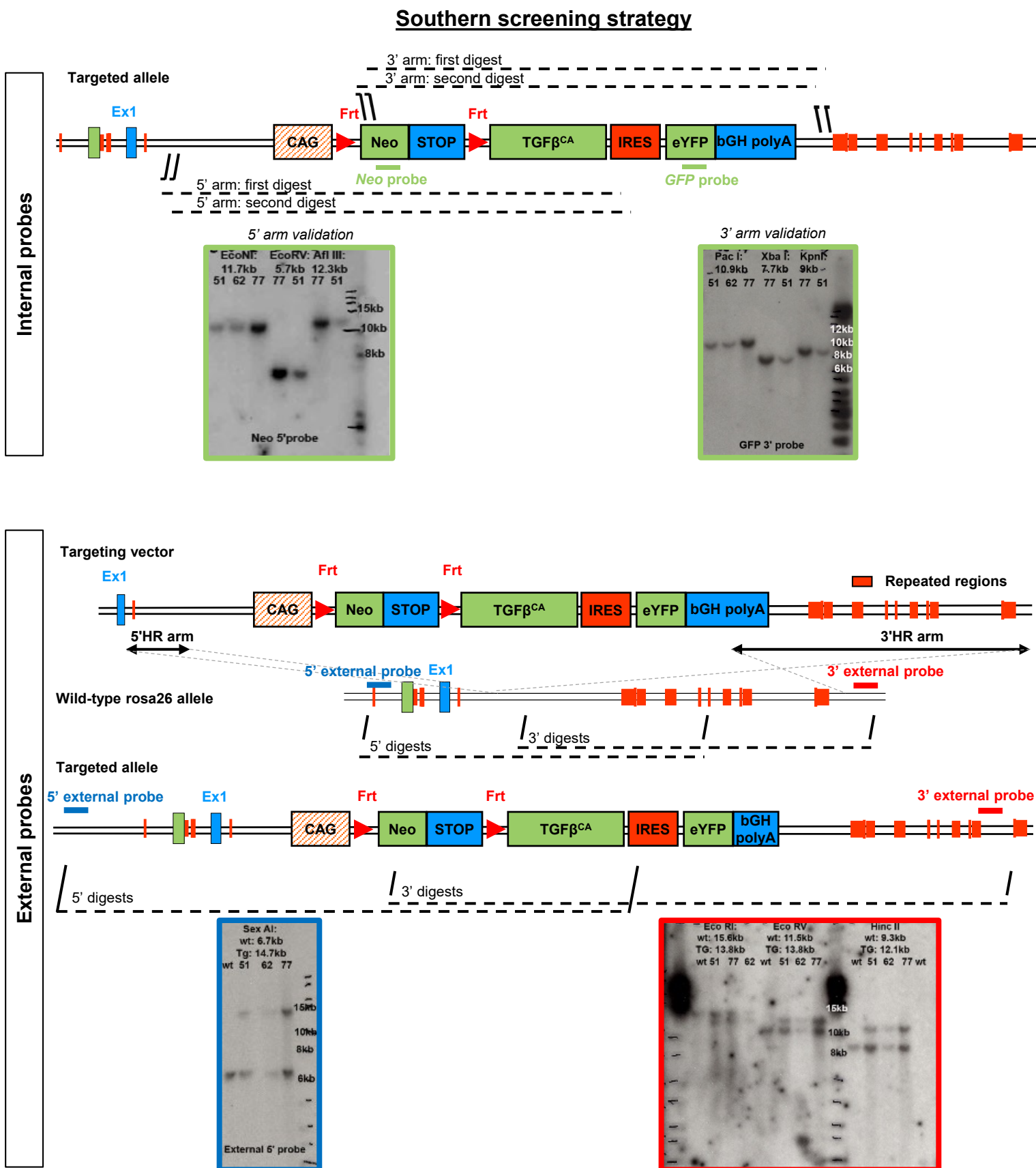


Fig. S3: Validation of *FSFTGFβ^{CA}* ES clones.** a) PCR screening strategy using 2 different set of primers (Ef/pCAG rev and pGFP fw/Er). b) Southern blot screening strategy. Top panel, 2 internal probes (Neo and GFP) and 6 different digests are used to validate correct HR event. 3 digests validate the 5' insertion, 3 other digests validate the 3' insertion. Bottom panel, 2 external probes (5' and 3'). 1 digest was used with 5' probe and 3 different digests were used with 3' probe. HR, Homologous Recombination.

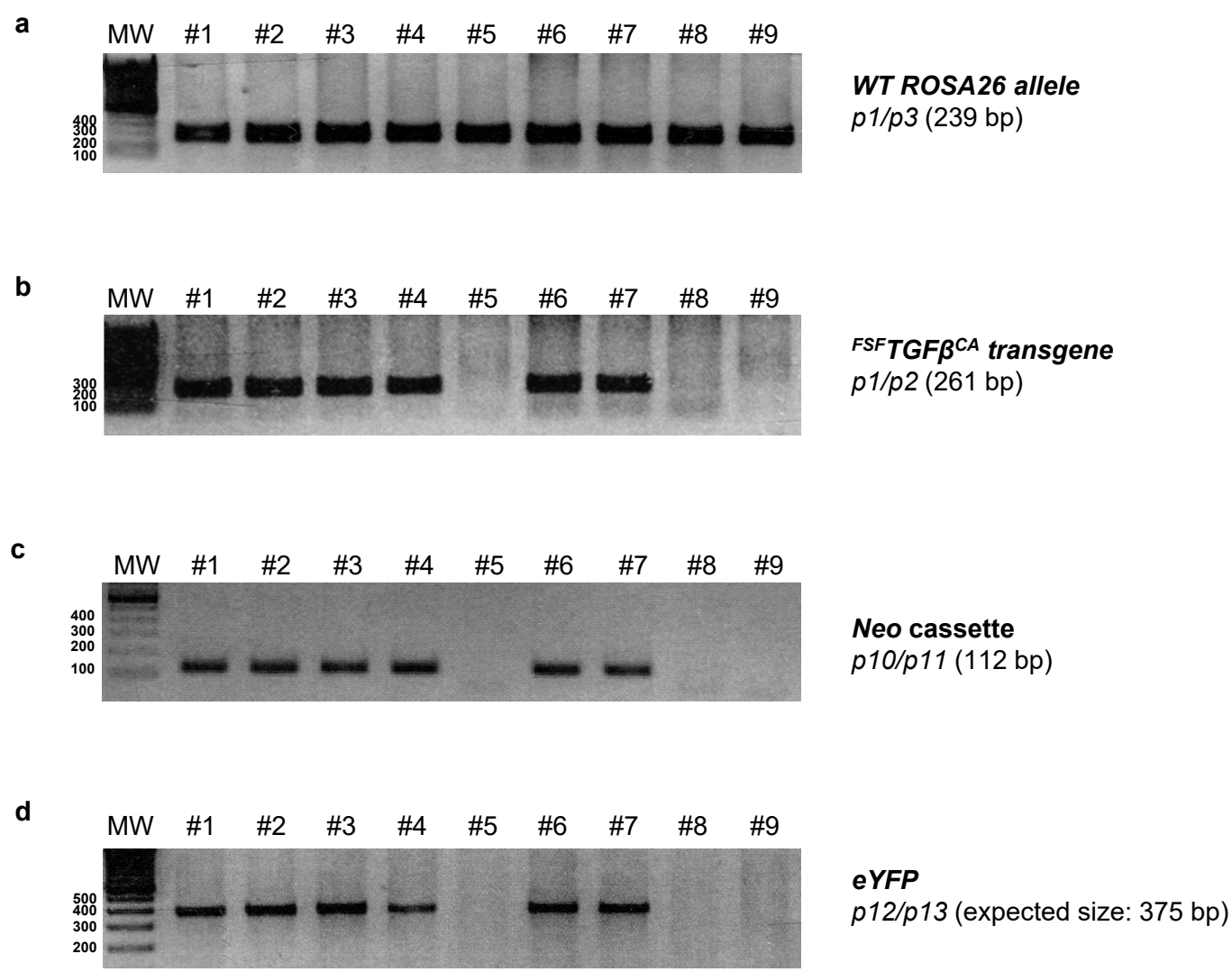


Fig. S4: Genotyping of F1 [*FSFTGFβ^{CA}*] heterozygous individuals. Primers are positioned on the sequence in Fig. S3a. a) *ROSA26* wild-type allele genotyping. b) *CAG* promoter genotyping. c) *Neo* transgene genotyping. d) *eYFP* transgene genotyping. In a, b, c and d, the name of primers are indicated on the right of each gel. Primers sequences are shown in table 1.