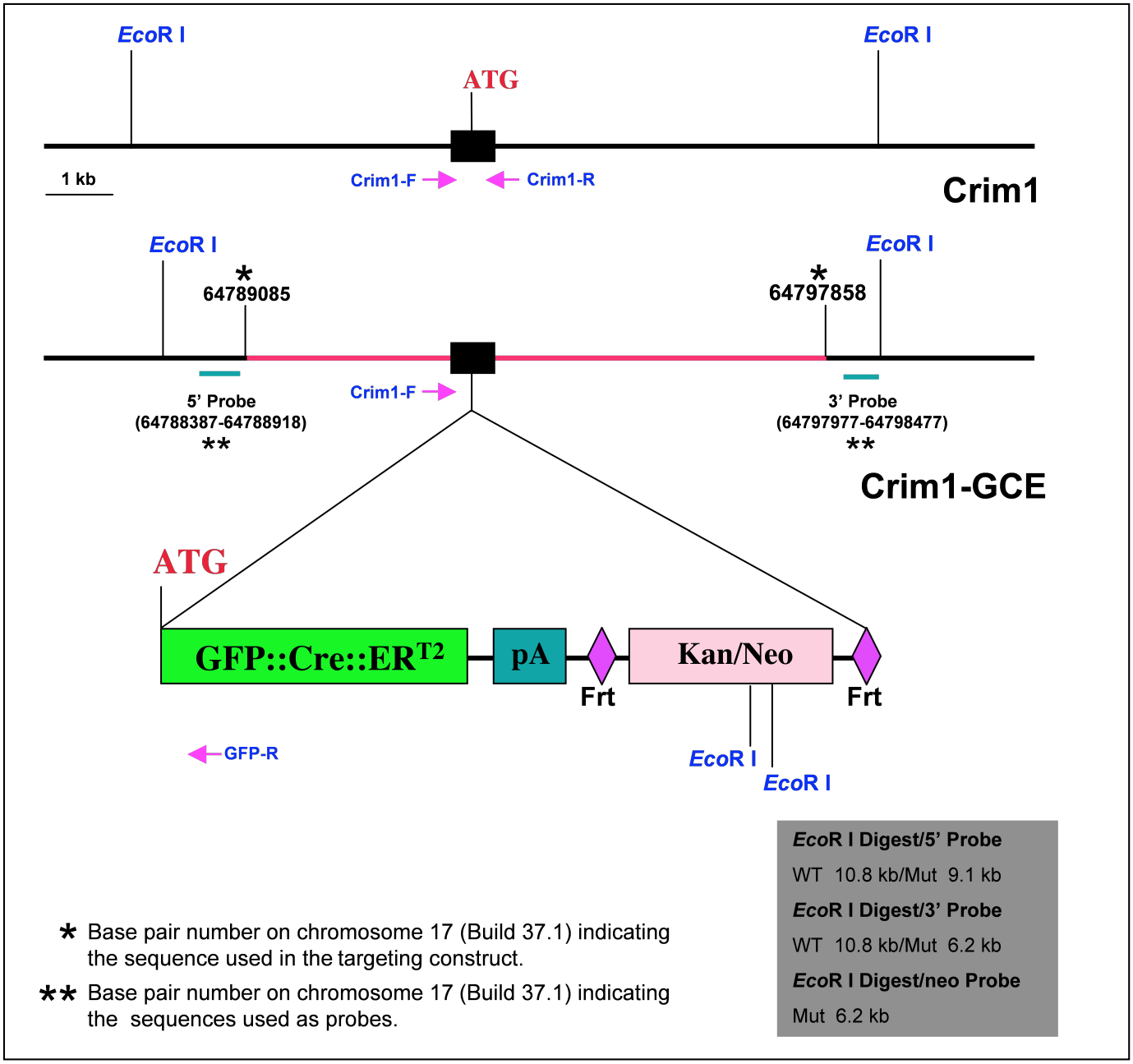
Crim1-GCE

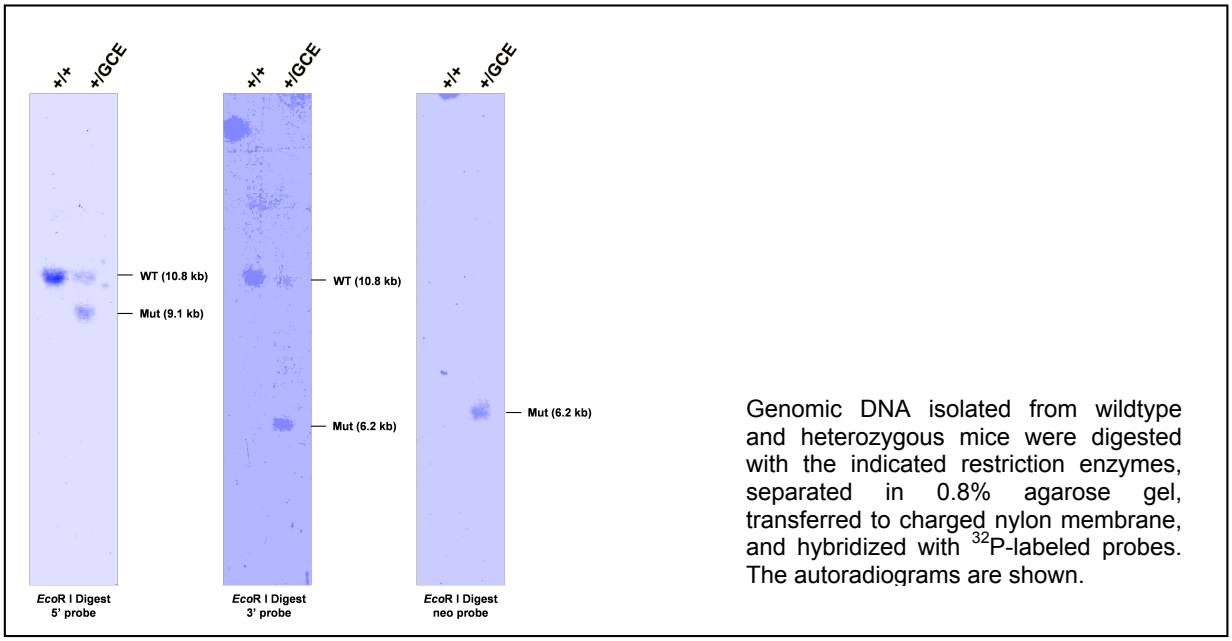
A. Rationale

Crim1, Cysteine rich transmembrane BMP regulator 1 (chordin-like) is expressed in a population of cells in the adrenal gland and kidney vasculature and may be involved in endothelial cell maintenance and integrity.
A strain of mice carrying eGFPCreERT2 knocked into the Crim1 locus was generated by the GUDMAP consortium to investigate microvasculature development.

B. Targeting Strategy



C. Southern Blot Analysis of the Targeted Allele in Mice



Genomic DNA isolated from wildtype and heterozygous mice were digested with the indicated restriction enzymes, separated in 0.8% agarose gel, transferred to charged nylon membrane, and hybridized with ^{32}P-labeled probes. The autoradiograms are shown.

D. PCR Genotyping

a. Primers

Crim1-F: 5' gtcgtctttccccggcaatc 3'
Crim1-R: 5' cttcgcagacgcccacttc 3'
GFP-R: 5' gtccagctcgaccaggatgg 3'

b. Expected Band Sizes

Crim1-F + Crim1-R: 742 bp
Crim1-F + GFP-R: 471 bp

D. Relevant Sequences

a. Genomic clone used for targeting construct

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Crim1-R

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b. The final construct (excluding plasmid backbone and the negative selection marker)

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Crim1-F

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Putative start of exon 1

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Start of GFP

GFP-R

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End of GFP

Start of Cre

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End of Cre

Start of ER

CATGATCAAACGCTCTAAGAAGAACAGCCTGGCCTTGTCCCTGACGGCCGACCAGATGGTCAGTGCCTTGTTGGATGCTGA
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End of ER

Start of SV40 polyA

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End of SV40 polyA
Frt
ataggaacttc aggtctgaagaggagtttacgtccagccaagctagcttggctgcaggtcgtggtacgaaattctaccgg
Start of Kan/Neo
gggaggcgcttttcccaaggcagtctggagcatgcgctttagcagccccgctgggcacttggcgctacacaagtggcctct
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End of Kan/Neo
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Frt
Crim1 continues
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Crim1-R
Exon/intron junction
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5′ probe

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3′ probe

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