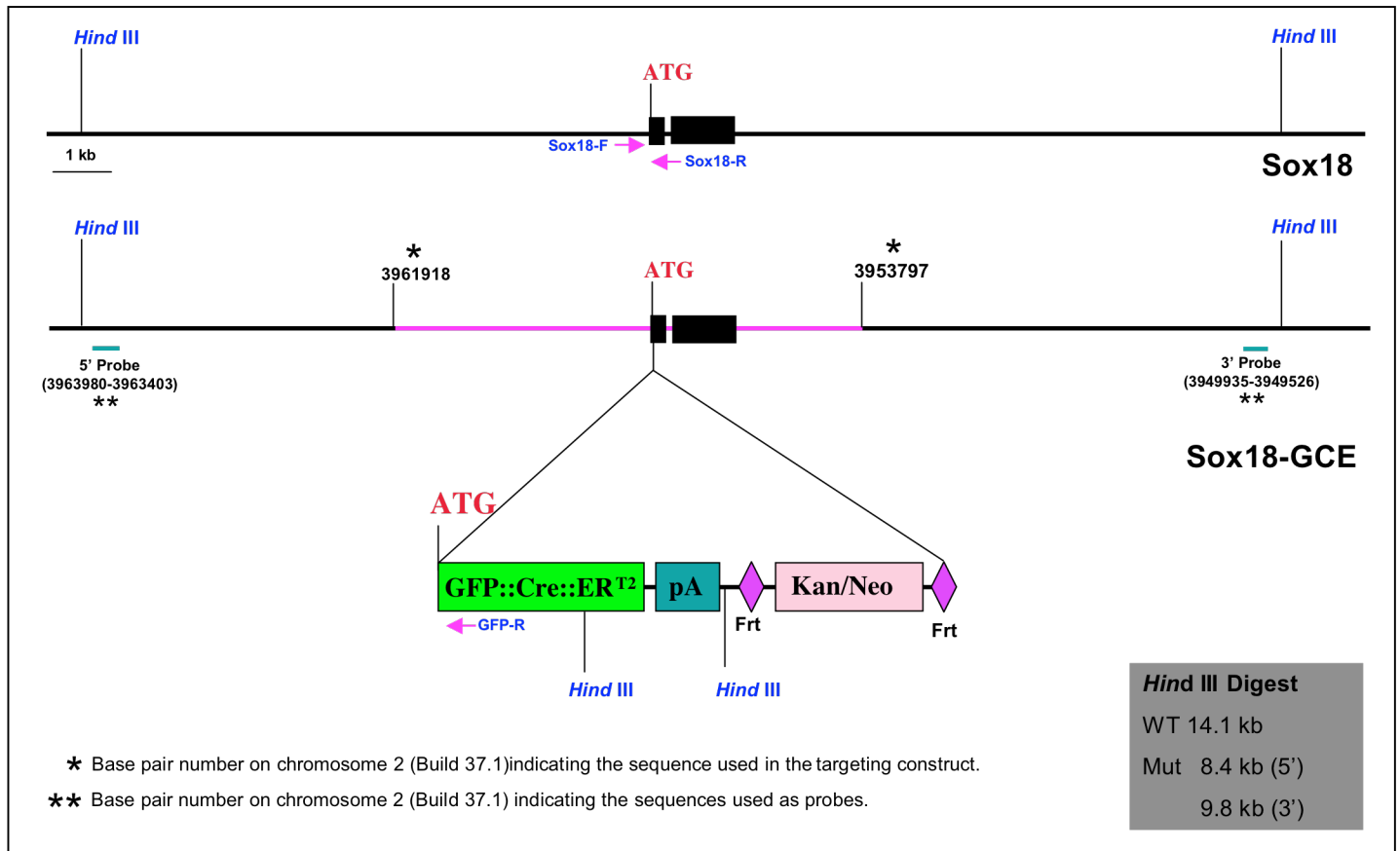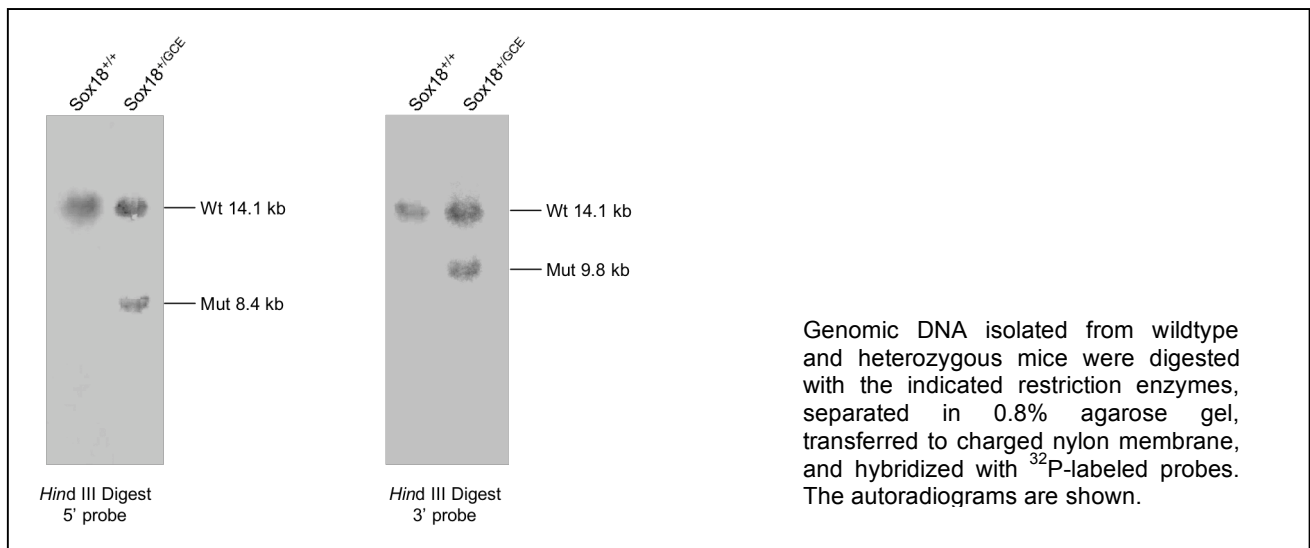# Sox18

A. Rationale

Nkx3.1 demarcates what is likely to be a rather specific aspect of vasculature or vascular associated cells and may be helpful in combination with Sox18 in getting at addressing different components of the vascular system deep medullary versus superficial cortical progenitors.

B. Targeting Strategy

Hind III   **Hind III**

ATG

Sox18-F →   ← Sox18-R

1 kb

Sox18

Hind III   *
3961918

*
3953797

Hind III

ATG

5' Probe
(3963980-3963403)
**

3' Probe
(3949935-3949526)
**

Sox18-GCE

ATG

GFP::Cre::ER^{T2} | pA | Kan/Neo

← GFP-R

Frt   Frt

Hind III   **Hind III**

★ Base pair number on chromosome 2 (Build 37.1) indicating the sequence used in the targeting construct.

★★ Base pair number on chromosome 2 (Build 37.1) indicating the sequences used as probes.

Hind III Digest

WT 14.1 kb

Mut 8.4 kb (5')

    9.8 kb (3')

C. Southern Blot Analysis of the Targeted Allele

Sox18$^{+/+}$ Sox18$^{+/GCE}$

— Wt 14.1 kb

— Mut 8.4 kb

Sox18$^{+/+}$ Sox18$^{+/GCE}$

— Wt 14.1 kb

— Mut 9.8 kb

Hind III Digest
5' probe

Hind III Digest
3' probe

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

E. Relevant Sequences

a. Genomic clone used for targeting construct

```
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<p align="center">Sox18-R</p>

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

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Sox18-F

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Start of Sox18 transcription
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Start of GFP

TGGTCGAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTGTCCGGCGAGGGCGAGGGCGATGCCACCTACGGCAAGC

GFP-R

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End of GFP

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Start of Cre
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End of Cre Start of ER

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GGCATCCGTGGAGGAGACGGACCAAAGCCACTTGGCCACTGCGGGCTCTACTTCATCGCATTCCTTGCAAAAGTATTACAT
CACGGGGGAGGCAGAGGGTTTCCCTGCCACAGCTTGATGA AGATCTGAGCTCCCTGGCGGAATTCGGATCTTATTAAAGC

End of ER Start of SV40 polyA

AGAACTTGTTTATTGCAGCTTATAATGGTTACAAATAAAGCAATAGCATCACAAATTTCACAAATAAAGCATTTTTTTCAC

TGCATTCTAGTTGTGGTTTGTCCAAACTCATCAATGTATCTTATCATGTCTGGTCGACGGTATCGATAAGCTTGATATCGA

ATTCC gaagttcctattctctagaaagtataggaacttc aggtctgaagaggagtttacgtccagccaagctagcttgg

polyA   Frt   Start of Kan/Neo

ctgcaggtcgtggtacgaaattctaccggggggaggcgcttttcccaaggcagtctggagcatgcgctttagcagccccgct
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End of Kan/Neo   Frt   Sox18 Cont'
CCCGGCTACGGCGCACAGGACGACCCGCCCTCCCGCCGCGACTGTGCATGGGCCCCTGGAATCGGGGCCGCTGCTGAGGCG
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        Sox18-R
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Exon/intron junction
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c. 5' Probe

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d. 3' Probe

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