Genotyping $Kdr^{im2.1Jrt}$

JDW 7/2012

CD309; FLK1; Flk-1; Flk1; VEGF receptor-2; VEGFR; VEGFR-2; VEGFR2; fetal liver kinase 1; vascular endothelial growth factor receptor-2;

MGI: 3629041


From the paper: An in-frame EGFP cDNA gene and a floxed PGK-neomycin resistance cassette replaced the translated portion of the first coding exon and the proximal part of the next intron, and placed EGFP under the transcriptional control of the endogenous promoter. The floxed PGK-neomycin promoter was subsequently removed by crossing to a cre-deleter strain.

JDW 42 (Kdr WT FOR) TGG AGA GCA AGG CGC TGC TAG C, In the ATG
JDW 43 (Kdr Common REV) CTT TCC ACT CCT GCC TAC CTA G
JDW 44 (Kdr EGFP MT FOR) CCC CCT GAA CCT GAA ACA TA
WT= 322 bp
MT= 600 bp

94C-5:00
94C-0:30
60C-0:30
72C-0:45
35 Cycles
72C-2:00
16C, forever

1.0 ul JDW 42 @ 20 uM
1.0 ul JDW 43 @ 20 uM
1.0 ul JDW 44 @ 20 uM
2.5 ul 10X Coral Red PCR Buffer
0.5 ul 10mM dNTPs
0.25 ul Qiagen Taq Polymerase
17.75 ul ddH$_2$O
1.0 ul genomic DNA
Other primers:

Kdr Common REV, in intron 1 oIMR 5223: TCC ACT CCT GCC TAC CTA GC
Kdr WT FOR in exon/intron 1, oIMR 5224: TGT GGG TAA GAA GCC CAC TC
Tm=60°C
Wild type = ~250 bp

The PCR conditions were 1 min at 94°C, 1 min at 66°C, and 1 min at 72°C for 35 cycles.

Run wt and mt reactions separately.

**For Flk1 EGFP**

<table>
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<tr>
<th>Primer Name</th>
<th>Tm</th>
<th>Genomic Location</th>
<th>GC Content</th>
<th>Tm Difference</th>
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<td>Fk1 WT Rev PRIMER</td>
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