

## B9d2 (MCZV; EPD0481\_2\_D09)

Allele: *B9d2*<sup>tm1a(EUCOMM)Wtsi</sup>

### Genotyping Information

These mice may be genotyped through a combination of separate PCR reactions that detect *LacZ*, the gene-specific wild type allele, and a mutant allele-specific short range PCR. Interpretation of the consolidated results produces the genotype of the mice.

For example: *LacZ* positive, mutant positive, wild type positive = heterozygous.

#### ➤ PCRs primer pairs and expected size bands

PCR type	Forward primer	Reverse primer	Expected size band (bp)
Mutant PCR	B9d2_89540_F	CAS_R1_Term x	134
Wild type PCR	B9d2_89540_F	B9d2_89540_R	186
LacZ PCR	LacZ_2_small_F	LacZ_2_small_R	108

#### ➤ Primer sequences

Primer name	Primer sequence (5' > 3')
CAS_R1_Term	TCGTGGTATCGTTATGCGCC
B9d2_89540_F	AGACAGGGAAACAGAGAGAAGC
B9d2_89540_R	GCCAGGGCTACACAGAGAAT
LacZ_2_small_F	ATCACGACGCGCTGTATC
LacZ_2_small_R	ACATCGGGCAAATAATATCG

This technical data sheet and information ("Datasheet") is supplied by Genome Research Limited ("GRL").

Although reasonable care is taken in the preparation of this Datasheet, GRL gives no warranties express or implied for any use of the Datasheet or for the accuracy of the Datasheet. GRL assumes no responsibility or liability for any decisions based upon the Datasheet. Without limiting the foregoing the Datasheet was prepared for mice supplied directly from GRL and where copies of this Datasheet are available from third party repositories or distribution centres ("Third Parties") GRL shall not be liable for any inconsistency between the mouse strain supplied by the Third Party and the Datasheet howsoever arising.

Last updated: 30/08/2011

➤ **Reaction**

Reagent	μl
DNA (~50-100 ng)	1.0
10x Buffer	2.0
MgCl <sub>2</sub> (50 mM)	0.6
PtTaq (Platinum® Taq (Invitrogen))	0.2
dNTPs (100 mM)	0.2
Primer 1 (10 μM)	0.4
Primer 2 (10 μM)	0.4
H <sub>2</sub> O	<u>15.2</u>
<b>Total</b>	<b>20.0</b>

➤ **Cycling conditions**

**Wild type and mutant PCRs**

Cycle	Temp	Time
1	94 °C	5 min
2	94 °C	30 sec
3	58 °C	30 sec
4	72 °C	45 sec
5	Go to '2' + 34 cycles	
6	72 °C	5 min
7	12 °C	forever

**LacZ PCR**

Cycle	Temp	Time
1	94 °C	5 min
2	94 °C	30 sec
3	60 °C	30 sec
4	72 °C	30 sec
5	Go to '2' + 34 cycles	
6	72 °C	5 min
7	12 °C	forever

This technical data sheet and information ("Datasheet") is supplied by Genome Research Limited ("GRL").

Although reasonable care is taken in the preparation of this Datasheet, GRL gives no warranties express or implied for any use of the Datasheet or for the accuracy of the Datasheet. GRL assumes no responsibility or liability for any decisions based upon the Datasheet. Without limiting the foregoing the Datasheet was prepared for mice supplied directly from GRL and where copies of this Datasheet are available from third party repositories or distribution centres ("Third Parties") GRL shall not be liable for any inconsistency between the mouse strain supplied by the Third Party and the Datasheet howsoever arising.

Last updated: 30/08/2011