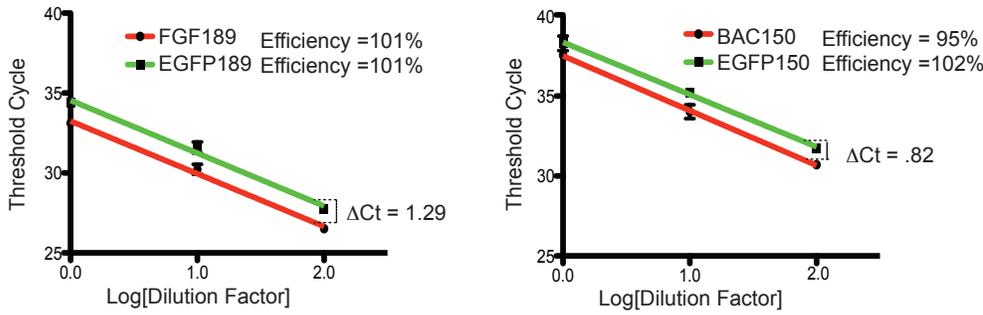


A



B

EGFP, fin clip, 9 months, 15 of 15 Sequences Mutant

```

ATGGTGAGCAAGGGCGAGGAGCTGTTACCGGGGTGGTGCCCATCTGGTTCGAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA  EGFP  X 0
ATGGTGAGCAAGGGCGAGGAGCTGT-----AAA (-65) X 8
ATGGTGAGCAAGGGCGAGGAGCTGTTTAC-----GGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-27)
ATGGTGAGCAAGGGCGAGGAGCTGTTTAC-----TGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-26)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCC-----GCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-12)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCA-----GCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-10)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCAT-----GCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-9)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCAAC-----AGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (+2, -9)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCATCTGGGGTGT-----GGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (+5, -7)
  
```

EGFP, fin clip, 9 months, 18 of 18 Sequences Mutant

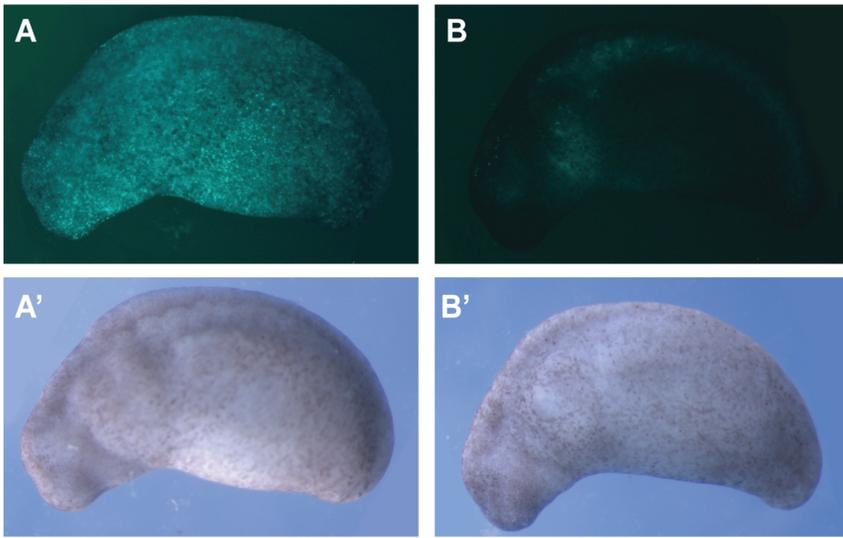
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ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCATCTGGTTCGAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA  EGFP  X 0
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ATGGTGAGCAAGGGCGAGGAGGCTG-----AAACGGCCACAAGTTCAGCGTGAAA (-43) X 2
ATGGTGAGCAAGGGCGAGGAGCTGTTTAC-----TGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-26)
ATGGTGAGCAAGGGCGAGGAGCTGTTTAC-----TCGAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-22)
ATGGTGAGCAAGGGCGAGGAGCTGTTTAC-----GTCGAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-19) X 2
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCATCTGGT-----GACGTAAACGGCCACAAGTTCAGCGTGAAA (-13)
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCAT-----GACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-13) X 3
ATGGTGAGCAAGGGCGAGGAGCTGTTTACCGGGGTGGTGCCCA-----GCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGAAA (-10) X 7
  
```

C



Supplementary Figure 1. (A). Two standard curves created from Real-Time PCR of EGFP transgene and size-matched control genomic DNA sequences from Tg(CAGG:EGFP) transgenic animal indicate that this line contains a single copy of the transgene. Under amplification conditions of near 100% efficiency, signal from EGFP reaches the threshold (Ct) one cycle behind sized-match control regions. (B) Sequencing of PCR products from animals depicted in Figure 1D and 1E reveal multiple indel events at targeted site in EGFP transgene. (C) At 9 months, animals depicted in Figure 1D and E (middle and right, respectively) next to an uninjected EGFP-positive (left) sibling.



Supplementary Figure 3. At 5 days post fertilization, variability in RGN-induced mutagenesis, corresponds to variability in expression of coinjected *nls-EGFP* mRNA. Embryos coinjected with *nls-EGFP* and *bambi*-directed RGN display uniform expression of EGFP (A), others display very low-level mosaic EGFP expression (B).