

## R26R Reporter Mice

### Oligonucleotide sequences: 5' → 3'

(188) MβCx7 For	GATGTGCTCCAGGCTAAAGTT
(189) MβCx7 Rev	AGAAACGGAATGTTGTGGAGT
R1295	GCGAAGAGTTTGTCTCAACC
R523	GGAGCGGGAGAAATGGATATG
R26F2	AAAGTCGCTCTGAGTTGTTAT

### PCR Mix:

Component	Volume
Tail DNA	5 $\mu$ l Tail DNA
10x MGB	2.5 $\mu$ l
DMSO	2.5 $\mu$ l
10 mM dNTP mix	1.25 $\mu$ l
R1295 primer	2.0 $\mu$ l
R523 primer	2.0 $\mu$ l
R26F2 primer	2.0 $\mu$ l
Taq polymerase	0.25 $\mu$ l
$\beta$ -mercaptoethanol	0.125
dH <sub>2</sub> O	7.375 $\mu$ l
Total	25 $\mu$ l

### 10X MGB (Modified Gitschier's Buffer)      Final concentrations

447 $\mu$ l	1.5 M Tris, pH 8.8	670mM Tris pH8.8 final
166 $\mu$ l	1 M (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	166mM Ammonium sulfate
67 $\mu$ l	1M MgCl <sub>2</sub>	67 mM Magnesium chloride
200 $\mu$ l	0.5% gelatin	0.1% gelatin
120 $\mu$ l	ddH <sub>2</sub> O	
1.0 ml	TOTAL	

Sterile filter with a syringe filter and store at -20°C

### Thermocycler profile:

93°C for 2 min (initial cycle only)

93°C for 30 sec

58°C for 40 sec

65°C for 1 min 10sec

-----◇ x40

4°C for 5 min

## R26R PCR Screening

### Gel analysis

1% agarose gel

20  $\mu$ l each reaction per lane

Wildtype: 600 bp

Floxed: 325 bp

