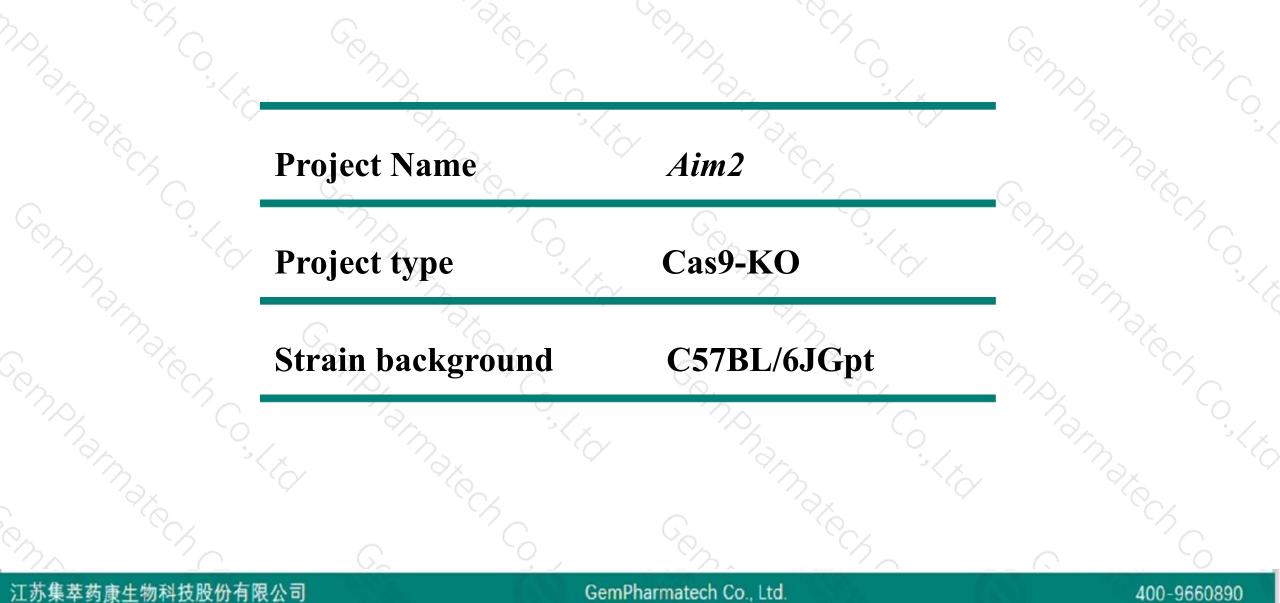


Aim2 Cas9-KO Strategy

Designer:Fengjuan Wang Reviewer:Lingyan Wu Design Date:2019-12-9

Project Overview

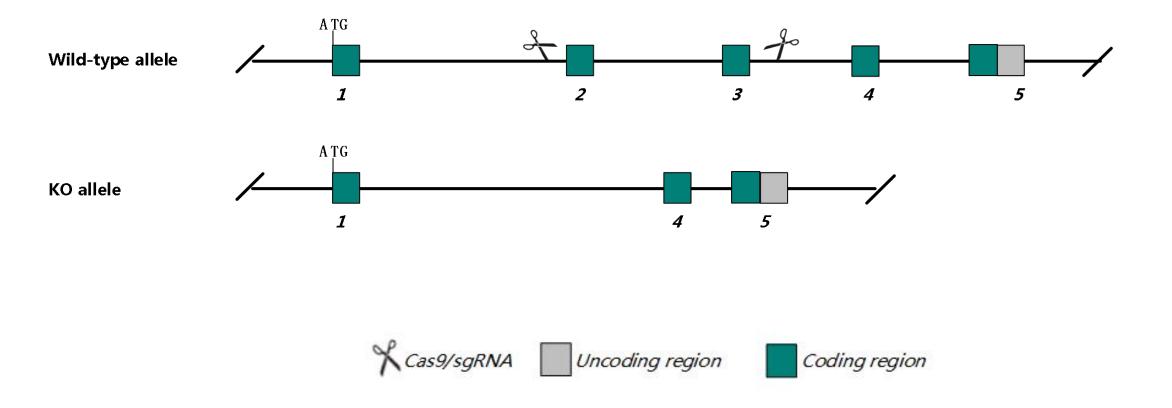




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Aim2 gene. The schematic diagram is as follows:





The Aim2 gene has 7 transcripts. According to the structure of Aim2 gene, exon2-exon3 of Aim2-204 (ENSMUST00000166137.2) transcript is recommended as the knockout region. The region contains 566bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Aim2* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- According to the existing MGI data, Mice homozygous for a gene trapped allele exhibit increased susceptibility to bacterial and viral infections with altered cytokine production and inflammatory cell death (pyrotosis).
 Transcripts *Aim2* -201, 203, 206 are not be affected.
- The Aim2 gene is located on the Chr1. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Notice

Gene information (NCBI)



\$?

Aim2 absent in melanoma 2 [Mus musculus (house mouse)]

Gene ID: 383619, updated on 9-Apr-2019

Summary

Official Symbol	Aim2 provided by MGI
•	absent in melanoma 2 provided byMGI
Primary source	
27.0	Ensembl:ENSMUSG00000037860
RefSeq status	
Organism	
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Lineuge	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	
	Broad expression in spleen adult (RPKM 1.0), CNS E11.5 (RPKM 0.8) and 22 other tissuesSee more
Orthologs	

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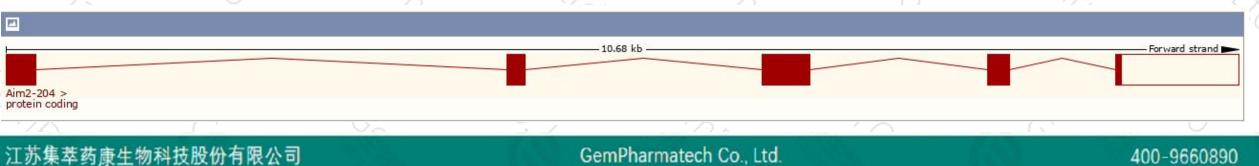
Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

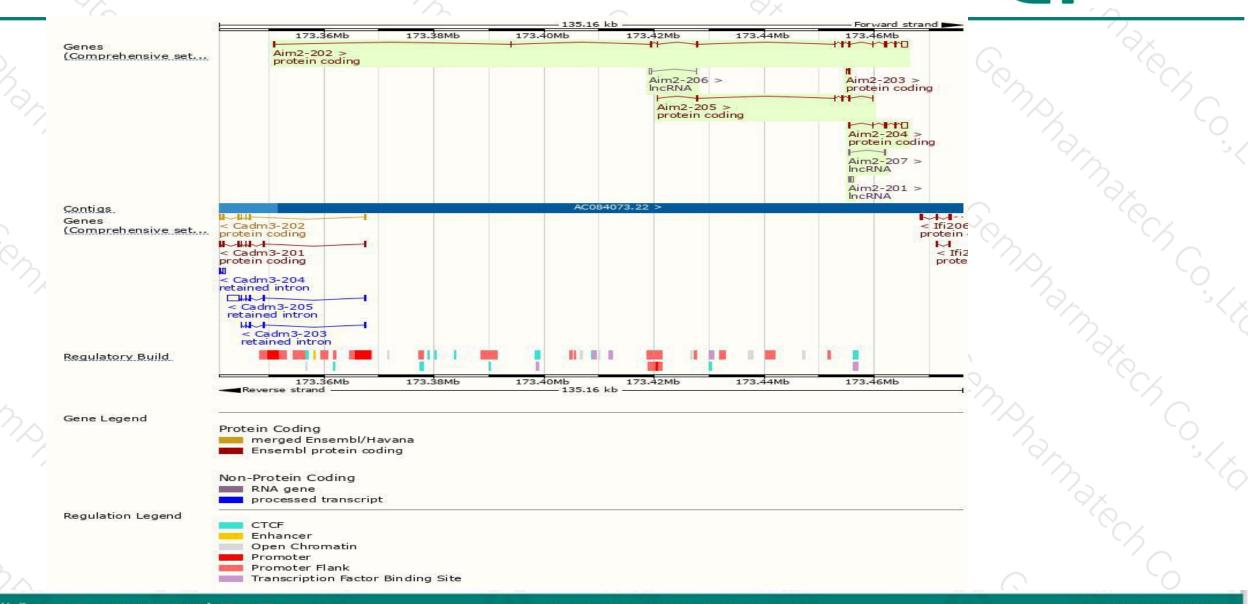
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Aim2-202	ENSMUST00000147604.7	2839	<u>354aa</u>	Protein coding	CCDS15529	<u>Q91VJ1</u>	TSL:5 GENCODE basic APPRIS P1
Aim2-204	ENSMUST00000166137.2	2079	<u>354aa</u>	Protein coding	CCDS15529	<u>Q91VJ1</u>	TSL:5 GENCODE basic APPRIS P1
Aim2-205	ENSMUST00000173023.1	744	<u>94aa</u>	Protein coding	1440	G3UZ36	CDS 3' incomplete TSL:1
Aim2-203	ENSMUST00000151176.1	326	<u>80aa</u>	Protein coding	127	D3Z341	CDS 3' incomplete TSL:5
Aim2-206	ENSMUST00000174263.1	383	No protein	IncRNA	170	-	TSL:5
Aim2-201	ENSMUST00000135370.1	372	No protein	IncRNA	(.)		TSL:3
Aim2-207	ENSMUST00000192575.1	363	No protein	IncRNA	1.20	-	TSL:1

The strategy is based on the design of Aim2-204 transcript, The transcription is shown below



Genomic location distribution

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Protein domain

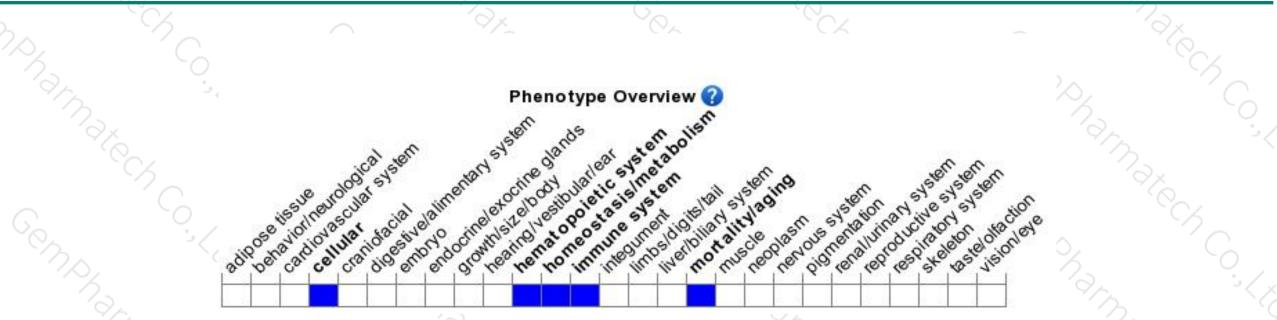


Variant Legend	missense varia	int									
Invient Logand		ant in the second se		synon	ymous variant						
ll sequence SNPs/i	Sequence variants (dbSNP and all other s	ources)	3	Ŭ	1	Ĩ.	11	ĩ	Ť.	
DD	cd08305										
ene3D	HIN-200 family 1.10.533,10		_		2.40.50,140						
ANTHER	PTHR12200:SF17										
ROSITE profiles	DAPIN domain				HIN-200/IF120x						
am.	DAPIN domain		-		HIN-200/IF1	20x					
1ART	DAPIN domain	superiamiy			55155141						
piled-coils (Ncoils) uperfamily	Death-like domain				SSF159141						
DB-ENSP mappings obiDB lite w complexity (Seg)											
NSMUSP00000132											

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Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Mice homozygous for a gene trapped allele exhibit increased susceptibility to bacterial and viral infections with altered cytokine production and inflammatory cell death (pyrotosis).



If you have any questions, you are welcome to inquire. Tel: 400-9660890



