



## Welcome to The Genotyping Protocol System

## Master Protocol

**Strain Name:** C57BL/6NJ-Myh10<sup>em1J/J</sup>  
**Stock Number:** 025812  
**Allele:** Myh10<sup>em1J</sup>-EP  
**Protocol Name:** Myh10<sup>em1J</sup>-EP  
**Method:** End Point Analysis  
**Created:** 24-November -2014 (JKELMEN) **Updated:** 08-March -2016 (LUCASM)

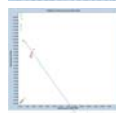
## Notes

Mutant = GGAGGATCCCGAGA[----]TCTCTTTGTGGAC  
WT = GGAGGATCCCGA GA[ggta]TCTCTTTGTGGAC  
Expected Results:  
Mutation is a deletion of ggta

## Attachments



View 25812.JPG



View 256281-EP w b6 labeled.JPG

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View Myh10 genomic.gcs

## Sequence

AGGCAATTACTTTTGGGTCCTTCTGTTTACAATGGCCCAGAGAACTGGACTGGAGGATCC  
CGA  
Sequence GA[ggta]TCTCTTTGTGGACAGGGCTGTCATCTACAACCCTGCCACTCAAGCTGACTGGAC  
AGC  
TAAAAAGCTGGTGTGGATTCCATCGGAACGCCATGGTTT

## Protocol Primers

| Primer | 5' Label | Sequence 5' --> 3'            | 3' Label              | Description | Reaction |
|--------|----------|-------------------------------|-----------------------|-------------|----------|
| 21934  | -        | TTA CAA TGG CCC AGA GAA CTG   | -                     | Forward     | -        |
| 21935  | -        | ACT AGC TGC CTC AAA ACC ATG   | -                     | Reverse     | -        |
| 21936  | Hex      | ATC CCG AGA GGT ATC TCT TTG T | Black Hole Quencher 1 | WT Probe    | -        |
| 21937  | 6-FAM    | GAT CCC GAG ATC TCT TTG TG    | Black Hole Quencher 1 | MUT Probe   | -        |

## Reaction/Components A

| Reaction Components | Volume Amt | Final Concentration | Total Volume Amt |
|---------------------|------------|---------------------|------------------|
|---------------------|------------|---------------------|------------------|

## Cycling

| Step # | Temp°C | Time  | Note |
|--------|--------|-------|------|
| 1      | 95     | 10min | -    |

| Reaction/Components A    |      |     |   | Cycling |    |       |                                |
|--------------------------|------|-----|---|---------|----|-------|--------------------------------|
| 2 X Kapa Probe Fast QPCR | 2.50 | .83 | - | 2       | 92 | 15sec | -                              |
| ddH2O                    | 1.07 | -   | - | 3       | 60 | 1 min | -                              |
| 100 uM 21934             | 0.02 | .33 | - | 4       | -  | -     | repeat steps 2-3 for 40 cycles |
| 100 uM 21935             | 0.02 | .33 | - | 5       | 4  | -     | Forever                        |
| 100 uM 21936             | 0.01 | .17 | - |         |    |       |                                |
| 100 uM 21937             | 0.01 | .17 | - |         |    |       |                                |
| DNA                      | 2.37 | -   | - |         |    |       |                                |

Number Of Reactions 1

Version 3.2