EMMA ID: *EM:16575*
Gene: *Ube2l6*
Common name: *Ube2l6-em1_2*
Allele: *Ube2l6* $^{em1(IMPC)Hmgu}$

Allele Information

For more information on production, guides and mutation, search for gene/project, go to project summary, go to production plan, go to production outcome and "more details"

https://www.gentar.org

IMPC mouse phenotype data, search by the gene name
http://www.mousephenotype.org/

Genotyping Information

Genotyping by end-point PCR based on gel is composed of a genespecific short range PCR using primers on wild type allele and a mutant allele-specific short range PCR. The combined results show the genotype of the mice. For example: mutant positive, wild type positive = Heterozygous. In addition to the expected product, the mutant assay may also amplify the endogenous wild type sequence, which will appear as a larger band on an agarose gel. The presence of this extra band will depend on the size of the original deletion.

PCR primer pairs and expected size bands

Assay	Forward Primer	Reverse Primer	Expected Size Band (bp)
Wild type	Ube2l6_F	Ube2l6_wtR	400
Mutant	Ube2l6_F	Ube2l6_R	295/764 (wt)

Because of a deletion size of 469 bp you will see an additional band at 764 bp with the primer pair Ube2l6_F/Ube2l6_R

Primer sequences

Primer Name	Sequence 5' --> 3'
Ube2l6_F	GGGCTCACTCAGGGAAAGAG
Ube2l6_wtR	TCATTGCTGATGAGGGGCAG
Ube2l6_R	GGATCACACCAGGTTTAGGCT

PCR setup (AllTaq)

Component	Volume (µl) 1x	Final conc.
DNA (~ 50-100 ng)	2	
Q-Solution (5x)	2,5	0,5
PCR-Buffer (5x)	5	1
DNTP mix (10 mM)	0,5	0,2
$MgCl_2$ (25 mM)	1,5	1,5
Primer 1 (10 pmol/µl)	1	0,4
Primer 2 (10 pmol/µl)	1	0,4
Primer 3 (10 pmol/µl)	1	0,4
Taq Polymerase (5 U/µl)	0,5	2,5 U/rxn
H_2O*	10	
Final volume	25	

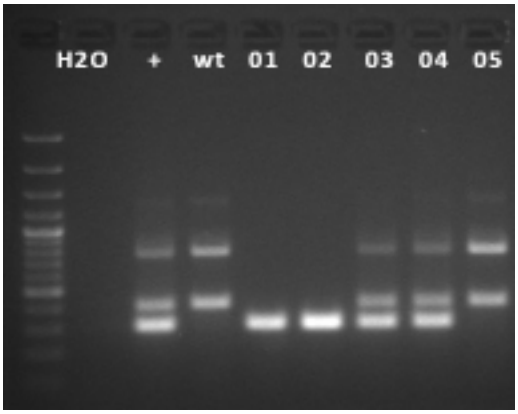
*** The amount of H_2O is adjusted with the number of primer.**

Amplification conditions

PCR Settings	Temperature (°C)	Time	# of cyles
1 Denaturation (Melting)	95°C	5 min	1
2 Amplification (Melting, Annealing, Polym.)	94°C 65-55 TD 72°C	30 sec 45 sec 45 sec	39
3 Polymerisation	72°C	10 min	1
4 Cooling	4°C	hold	1

Touch-Down cycling protocol: first 10 cycles anneal at 65°C, decreasing 1°C per cycle, next 30 cycles aneal at 55°C.These PCR conditions have been optimized for our methods and preparation

Gel Image



+ het control
01,02 hom
03,04 het
05 wt

Separated by gel electrophoresis on a 2% agarose gel.