

C57BL/6NJ-Avpr1a^{em1(IMPC)J}/Mmjax

Stock No: 032811

Protocol 34348: Probe Assay - Avpr1a^{<em1(IMPC)J>}

Version 1.0

Notes

Taqman qPCR protocols are run on a real time PCR instrument. Use an appropriate instrument specific Fluorophore/Quencher combination.

The genotyping protocol(s) presented here have been optimized for reagents and conditions used by The Jackson Laboratory (JAX). To genotype animals, JAX recommends researchers validate the assay independently upon receipt of animals into their facility. Reaction cycling temperature and times may require additional optimization based on the specific genotyping reagents used.

Expected Results

>[chr10:122448339+122448466](#) 128bp CGCTGCATGACTTTAAATTGG CAGGGGTGGAGCTGAGAAT

Mut= 129 bp

Wt= 128 bp

Fam=Mut

Hex=Wt

Sequence

Wt Sequence (deletion in lower case):

```
GGGCCTGGGGGCTTGAAGGGTGGACCCCAAATCCAGG
CGCACTGAGTTCGCAGCAGCAGCAGCAGCGCGATCTCTA
CGTACGCTTTTACTACACCTGTTGACGCGCTGCATGACTT
TAAATTGGCTTATAAAGGAGGCTGGAGAAAAttccgaggagcca
gagaagttggggcagatccggaagcgcgggctctgcagaggaatctaggatt
ctcagctccaccctgcagcaccaaagagaaaaggacaaatcaccacgactg
gaggggtgggtgtaggggacagtcctccagccactaagatgagaaggagcgcatc
ctgaggcagcctaagactgcttggggagcagggagagtccgctccctgcctcggg
caagccggagcagggacacgcaagaacatccatccaaaaccactctgagcgc
gctggtgattgcgagcgaagagagcagggtaaggaaggacggtgctgcagaa
cttaggacgcggtcttcgggtcctcggaggagaaggaagcagcccacgagg
aacaggggtccccggtctgtgcgacagcatgagttcccgagggctcccacgat
ctgcccggggcaactccagcccgtggtggcctctgaccaccgagggcgccaac
agcagccgggaagcagctgggctcggggaaggcggcagcccggggggac
gtacgcaacgaggagctggcgaagctggaggtaaccgtctgctggctgatttctg
gtggccgtgctgggtaatagcagtgctgctgctgctgcagcagccacgcaa
gacatcccgatgcatctctcatccgacacctcagcctggcagacctggcgctgc
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agtgaagatgacctgtgattgaagcgcctacatcctctgctggacaccttctcatc
gtccagatggtgctgctgggataccaattctgttgaccggtatgactgagactac
tatggaagagggtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgt
gggggggagttgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgtgt
GGTAAAGCTTCATGTAAGAAGCTTAAATGAACAAAATTGC
AAATTCCTGGGACCTCCACCCCAGGTACACTGGAGCCCA
GGATTACAGCATCTGCAGCATGTGTGACCACCCTAAGGCC
ACCAGCTGTGCCCTAGAGTCACTAGTTACAAAACACTTGTG
ACATTTTGCCAAATTAC
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This mutation is a 1550 bp deletion beginning at Chromosome 10 position 122448382 bp and ending after 122449931 bp (GRCh38/mm10).

JAX Protocol

Protocol Primers

PRIMER	5' LABEL	SEQUENCE 5' → 3'	3' LABEL	PRIMER TYPE	REACTION	NOTE
43365		CGC TGC ATG ACT TTA AAT TGG		Common	A	
43366		CAG GGG TGG AGC TGA GAA T		Wild type Reverse	A	
43367		ATC CTG GGC TCC AGT GTA CC		Mutant Reverse	A	
43368	Fluorophore-1	CGA GGA GCC CAG AGA AGT TT	Quencher-1	WT Probe		
43369	Fluorophore-2	CAA ATT CCT GGG ACC TCC A	Quencher-2	MUT Probe		

Reaction A

COMPONENT	FINAL CONCENTRATION
Kapa Probe Fast QPCR	1.00 X
ddH2O	
43365	0.40 uM
43366	0.40 uM
43367	0.40 uM
Wt Probe	0.15 uM
Mutant Probe	0.15 uM
DNA	

Cycling

STEP	TEMP °C	TIME	NOTE
1	95.0	--	
2	95.0	--	
3	60.0	--	
4		--	repeat steps 2-3 for 40 cycles
5	4.0	--	Forever

JAX uses a very high speed Taq (~1000 bp/sec), use cycling times recommended for your reagents.



