

Genotyping Protocol for *lacZ* transgenic mice, including *mef2c-L8-lacZ*, *mef2c-F6-lacZ* and many others (Black Lab)

By Southern Blot (preferred)

Using a 352 bp fragment, which was amplified by PCR using the primers *lacZ*probe5 (5'-gtggtggttatgccgatc-3') and *lacZ*probe3 (5'-taccacagcggatggttcgg-3'), from the *lacZ* cDNA

Sequence of the probe:

```
gtggtggttatgccgatcgcggtcacactacgtctgaacgtcgaaaacccgaaactgtggagcgccgaaa
tcccgaatctctatcgtgcggtggttgaactgcacaccgccgacggcacgctgattgaagcagaagcct
gcgatgtcggtttccgcgagggtgcggttgaaaatggtctgctgctgctgaacggcaagccggttgctga
ttcgaggcgttaaccggtcacgagcatcatcctctgcatggtcaggatcatggatgagcagacgatgggtgc
aggatatcctgctgatgaagcagaacaactttaacgccgtgctgctgctgattatccgaaccatccgc
tgtagta
```

For the majority of Black lab-generated transgenic mice, this is typically analyzed on a *Pst*I digest. We analyze *mef2c-L8-lacZ* on a *Sac*I digest.

By PCR (faster; efficient for yolk sacs from young embryos)

Using the following primers:

LACZ5: 5'-cggatgaatggtgctgctgcttggga-3' (22-mer)

LACZ3: 5'-accaccgcacgatagagattc-3' (21-mer)

with 1 μ l of tail DNA (from a 50 μ l tail prep) under the following conditions:

94°C 5 min one cycle melt

94°C 30 sec

50°C 30 sec 30-36 cycles

72°C 1 min

72°C 7 min one cycle polish

This will detect a 385 bp for all of our *lacZ* transgenic lines