



## Welcome to The Genotyping Protocol System

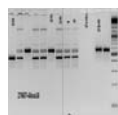
## Master Protocol

**Strain Name:** C57BL/6NJ-*Abca16*<sup>em1J/J</sup>  
**Stock Number:** 027607  
**Allele:** *Abca16*-6894-KOMP CRISPR  
**Protocol Name:** *Abca16*<sup>em1J</sup>  
**Method:** Standard PCR  
**Created:** 23-October -2015 (DTG)    **Updated:** 12-January -2016 (LUCASM)

## Notes

Expected Results:  
 Mutant = 480 bp  
 Heterozygote = 480 bp and 572 bp  
 Wild type = 572 bp  
 Readthrough band at ~1100

## Attachments



View [27607-abca16-gel.jpg](#)

- View [Abca16 genomic.gcs](#)

## Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
23607	-	TCC TTG AGA ATC TTT TCA TGC T	-	Forward	-
23608	-	CCC ACT TGC AGG TTT TTC CT	-	Reverse	-

## Reaction/Components A

Reaction Components	Volume Amt	Final Concentration	Total Volume Amt
ddH2O	4.55	-	-
5 X Kapa 2G HS buffer	2.40	1	-
25 mM MgCl2	0.96	2	-
10 mM dNTPS-kapa	0.24	.2	-
20 uM 23607	0.30	.5	-
20 uM 23608	0.30	.5	-
5 mM 10x Loading Dye	1.20	.5	-
2.5 U/ul Kapa 2G HS taq polymerase	0.05	.01	-
DNA	2.00	-	-

## Cycling

Step #	Temp °C	Time	Note
1	94	2 min	-
2	94	20sec	-
3	65	15sec	-0.5 C per cycle decrease
4	68	10sec	-
5	-	-	repeat steps 2-4 for 10 cycles
6	94	15sec	-
7	60	15sec	-
8	72	10sec	-
9	-	-	repeat steps 6-8 for 28 cycles
10	72	2 min	-

<b>Cycling</b>
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11	10	-	hold
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Number Of Reactions 1

Version 3.2
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